

Use Of Sawdust From Different Tree Species And Some Non-Wood Agro-Wastes In The Cultivation Of *Pleurotus Pulmonarius*

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Abstract:

Background: Large volumes of agro-waste are produced globally as a result of increased population growth. Most of this waste is lignocellulosic in nature and slow to decompose, leading to accumulation of these wastes, pollution, and associated health risks. Cultivating *Pleurotus pulmonarius*, a nutritious and medicinal mushroom, on these wastes offers a sustainable recycling method. This study evaluates performance of *P. pulmonarius* on different sawdust types and non-wood agro-wastes with a view to identify the best substrate for its production.

Materials and Methods: Spawn of *Pleurotus pulmonarius* sourced from Federal Institute of Industrial Research, Oshodi (FIIRO), Lagos State, was first of all multiplied using sorghum grains and cultivated on sawdust substrates from five different tree species (*Azizelia africana* (AA), *Pinus* sp. (PS), *Gmelina arborea* (GA), *Swientia mahagani* (MA) and *Milicia excelsa* (ME)) and two non-wood agro-wastes (plantain mid-rib (PM) and corn cobs (CC)). The experiment incorporated a completely randomized design replicated thrice. Data collected included spawn run, pinhead formation duration, yield and yield components, proximate composition, mineral element and heavy metal (zinc, iron, lead, nickel, mercury and cadmium) concentration of the substrates including those of the harvested fruiting bodies. SPSS 20 was employed in data analysis at $P < 0.05$.

Results: The substrates differed significantly ($P < 0.05$) in their nutrient composition. They contain higher quantities of carbohydrate and fibre but low protein. Carbohydrate concentration ranged from $42.19 \pm 0.28\%$ in PM to $60.78 \pm 0.31\%$ in GA; fibre ranged from $27.80 \pm 0.35\%$ in GA to $44.48 \pm 0.46\%$ in PM; protein was from $4.17 \pm 0.39\%$ in GA to $5.68 \pm 0.32\%$ in CC. The substrates were also high in phosphorus and potassium. The heavy metal (excepting zinc and iron) concentration of the substrates were low, below 1.00mg/kg. *pulmonarius* cultivated on PM yielded significantly ($P < 0.05$) the highest total fresh weight (84.97 ± 0.20 g/kg) while that on ME yielded the lowest total fresh (30.75 ± 0.40 g/kg). The nutrient content of the harvested *P. pulmonarius* from the different substrates varied significantly ($P < 0.05$). The mushroom was high in fibre, protein and carbohydrate but low in fat content. The ranges of these nutrients (%) in the harvested mushroom were fibre: $11.93 \pm 1.30 - 20.70 \pm 0.57$ on ME and PM, respectively; protein: $17.25 \pm 2.00 - 29.97 \pm 1.25$ on PM and PS, respectively; carbohydrate: $37.28 \pm 0.11 - 46.70 \pm 1.64$ on PS and PM, respectively; fat: $1.40 \pm 0.14 - 2.95 \pm 0.21$ on GA and PM, respectively. It was also high in mineral elements most especially phosphorus, potassium and magnesium. The heavy metal concentration of the mushroom grown on all the substrates were below their threshold limit making them safe for consumption.

Conclusion: All the substrates contain nutrients, which supported the growth and fructification of *P. pulmonarius* with PM adjudged the best.

Key Word: *Pleurotus pulmonarius*, agro-waste, yield, nutrient content, heavy metal

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I. Introduction

Oyster mushrooms are mushrooms belonging to the genus, *Pleurotus* and phyla Basidiomycetes. There are about forty species of this genus including *P. pulmonarius*. All oyster mushroom species are edible¹. Oyster mushrooms have a fragile fruit body and can be recognized by their short, eccentric stalks that is not attached centrally. The name Oyster was derived from the marine bivalve mollusks, oyster, as these mushrooms have a white, shell-like appearance and flavour similar to that of the popular bivalve mollusks.

Pleurotus have excellent nutritional profiles. They are rich in fiber, vitamins (thiamine, riboflavin, ascorbic acid, ergosterine, and niacin), micro and macro-elements (phosphorus and iron), protein with important content of essential amino acids especially, lysine and leucine, which are lacking in most staple cereal foods². Their protein content is in the range of 25 – 27 g/100g and can therefore be used as an excellent source of protein supplement³. They have higher mineral contents than meat, fish, eggs, cheese and most vegetables⁴. They also have low fat (but with excellent important fatty acids) and carbohydrate contents, an indication that they are good for diabetic and obese people⁵. In addition to their excellent nutritional value, *Pleurotus* spp. have therapeutic value. They are rich source of antioxidants⁶. There are also reported to have anti-inflammatory, hepatic-protective, anti-diabetic, anti-tumor, anti-microbial properties^{7,8}.

Pleurotus pulmonarius (commonly known as the phoenix oyster mushroom) like every other oyster mushroom is a lignocellulolytic fungus. It has the ability to breakdown cellulose, hemicelluloses and lignin from wood and thus can be grown on various agricultural and forestry by-products⁹. This makes it an ideal candidate for bioconversion of organic wastes into high-value food. These wastes, which include sawdust, corn cobs, cereal straws and husks, palm bunch, coconut husk, cocoa pod, etc. are abundant in both rural and urban areas. They have insignificant or no commercial value, and certainly no food value, at least in their original form. Disposal of these wastes is a very serious problem. In Nigeria, these wastes are carelessly disposed off to the surrounding environment either by dumping or burning. These cause environmental pollution, and consequently health hazards. The use of such lignocellulosic wastes not only provides a cost-effective substrate for mushroom cultivation but also contributes to waste management and environmental conservation.

Mushroom substrate composition can significantly influence the yield, nutritional value, and growth rate of mushrooms. For instance, research conducted by Okwulehie and Nosike showed that the organic constituent in the bark of some Nigerian trees supported the growth and yield of *P. pulmonarius* with the best result achieved with *Treculia africana*¹⁰. In another study, Hoa HT *et al.* reported significant differences in the productivity, nutritional and mineral compositions of two oyster mushrooms, *P. ostreatus* and *P. cystidiosus* cultivated on seven different substrate formulas¹¹. Agba *et.al.* in their study on the impact of agro wastes in sole and in combination on the growth and yield of *P. ostreatus* found that sole sawdust was the best for the cultivation of the mushroom¹². Therefore, identifying the best agro-waste for a particular mushroom could enhance both the ecological and economic aspects of mushroom production. Unlike *P. ostreatus* where much has been done, there is scarcity of information on the cultivation of *P. pulmonarius*. In addition, there is dearth of information on the use of plantain mid-rib in mushroom cultivation. This study aims to determine the most effective substrates for maximizing the yield of *P. pulmonarius* and promoting sustainable environmental protection without compromising the nutritive value of the mushroom.

II. Material And Methods

Study location

The research study was conducted in the mushroom house of Department of Plant Science and Biotechnology, Abia State University, Uturu, Abia State, Nigeria.

Sample Collection

The mother spawn of *P. pulmonarius* used for this study was gotten from the Federal Institute of Industrial Research, Oshodi, Lagos State. The substrates were sawdust from five different tree species (*Azefelia africana*, *Pinus* sp, *Gmelina arborea*, *Swietenia mahagoni* and *Milica excelsa*) and two non-wood wastes (plantain mid rib and corn cobs). The sawdust from *Azefelia africana* and *Pinus* sp were collected from timber markets, Umuahia, that from *Gmelina arborea* was sourced from Okigwe while those from *Swietenia mahagoni* and *Milica excelsa* were sourced from Port-Harcourt. The non-wood wastes were collected from Okigwe.

Multiplication of Mother Spawn

The mother spawn of the two *Pleurotus* species were multiplied using sorghum grains according to the methods described by Narh Mensah D *et al.* and Okwulehie IC *et al.*^{13,14}. 1 kg of sorghum grains were winnowed to get rid of chaff and bad seed. They were thoroughly washed and boiled until they became tender but not allowed to break. They grains were allowed to cool. A little quantity of calcium carbonate was added to the grains to prevent them from sticking together. After cooling, the grains were loosely packed in 20 clean and sterilized empty salad bottles that could withstand heat. The bottles containing the grains were plugged with non-absorbent cotton wool, covered with aluminum foil and sterilized in an autoclave for an hour at a temperature of 121°C. The bottles were allowed to cool and made free of condensed water before inoculating the *P. pulmonarius* mother spawn. The inoculated bottles were incubated at room temperature (25°C), and the bottles were shaken every 3 – 4 days for one minute to loosen the grains being colonized by the growing mycelia.

Substrate preparation and pasteurization

The methods of Kimenju JW *et al.* and Sharma K were adopted^{15,16}. The plantain mid-rib and corn cobs were chopped to about 2-3 cm long. About 9 kg each of the substrates were separately soaked overnight in a basin of clean tap water. The water was changed 1 to 2 times to prevent the substrates from fermenting before pasteurization. The soaked substrates were drained of excess water until about 70% water content was attained. After which, 1 kg of each of the substrates were dispensed separately in three replicates into heat-resistant polythene bags and then pasteurized for six (6) hours in a gas-heated drum.

Inoculation of the spawn and incubation

The substrates after cooling were transferred separately into sterilized perforated transparent buckets and properly labelled. *P. pulmonarius* spawns were inoculated into each of the substrates contained in the buckets. After inoculation, the buckets were transferred to the cropping room, kept in wooden racks and covered with black polythene bag to serve as dark incubation for ramification to take place. The cropping room were constantly flooded to maintain good humid condition necessary for vegetative growth of fungi. The polythene bags were removed after full colonization of the substrate by the mycelium and prior to primordial formation.

Experimental design

The experiment incorporated a completely randomized design with three replications. The treatments were sawdust from the five different tree species (*Azafia africana*, *Pinus* sp, *Gmelina arborea*, *Swietenia mahagoni* and *Milica excelsa*) and the two non-wood agro-wastes (plantain mid rib and corn cobs). This gave a total of 7 treatments and 21 experimental units.

Data collection

Data were collected on the following:

Nutrient composition: Proximate and mineral composition (calcium, magnesium, sodium, iron, zinc, phosphorus and potassium) of each of the substrates were determined prior to use using the method described by AOAC¹⁷. Also, proximate and mineral contents of the fruiting bodies harvested from each of the substrates were determined as stated earlier.

Heavy metal composition: The heavy metal (mercury, nickel, lead, cadmium) content of each of the substrates before use was determined using the method of AOAC¹⁷. Also, the heavy metal content of the fruiting bodies of the mushrooms harvested from each of the substrates were determined using the same method as above.

Duration of spawn run (mycelium growth period): This is the number of days it took the mycelium to fully colonize the substrate.

Period of pin head formation: This is the time (days) taken for the pin heads to appear after full colonization of the mycelium.

Cap/pileus diameter: This was measured with a ruler from one end of the cap through the middle to the other end. This were done prior to the harvest of the mushroom at maturity.

Stipe length: This was measured with a ruler from the base of the fruiting body on the substrate to its cap. This were done prior to the harvest of the mushroom at maturity.

Stipe girth: This was measured with vernier calipers

Number of fruiting bodies: The number of fruiting bodies produced in each substrate was counted and the value recorded.

Yield measured as fresh and dry weights of the mushroom: This is the weight (g) of mushroom produced per bucket of substrate per harvest time. Mature mushrooms were harvested manually by hand plucking; this was done in flushes of one and two. Thereafter, the fresh weight of the mushrooms for each flush in each replicate was weighed immediately after harvesting using an electronic balance, and recorded as the fresh weight. After which, they were separately packed in a properly labeled envelopes and oven-dried at a temperature of 60°C until a constant weight was achieved and the weight were recorded as the dry weight.

Data analysis

Data collected were analyzed statistically using SPSS 20 at probability level of 0.05. Significant means were partitioned using Fisher's Least Significant Difference (FLSD) at $P < 0.05$.

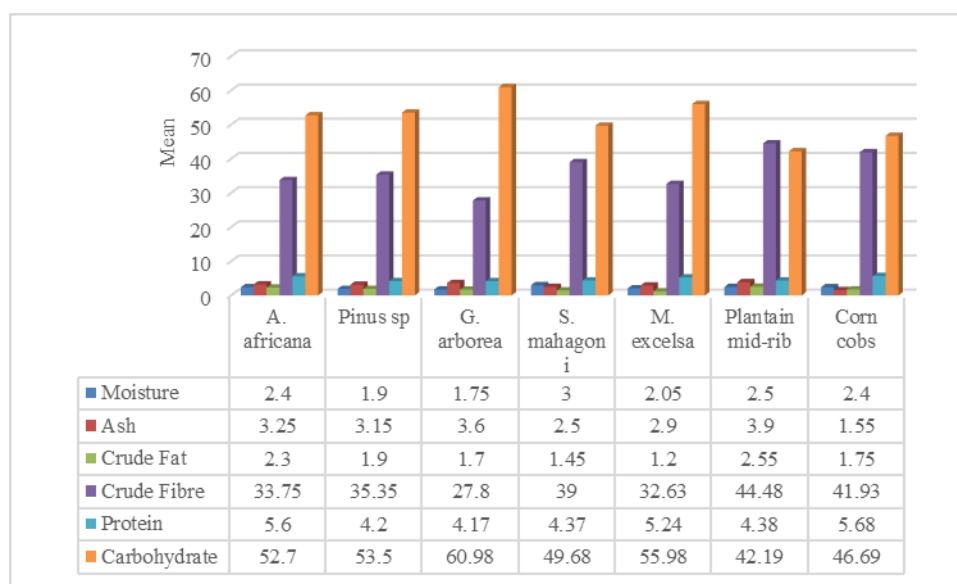
III. Result

Proximate composition of the substrates

Significant differences were found in the proximate composition of the substrates (Table no 1). *Swietenia mahagoni* (MA) had significantly ($P < 0.05$) the highest moisture content, this was followed by plantain mid-rib (PM), which had moisture content that was significantly different from the rest except from corn cobs (CC) and *Azfelia africana* (AA). *Gmelina arborea* (GA) had the least moisture content though it was not significantly different from that of *Pinus* sp (PS). PM had significantly the highest ash content followed by GA while CC had significantly the least. For crude fat content, PM again had significantly the highest concentration followed by AA while *Milicia excelsa* (ME) had significantly the least. In terms of crude fibre content, PM had significantly the highest concentration, this was followed by CC whereas GA had significantly the least. Although CC had the highest protein content, it was statistically similar to those of AA and ME, which were significantly different from those of the rest. GA had the least protein content that was not significantly different from those of the remaining three substrates. In the case of carbohydrate concentration, GA had the significantly the highest concentration followed by ME and the least was PM.

Table no 1: Proximate composition (%) of the different substrates before spawn inoculation

Substrate	Moisture	Ash	Crude Fat	Crude Fibre	Protein	Carbohydrate
<i>A. africana</i>	2.40±0.00 ^{bc}	3.25±0.07 ^c	2.30±0.14 ^b	33.75±0.21 ^c	5.60±0.49 ^a	52.70±0.78 ^c
<i>Pinus</i> sp	1.90±0.14 ^d	3.15±0.07 ^c	1.90±0.14 ^c	35.35±0.21 ^d	4.20±0.00 ^b	53.50±0.14 ^c
<i>G. arborea</i>	1.75±0.07 ^d	3.60±0.14 ^b	1.70±0.00 ^c	27.80±0.85 ^f	4.17±0.39 ^b	60.98±1.31 ^a
<i>S. mahagoni</i>	3.00±0.14 ^a	2.50±0.00 ^e	1.45±0.07 ^d	39.00±0.99 ^e	4.37±0.25 ^b	49.68±0.81 ^d
<i>M. excelsa</i>	2.05±0.07 ^{cd}	2.90±0.14 ^d	1.20±0.00 ^e	32.63±0.11 ^e	5.24±0.00 ^a	55.98±0.32 ^b
Plantain mid-rib	2.50±0.00 ^b	3.90±0.14 ^a	2.55±0.07 ^a	44.48±0.46 ^a	4.38±0.25 ^b	42.19±0.28 ^f
Corn cobs	2.40±0.14 ^{bc}	1.55±0.07 ^f	1.75±0.07 ^c	41.93±0.32 ^b	5.68±0.32 ^a	46.69±0.28 ^e
Mean	2.29±0.41	2.98±0.75	1.84±0.45	36.42±5.55	4.81±0.68	51.68±1.94
P value	<0.000	<0.000	<0.000***	<0.000	0.004	<0.000

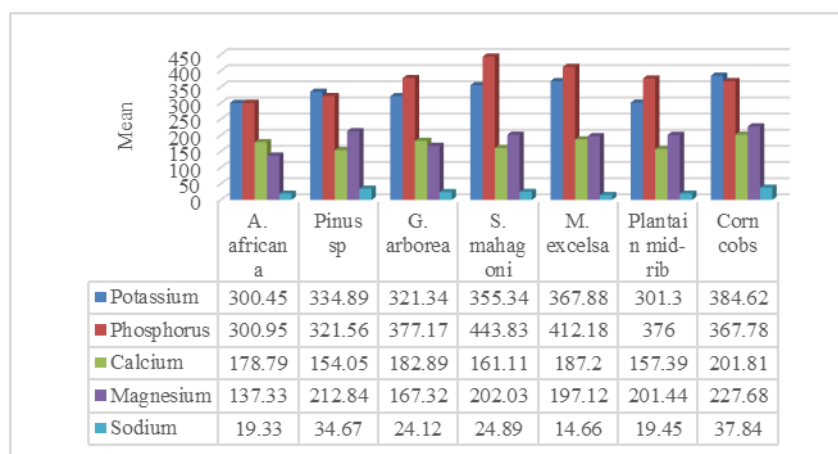


Mineral concentration of the substrates

The mineral concentration of the substrates varied significantly (Table no 2). CC had significantly the highest potassium concentration, followed by ME and the least was AA, which did not vary significantly from PM. For phosphorus, MA had significantly the highest, this was followed by ME while AA had significantly the least. CC recorded significantly the highest calcium content, followed by ME with PS having significantly the least. CC again had the highest magnesium content, followed by PS and the least was AA. In the same vein, CC had significantly the highest sodium content, this was closely followed by PS and ME had the least, which differed significantly from the rest.

Table no 2: Mineral concentration (mg/kg) of the different substrates before fungi inoculation

Substrate	Potassium	Phosphorus	Calcium	Magnesium	Sodium
<i>A. africana</i>	300.45±1.41 ^f	300.95±0.23 ^f	178.79±0.79 ^d	137.33±0.00 ^f	19.33±0.16 ^d
<i>Pinus</i> sp	334.89±0.33 ^d	321.56±0.32 ^e	154.05±0.39 ^e	212.84±1.03 ^b	34.67±0.64 ^b
<i>G. arborea</i>	321.34±0.47 ^e	377.17±1.34 ^c	182.89±0.31 ^c	167.32±0.00 ^e	24.12±0.00 ^c
<i>S. mahagoni</i>	355.34±0.00 ^c	443.83±1.02 ^a	161.11±0.00 ^e	202.03±0.54 ^c	24.89±0.33 ^c
<i>M. excelsa</i>	367.88±0.48 ^b	412.18±0.66 ^b	187.20±1.07 ^b	197.12±0.00 ^d	14.66±0.45 ^e
Plantain mid-rib	301.30±0.25 ^f	376.00±0.33 ^c	157.39±0.86 ^f	201.44±0.47 ^c	19.45±0.00 ^d
Corn cobs	384.62±0.70 ^a	367.78±0.78 ^d	201.81±0.84 ^a	227.68±0.63 ^a	37.84±0.56 ^a
Mean	337.97±31.41	371.35±47.11	174.75±17.04	192.25±29.14	24.99±8.13
P value	<0.000	<0.000	<0.000	<0.000	<0.000

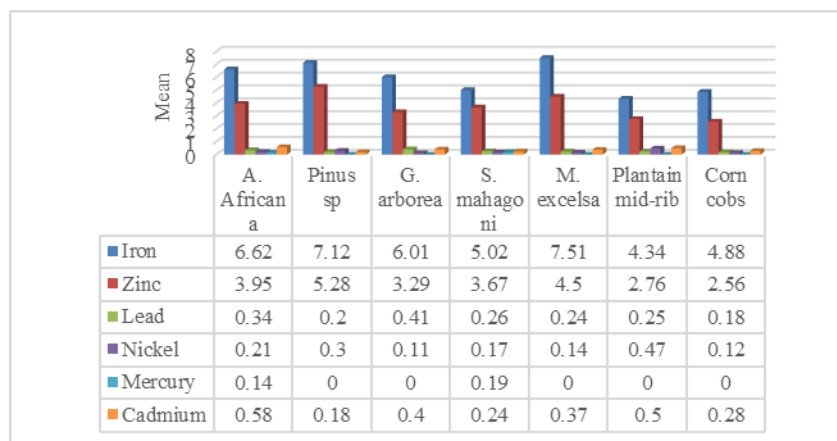


Heavy Metal concentration of the substrates

The heavy metal concentration of the substrates was also found to differ significantly among the various substrates (Table no 3). Aside iron and zinc which are needed for the body's metabolism, the other heavy metals assessed were very much in low concentration which is more or less negligible. ME had the highest iron concentration that varied significantly from the others except PS. PS and AA had iron contents that were statistically similar but statistically different from the rest. The least iron content was found in PM though it was not significantly different from CC and MA. For zinc content, PS had the highest zinc content that was not significantly different from that of ME but the zinc contents of PS and ME were significantly different from the rest of the substrates. The lowest zinc concentration was recorded in CC but it did not vary significantly from those of PM and GA. In terms of lead content, GA had significantly the highest content followed by AA and the least was CC, which was not significantly from PS. PM had significantly the highest nickel content, followed by PS and the least was GA, which had statistically similar nickel contents with CC, ME and MA. With the exception of AA and MA, mercury was not detected in the substrates. MA had significantly higher mercury content than AA. Significantly highest cadmium content was found in AA, this was closely followed by PM and the least was in PS.

Table no 3: Heavy Metal Concentration (mg/kg) of the different substrates before spawn inoculation

Substrate	Iron	Zinc	Lead	Nickel	Mercury	Cadmium
<i>A. Africana</i>	6.62±0.39 ^{bc}	3.95±0.23 ^b	0.34±0.03 ^b	0.21±0.00 ^c	0.14±0.03 ^b	0.58±0.06 ^a
<i>Pinus</i> sp	7.12±0.15 ^{ab}	5.28±0.23 ^a	0.20±0.01 ^{de}	0.30±0.02 ^b	0.00±0.00 ^c	0.18±0.01 ^e
<i>G. arborea</i>	6.01±0.16 ^c	3.29±0.00 ^{bcd}	0.41±0.01 ^a	0.11±0.00 ^d	0.00±0.00 ^c	0.40±0.02 ^c
<i>S. mahagoni</i>	5.02±0.17 ^d	3.67±0.16 ^{bc}	0.26±0.03 ^c	0.17±0.02 ^{cd}	0.19±0.00 ^a	0.24±0.02 ^{de}
<i>M. excelsa</i>	7.51±0.54 ^a	4.50±0.40 ^a	0.24±0.01 ^c	0.14±0.01 ^d	0.00±0.00 ^c	0.37±0.02 ^c
Plantain mid-rib	4.34±0.00 ^d	2.76±0.00 ^{cd}	0.25±0.02 ^c	0.47±0.05 ^a	0.00±0.00 ^c	0.50±0.04 ^b
Corn cobs	4.88±0.15 ^d	2.56±0.31 ^d	0.18±0.01 ^e	0.12±0.01 ^d	0.00±0.00 ^c	0.28±0.00 ^d
Mean	5.93±1.18	3.71±0.94	0.27±0.08	0.22±0.12	0.17±0.08	0.36±0.14
P value	<0.000	<0.000	<0.000	<0.000	<0.000	<0.000

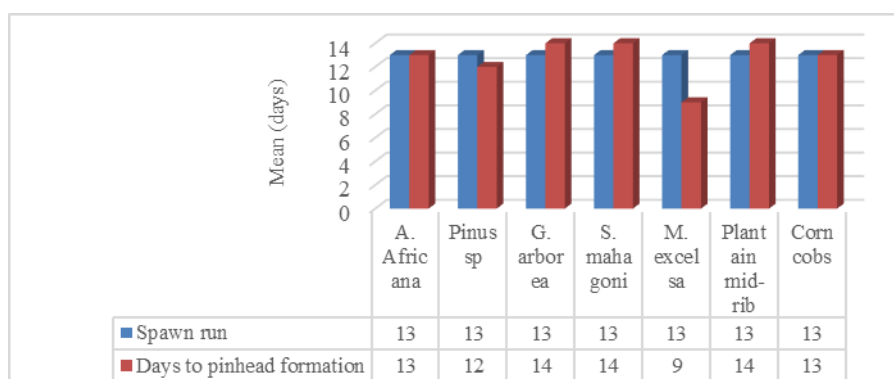


Effects of the substrates on spawn run and pin-head formation of *P. pulmonarius*

Spawn run was the same for all the substrates while days to pin-head formation of the mushroom differed (Table no 4). Pin-head formation of *P. pulmonarius* was fastest in ME (9 days) followed by PS (12 days) and longest in GA, MA and PM, which took 14 days.

Table no 4: Effects of substrates on spawn running and days to pinhead formation

Substrate	Spawn run	Days to pinhead formation
<i>A. africana</i>	13±0.00	13±0.00
<i>Pinus sp</i>	13±0.00	12±0.02
<i>G. arborea</i>	13±0.00	14±0.01
<i>S. mahagoni</i>	13±0.00	14±0.03
<i>M. excelsa</i>	13±0.00	9±0.00
Plantain mid-rib	13±0.00	14±0.02
Corn cobs	13±0.00	13±0.01
Mean	13±0.00	12.7±0.03
P value	0.82	0.41



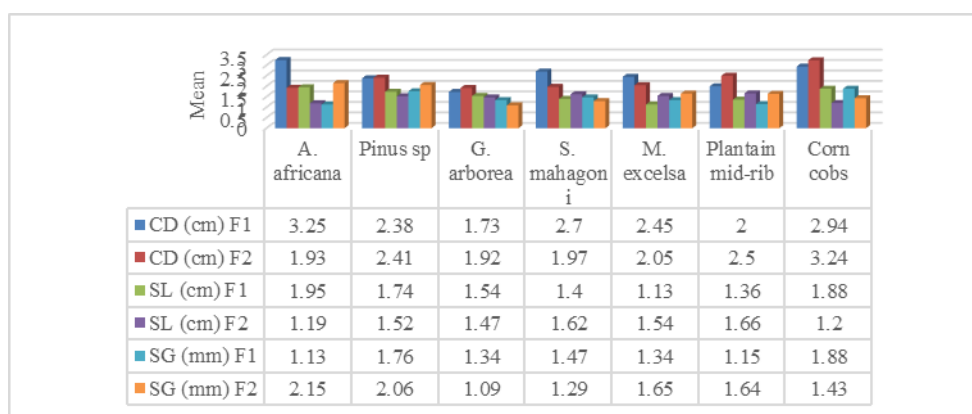
Growth response of *P. pulmonarius* cultivated on the different substrates

Results revealed that significant variations were not found in cap diameter and stipe lengths of the mushroom harvested from the different substrates in both flushes (Table no 5). However, in the first flush, maximum cap diameter was recorded for *P. pulmonarius* grown on AA, this was closely followed by CC and the minimum was recorded on GA. In the second flush, maximum cap diameter was recorded for *P. pulmonarius* grown on CC, followed by PM and the minimum was recorded on GA. For the stipe length, in the first flush, the highest stipe length was recorded for *P. pulmonarius* grown on AA, this was followed by CC and the lowest was on ME. There were significant variations found in stipe girth in both flushes (Table no 5). In the first flush, *P. pulmonarius* grown on CC had the highest stipe girth, which was not significantly different from that grown on PS but differed significantly from the rest. The least stipe girth was recorded for *P. pulmonarius* grown on AA but this did not differ significantly from those recorded for PM, ME, GA and MA. In the second flush, the highest stipe girth was recorded for *P. pulmonarius* grown on AA and it was statistically similar to that recorded on PS, which were significantly different from the others. This was followed by those recorded on ME and PM, which were statistically similar but significantly different from the rest. The lowest stipe girth was recorded on GA and

it was not significantly different from MA. Generally, the cap size and stipe length were higher in the first flush than in the second flush. On the contrary, the stipe girth was higher in the second flush than in the first.

Table no 5: Growth response of *P. pulmonarius* grown on the different substrates

Substrate	CD (cm) F1	CD (cm) F2	SL (cm) F1	SL (cm) F2	SG (mm) F1	SG (mm) F2
<i>A. africana</i>	3.25±0.05 ^a	1.93±0.43 ^a	1.95±1.75 ^a	1.19±0.32 ^a	1.13±0.18 ^c	2.15±0.77 ^a
<i>Pinus</i> sp	2.38±0.59 ^a	2.41±0.08 ^a	1.74±0.29 ^a	1.52±0.17 ^a	1.76±0.10 ^{ab}	2.06±0.23 ^a
<i>G. arborea</i>	1.73±0.75 ^a	1.92±0.85 ^a	1.54±0.20 ^a	1.47±0.12 ^a	1.34±0.12 ^c	1.09±0.66 ^d
<i>S. mahagoni</i>	2.70±0.04 ^a	1.97±1.29 ^a	1.40±0.11 ^a	1.62±0.42 ^a	1.47±0.12 ^{bc}	1.29±0.08 ^{cd}
<i>M. excelsa</i>	2.45±0.94 ^a	2.05±1.17 ^a	1.13±0.16 ^a	1.54±0.03 ^a	1.34±0.15 ^c	1.65±0.36 ^b
Plantain mid-rib	2.00±0.55 ^a	2.50±2.04 ^a	1.36±0.26 ^a	1.66±0.13 ^a	1.15±0.40 ^c	1.64±0.27 ^b
Corn cobs	2.94±0.43 ^a	3.24±0.48 ^a	1.88±1.19 ^a	1.20±0.30 ^a	1.88±0.35 ^a	1.43±0.13 ^{bc}
Mean	2.49±0.12	2.29±1.02	1.57±0.74	1.46±0.28	1.44±0.34	1.62±0.52
P value	0.743	0.740	0.870	0.174	0.008	0.048

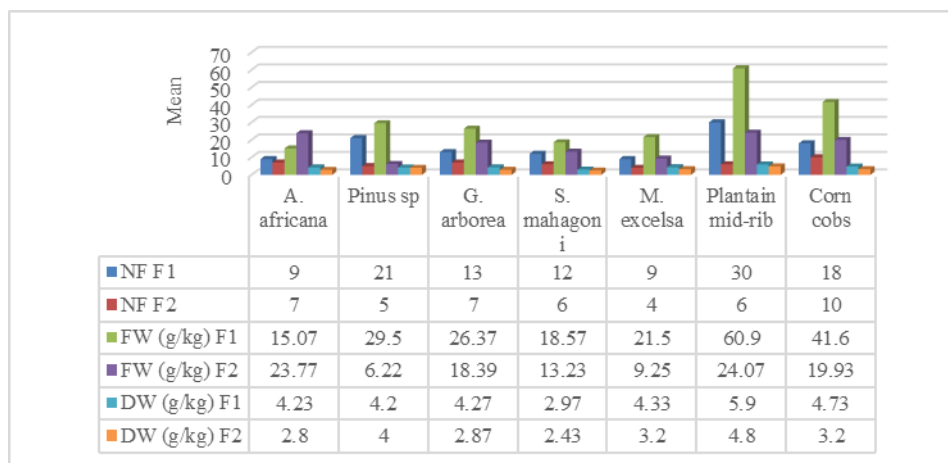


Yield and yield attributes of *P. pulmonarius* grown on the different substrates

Results revealed significant variations in the number of fruiting bodies, fresh weight and dry weight of *P. pulmonarius* cultivated on the different substrates in both flushes with the exception of dry weight in the second flush (Table no 6). In the first flush, the number of fruiting bodies produced by *P. pulmonarius* was significantly highest on PM. This was closely followed by PS and CC, which had statistically similar values and the least were on ME and AA. In the second flush, the number of fruiting bodies produced by *P. pulmonarius* was significantly highest on CC, followed by AA and GA, which had same values that differed significantly from ME, which had the least. For the fresh weight and in the first flush, *P. pulmonarius* grown on PM had significantly the highest fresh weight, followed by CC and the least was on AA, which did not vary different from MA. In the second flush, *P. pulmonarius* grown on PM again had the highest fresh weight and was not significantly different from that on AA but was from the others. The least fresh weight was recorded for PS, which was statistically similar with ME. In the case of the dry weight, in the first flush, *P. pulmonarius* grown on PM had significantly the highest dry weight, followed by CC, and the least was on MA. In the second flush, no significant variation was observed. However, the highest dry weight of the mushroom was recorded on PM followed by PS and the least was MA.

Table no 6: Yield and yield attributes of *Pleurotus pulmonarius* grown on the different substrates

Substrate	NF F1	NF F2	FW (g/kg) F1	FW (g/kg) F2	DW (g/kg) F1	DW (g/kg) F2
<i>A. africana</i>	9.00±1.21 ^d	7.00±0.51 ^b	15.07±1.05 ^c	23.77±3.18 ^a	4.23±1.00 ^b	2.80±1.85 ^a
<i>Pinus</i> sp	21.00±1.32 ^b	5.00±0.44 ^{bc}	29.50±1.99 ^c	6.22±0.33 ^d	4.20±1.00 ^b	4.00±0.20 ^a
<i>G. arborea</i>	13.00±0.62 ^c	7.00±1.08 ^b	26.37±3.10 ^{cd}	18.39±1.13 ^b	4.27±0.55 ^b	2.87±1.03 ^a
<i>S. mahagoni</i>	12.00±1.15 ^c	6.00±1.73 ^{bc}	18.57±0.05 ^c	13.23±0.40 ^c	2.97±0.23 ^c	2.43±0.49 ^a
<i>M. excelsa</i>	9.00±0.51 ^d	4.00±1.15 ^c	21.50±0.63 ^d	9.25±2.21 ^d	4.33±0.72 ^b	3.20±0.70 ^a
Plantain mid-rib	30.00±1.39 ^a	6.00±1.53 ^{bc}	60.90±2.13 ^a	24.07±2.83 ^a	5.90±1.56 ^a	4.80±0.43 ^a
Corn cobs	18.00±1.29 ^b	10.00±1.08 ^a	41.60±1.82 ^b	19.93±1.19 ^b	4.73±2.48 ^b	3.20±1.23 ^a
Mean	16.00±1.27	7.00±0.51	30.50±2.72	16.41±0.54	4.38±1.35	3.33±1.36
P value	0.016	0.045	0.025	0.001	0.029	0.414

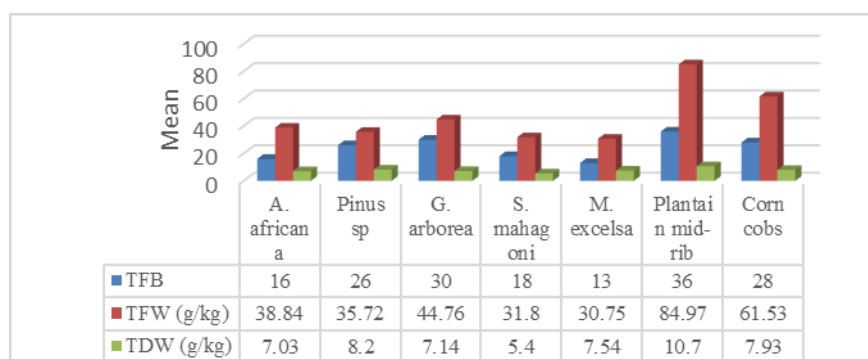


Total yield

Table no 7 shows the total number of fruiting bodies (TFB) produced in the two flushes by *P. pulmonarius* cultivated on the different substrates including the total fresh weight (TFW) and total dry weight (TDW) of the fruiting bodies in the two flushes. Results showed that significant ($P < 0.05$) variations existed among the substrates in the total number of fruiting bodies, the total fresh and dry weights. *P. pulmonarius* cultivated on plantain mid-rib produced the highest number of fruiting bodies that varied significantly ($P < 0.05$) from the others closely followed by that on GA, which did not differ significantly from that on CC but was from the rest and the least was that on ME. For the total fresh weight, *P. pulmonarius* grown on PM had significantly the highest value closely followed by that on CC and the lowest was on ME. Similarly, *P. pulmonarius* grown on PM had significantly the highest total dry weight of fruiting bodies, this was followed by that on PS and the least was that on MA.

Table no 7: Total number of fruiting bodies, fresh weight and dry weight of *P. pulmonarius* cultivated on the various substrates

Substrate	TFB	TFW (g/kg)	TDW (g/kg)
<i>A. africana</i>	16±0.22 ^d	38.84±1.20 ^d	7.03±0.02 ^c
<i>Pinus sp</i>	26±0.14 ^c	35.72±0.24 ^e	8.20±0.00 ^b
<i>G. arborea</i>	30±0.40 ^b	44.76±1.00 ^c	7.14±0.12 ^c
<i>S. mahagoni</i>	18±0.10 ^d	31.80±0.02 ^f	5.4±0.01 ^d
<i>M. excelsa</i>	13±0.02 ^e	30.75±0.40 ^f	7.54±0.80 ^c
Plantain mid-rib	36±0.25 ^a	84.97±1.20 ^a	10.7±0.22 ^a
Corn cobs	28±0.08 ^{bc}	61.53±0.45 ^b	7.93±0.20 ^{bc}
Mean	28±0.10	52.01±1.00	8.87±0.05
P value	0.026	0.0001	0.029



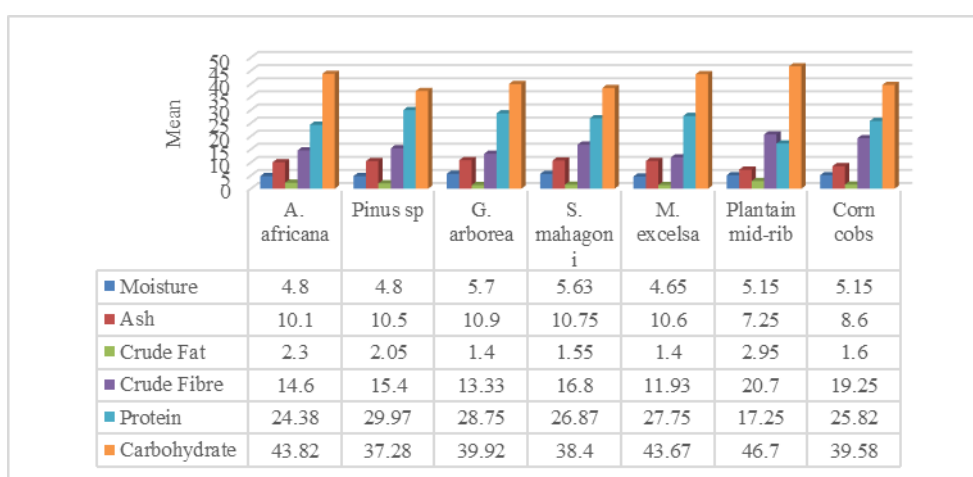
Proximate composition of *P. pulmonarius* grown on the different substrates

Results on proximate composition of *P. pulmonarius* grown on the different substrates showed they were significant differences among the substrates (Table no 8). *P. pulmonarius* cultivated on GA had the highest moisture content that was not significantly different from that on MA but was from the others. The least was recorded on ME but it was not significantly different from AA and PS. For ash content, the highest value was found in the mushroom grown on GA, which varied significantly from CC and PM only. The least was recorded on PM. In the case of the crude fat content, significantly highest value was recorded for *P. pulmonarius* cultivated

on PM, this was followed by AA, which had statistically similar values with PS. Again the least crude fat content was found in the mushroom grown on ME. In the same vein, crude fibre was significantly highest in *P. pulmonarius* grown on PM, followed by CC and the least was ME. Protein content was significantly highest in the mushroom grown on PS, followed by GA and then PM, which was the least. On the other hand, the carbohydrate content was significantly highest in the mushroom grown on PM, followed by AA and the least was PS.

Table no 8: Proximate composition (%) of *P. pulmonarius* grown on different substrates

Substrate	Moisture	Ash	Crude Fat	Crude Fibre	Protein	Carbohydrate
<i>A. africana</i>	4.80±0.14 ^c	10.10±0.14 ^b	2.30±0.14 ^b	14.60±0.00 ^d	24.38±0.25 ^f	43.82±0.39 ^b
<i>Pinus</i> sp	4.80±0.00 ^c	10.50±0.00 ^{ab}	2.05±0.07 ^b	15.40±0.07 ^d	29.97±0.25 ^a	37.28±0.11 ^e
<i>G. arborea</i>	5.70±0.14 ^a	10.90±0.14 ^a	1.40±0.14 ^c	13.33±0.11 ^e	28.75±0.00 ^b	39.92±0.53 ^c
<i>S. mahagoni</i>	5.63±0.11 ^a	10.75±0.21 ^a	1.55±0.21 ^c	16.80±0.14 ^c	26.87±0.25 ^d	38.40±0.42 ^d
<i>M. excelsa</i>	4.65±0.21 ^c	10.60±0.14 ^a	1.40±0.00 ^c	11.93±1.03 ^f	27.75±0.00 ^c	43.67±0.67 ^b
Plantain mid-rib	5.15±0.07 ^b	7.25±0.35 ^d	2.95±0.21 ^a	20.70±0.57 ^a	17.25±2.00 ^g	46.70±1.64 ^a
Corn cobs	5.15±0.07 ^b	8.60±0.14 ^c	1.60±0.14 ^c	19.25±0.10 ^b	25.82±0.11 ^e	39.58±0.28 ^c
Mean	5.13±0.41	9.81±1.32	1.89±0.57	16.00±3.04	25.83±4.04	41.34±3.31
P value	<0.000	<0.000	<0.000	<0.000	0.004	<0.000

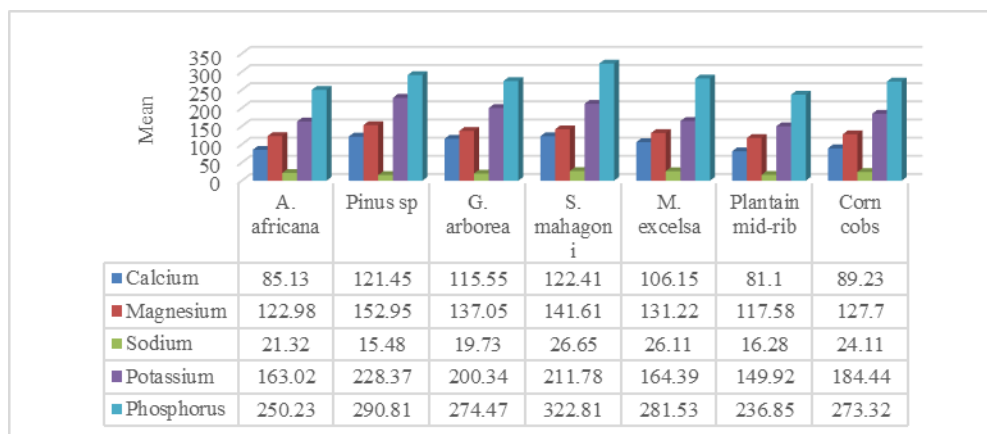


Mineral composition of *P. pulmonarius* cultivated on the different substrates

The mineral content of *P. pulmonarius* harvested from the different substrates varied significantly ($P < 0.05$) (Table no 9). Calcium content was highest in *P. pulmonarius* grown on MA although this did not vary significantly from PS but did vary from the others, with the least on PM. Magnesium content was significantly highest in the mushroom from PS followed by GA and the least on PM. For sodium content, the highest value was recorded on MA, which had similar statistical value with ME that differed significantly from the rest. The least sodium content was recorded on PS. Potassium content of the mushroom grown on the different substrates followed the same trend as that of magnesium content with PS having significantly the highest value followed by GA and the least on PM. Phosphorus content was significantly highest in the mushroom on MA, followed by PS and the least on PM.

Table no 9: Mineral composition (mg/kg) of *P. pulmonarius* cultivated on the different substrates

Substrate	Calcium	Magnesium	Sodium	Potassium	Phosphorus
<i>A. africana</i>	85.13±0.15 ^{df}	122.98±0.20 ^f	21.32±0.28 ^c	163.02±0.19 ^e	250.23±0.00 ^e
<i>Pinus</i> sp	121.45±1.88 ^a	152.95±0.00 ^a	15.48±0.27 ^f	228.37±0.50 ^a	290.81±2.16 ^b
<i>G. arborea</i>	115.55±0.33 ^b	137.05±0.10 ^c	19.73±0.32 ^d	200.34±0.00 ^c	274.47±0.35 ^d
<i>S. mahagoni</i>	122.41±0.63 ^a	141.61±0.53 ^b	26.65±0.00 ^a	211.78±0.81 ^b	322.81±0.37 ^a
<i>M. excelsa</i>	106.15±0.00 ^c	131.22±0.00 ^d	26.11±0.00 ^a	164.39±0.39 ^e	281.53±0.53 ^c
Plantain mid-rib	81.10±0.00 ^f	117.58±0.65 ^e	16.28±0.09 ^e	149.92±0.28 ^f	236.85±0.00 ^f
Corn cobs	89.23±0.04 ^d	127.70±0.59 ^c	24.11±0.00 ^b	184.44±0.45 ^d	273.32±0.18 ^d
Mean	103.00±1.08	133.01±1.49	21.38±0.32	186.04±2.64	275.72±2.79
P value	<0.000	<0.000	<0.000	<0.000	<0.000

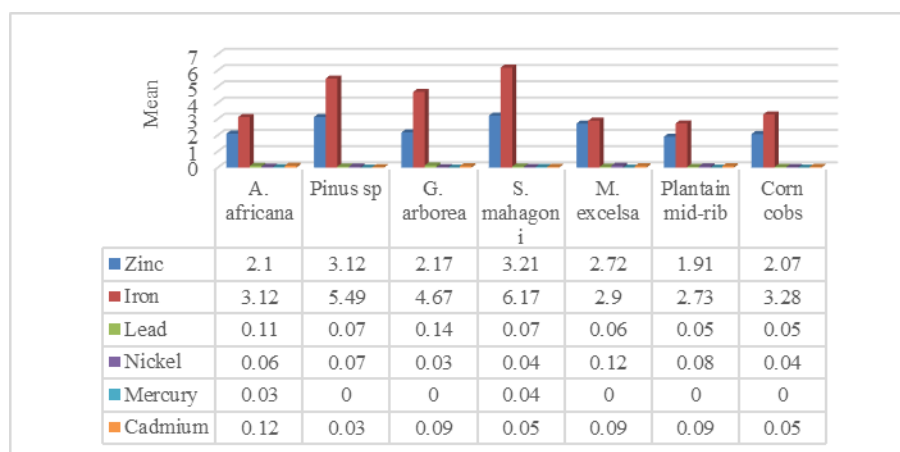


Heavy metal composition of *P. pulmonarius* cultivated on the different substrates

Significant variation existed among the substrates on heavy metal composition of the harvested mushroom (Table no 10). *P. pulmonarius* harvested from MA had the highest zinc concentration, which did not vary significantly from that on PS but did from the rest. This was followed by ME and the lowest was on PM. Similarly, iron concentration was significantly highest in the mushroom on MA, followed by PS and again the lowest was on PM. Lead content was significantly highest in the mushroom on GA, followed by AA and the lowest was again on PM, although the value did not vary significantly from that on CC. The highest nickel concentration was recorded on *P. pulmonarius* grown on ME. This was found to vary significantly from the others. Following this was PM and the least was on GA. Mercury was not detected in the *P. pulmonarius* grown on the different substrates except on MA and AA with mercury concentration being higher in MA than AA. In the case of cadmium, *P. pulmonarius* grown on AA had significantly the highest concentration of cadmium, followed by GA, ME and PM, which had same values while the least was on PS.

Table no 10: Heavy Metals concentration (mg/kg) of *P. pulmonarius* grown on the different substrates

Substrate	Zinc	Iron	Lead	Nickel	Mercury	Cadmium
<i>A. africana</i>	2.10±0.02 ^d	3.12±0.16 ^d	0.11±0.00 ^b	0.06±0.01 ^c	0.03±0.01	0.12±0.01 ^a
<i>Pinus sp</i>	3.12±0.01 ^a	5.49±0.23 ^b	0.07±0.00 ^c	0.07±0.01 ^{bc}	0.00±0.00	0.03±0.00 ^d
<i>G. arborea</i>	2.17±0.08 ^c	4.67±0.00 ^c	0.14±0.01 ^a	0.03±0.00 ^d	0.00±0.00	0.09±0.01 ^b
<i>S. mahagoni</i>	3.21±0.00 ^a	6.17±0.22 ^a	0.07±0.01 ^c	0.04±0.01 ^d	0.04±0.01	0.05±0.01 ^c
<i>M. excelsa</i>	2.72±0.09 ^b	2.90±0.17 ^{de}	0.06±0.01 ^d	0.12±0.01 ^a	0.00±0.00	0.09±0.01 ^b
Plantain mid-rib	1.91±0.06 ^d	2.73±0.08 ^e	0.05±0.00 ^e	0.08±0.01 ^b	0.00±0.00	0.09±0.00 ^b
Corn cobs	2.07±0.08 ^d	3.28±0.22 ^d	0.05±0.01 ^c	0.04±0.00 ^d	0.00±0.00	0.05±0.00 ^c
Mean	2.47±0.52	4.05±1.33	0.08±0.03	0.06±0.03	0.01±0.00	0.07±0.03
P value	<0.000	<0.000	<0.000	<0.000	<0.000	<0.000



IV. Discussion

The nutrient composition of substrates employed in the cultivation of mushrooms plays a fundamental role in determining the nutritional quality and yield of fruiting bodies of mushroom. The nutrient composition of the substrates used in this study varied. They had more carbohydrate (42.19 – 60.98 %) and crude fibre (27.80 –

44.48 %) compared to the other food constituents with crude fat (1.20 – 2.55 %) being the least. This result agrees with that of Obiaigwe JA *et al.* that mushroom substrates have higher contents of carbohydrate and fibre¹⁸. Carbohydrates and fibres are critical in oyster mushroom cultivation. Carbohydrates in the form of polysaccharides (such as cellulose and hemicellulose) are abundant in lignocellulosic materials used in mushroom cultivation. Carbohydrate in mushroom substrate serves as a source of carbon for mycelia growth. Oyster mushrooms are able to degrade this polysaccharide for their growth and fruiting¹⁹. It is reported that substrates with high fiber content provide structural support and are rich in lignocellulosic material, which the mushroom can degrade and utilize²⁰. Generally, carbohydrate was higher in the sawdust than in the agro-wastes (PM and CC). On the contrary, crude fibre was higher in the agro-wastes than in the sawdust. Although sawdust is a fibrous material, it does not have digestible fibre. This may explain the higher fibre content in the non-wood agro wastes in relation to the sawdust wastes. The fibre and protein contents of 41.93% and 5.68%, respectively obtained in CC in this study was higher than the values of 34.00% and 3.92%, respectively reported by Yusuf KO *et al.*²¹.

The substrates had appreciable amounts of minerals with higher contents of potassium and phosphorus than the other minerals. This will impact on the nutrient composition of the mushroom. Heavy metals (Pb, Ni, Hg and Cd) were all less than 1.00 mg/kg in all the substrates. Mercury was only detected in two of the substrates, AA and MA. This is an indication that the substrates are safe for mushroom cultivation. All the substrates (both the sawdust and non-wood wastes) used in this study supported the growth and fructification of *P. pulmonarius* (Plate 2) although the response varied significantly amongst the substrates. This is attributed to the substrate's composition particularly to the high contents of carbohydrate and fibre. The presence of substantial amounts of carbohydrate in mushroom substrates has been reported to support vigorous proliferation of mushroom mycelia and growth²². These results collaborate with the findings of Agba MO *et al.*¹², Hoa HT *et al.* and Jonathan SG *et al.*, on *P. ostreatus*, *P. cystidiosus* and *P. pulmonarius*, respectively^{12,11,23}. Oyster mushrooms are capable of degrading cellulose, hemicellulose and lignin which are major components of many agricultural and forestry wastes by secreting lignocellulolytic enzymes, hence they can be grown on various agro- and forestry by-products^{9,20}.

The period for full colonization of the mycelia on the substrate referred to as spawn run obtained in this study (13 days) was shorter than that (35 days) reported by Akinmusire OO *et al.* on the same mushroom and on sawdust substrate²⁴. Fast colonization of mushroom mycelia on substrates will greatly reduce growth of competitive microorganisms and consequently spawn contamination²³. Pin-head (primordium initiation) was observed following the invasion of the substrates by *P. pulmonarius* mycelia. The results of this study showed that pin-head appeared earlier in *Milicia excelsa* (ME) than the other substrates where it appeared between 12 (PS) and 14 days (in GA, MA and PM). The early appearance of pin-head in ME could be as a result of the chemical composition of the substrate, particularly because of its relatively high nitrogen contents. Chiejina and Osibe reported that growth of mushroom mycelia depends on the nitrogen content of the substrate²⁵. Naraian R *et al.* reported that mycelia growth and primordia development depend heavily on the type of lignocellulosic materials especially on the C:N of the substrates²⁶.



Plate no 2: Growth of *P. pulmonarius* in the substrates

Fructification of *P. pulmonarius* occurred in all the substrates (Plate no 3). This indicates that these substrates contained nutrients that supported the growth of the mushrooms²⁷. In addition, *Pleurotus* spp. have a high saprophytic ability and can successfully grow on a variety of lignocellulosic substrates^{9,28,29}. Fruiting bodies of *P. pulmonarius* were harvested from the various substrates in two flushes. Results of this study revealed that the substrates had no significant effect on the growth of the mushroom except on the stipe girth in both flushes. This is contrary to the result of Markson AA *et al.* who reported significant variations in the growth of *P. ostreatus* mushroom grown on different substrates³⁰. Ajonina and Tatah stated that the cap size is one of the contributing characteristics of mushroom yield³¹. *P. pulmonarius* cap size ranged from 1.73 cm (in GA) – 3.25 cm (AA) in the first flush and then 1.92 cm (GA) – 3.24 cm (CC) in the second flush. The stipe length on the other hand ranged from 1.13 cm (in ME) – 1.95 cm (AA) in the first flush. In the second flush, the stipe length ranged from 1.19 cm

(AA) – 1.62 cm (MA). These were lower than the values reported by Okwulehie IC *et al.* who obtained highest cap size of 3.83 cm and stipe length of 2.77 cm in *P. pulmonarius* on oil palm bunch substrate treated with 0.4% HCl and 0.3% HCl, respectively³².



Plate no 3: Fruiting bodies of *P. pulmonarius* on the various substrates

The number of fruiting bodies produced by the mushroom may play a role in the size of the fruiting bodies in terms of pileus diameter, stipe length and stipe girth in addition to the substrate quality. Higher number of fruiting bodies may result in smaller sizes of the fruiting bodies and vice versa as they will be more competition for nutrients. Kimenju JW *et al.* also reported that the fewer the mushroom the wider the pileus¹⁵. *P. pulmonarius* cultivated on GA produced fruiting bodies with the least cap size in both flushes. GA substrate had lower protein (nitrogen content) and fibre contents in relation to the other substrates. This may partly explain the lowest cap size observed in the fruiting bodies of *P. pulmonarius* harvested from GA. According to Agba MO *et al.* preferred mushrooms with marketable quality are those with bigger pileus, bigger stipes but short stipes¹².

The substrates had a significant influence on the number of fruiting bodies of *P. pulmonarius* produced. In the first flush, number of effective fruiting bodies of the mushroom ranged from 9 (in AA and ME) – 30 (in PM) and then 4 (in ME) – 10 (in CC) in the second flush. These values were higher than values obtained from previous studies^{33,34}. Shah ZA *et al.* reported average number of fruiting bodies in the range of 7 – 22 for oyster mushrooms³⁵. The number of fruiting bodies produced by *P. pulmonarius* on the various substrates was higher in the first flush than in the second flush. This agrees with the findings of Frimpong-Manson S *et al.* who reported that the number of fruiting bodies produced per flush decreased from flush to flush³⁶. Uptake of the available nutrients in the substrates by the harvested mushrooms in the first flush will cause a reduction in the nutrient content of the substrate. This may explain the reason for the decrease in the number of effective fruiting bodies in the second flush. Total number of effective fruiting bodies harvested from the substrates decreased in this order- PM>CC>PS>GA>MA>AA>ME. The variation in the number of fruiting bodies observed in this study may be attributable to the difference in substrate and substrate composition. Mondal SR *et al.* obtained effective fruiting bodies of oyster mushroom, *P. florida* in the range of 8.5 to 37.25 and opined that the number produced depended on substrate types used for cultivation³⁷.

Mushroom weight either fresh or dry weight is a vital agronomic parameter for assessing the effectiveness of fungi as bio-agents in the conversion of inedible organic wastes directly into nutritious human food³⁸. The fresh weight of mushroom is a direct indicator of commercial productivity. The fresh weight in both flushes was consistently highest for the mushroom grown on PM. It was lowest on MA and PS in first and second flush, respectively. Total fresh weight of the mushroom on the different substrates decreased in this order: PM>CC>GA>PS>MA>ME>AA. The high fresh weight observed in the mushroom grown on PM could be due to other factors aside the composition of the substrate. Frimpong-Manso S *et al.* reported highest weight of an oyster mushroom, *P. ostreatus* on rice husk and attributed it to physical nature, high porosity and high level of aeration of the rice husk and not necessarily on the substrate composition³⁶. The results of this study indicated that *P. pulmonarius* grown on the non-wood agro-wastes (PM and CC) recorded higher mushroom fresh weight in comparison with the sawdust from the tree hardwood species. This result agrees with the findings of Aswathy S *et al.* who reported significantly higher yields of *Pleurotus tuber-regium* cultivated on paddy straw substrate compared to sawdust³⁹. It also agrees with the result of Philippoussis A *et al.* who found that *Pleurotus* species had higher fresh weights on cereal straw than on beech or oak sawdust⁴⁰. Non-wood agro-wastes are rich in easily degradable lignocellulosic materials that promote faster colonization and fruiting. On the contrary, sawdust, especially from hardwood species, is often more recalcitrant due to its high lignin content and low nitrogen levels. This slows down mushroom development and reduces overall yield^{35,41}. In addition, non-wood agro-wastes tend to have better moisture-holding capacities than sawdust, which supports optimal humidity levels needed for mushroom development. This property enhances the microenvironment for fruiting, thereby increasing fresh weight⁴². The dry weight of the mushroom in the first flush ranged from 2.97 – 5.90 g/kg on MA/PM, respectively.

In the second flush, it ranged from 2.43 to 4.8 On MA and PM, respectively. It also decreased in this order: PM>PS>CC>ME>GA>AA>MA. Similar to the fresh weight, *P. pulmonarius* grown on PM had the highest dry weight. This supports previous research that the choice of substrate significantly affects mushroom yield⁴².

The nutrient composition of *P. pulmonarius* grown on the different substrates varied. This is attributable to the difference in the substrate compositions as reported earlier in this study. Mushrooms derive their food from the substrate on which they grow, therefore substrates will have an impact on the nutrient composition of mushrooms. Proximate analysis results showed that *P. pulmonarius* had an appreciable quantities of food nutrients. It has high contents of ash (7.25 – 10.90 %), crude fibre (11.93 – 20.70%), protein (17.25 – 29.97 %) and carbohydrate (37.28 – 46.70 %) but low content of crude fat (1.40 – 2.95 %). This is an indication that the substrates, which were reportedly high in crude fibre and carbohydrate had a positive impact on the nutrient composition of the harvested mushroom. This result is in conformity with the findings of some workers who reported that mushrooms have high nutritional components that make them viable as a food source^{2,3,5}. The range of ash, fibre and protein contents obtained in this study is similar to the range of 2.1 – 9.4 %, 6.21 – 54.0 % and 16.07 – 25.15 %, respectively reported by Irshad A⁴³.

Protein is essential for good health since it helps the body grow, repair, and maintain body tissue. The protein and fibre contents obtained in this study are within the range reported by Cheung PCK, who obtained protein content in oyster mushrooms in the range of 10.5 – 30.4 % and crude fiber range of 1.44 – 44.0 %⁴⁴. The high fiber content of this mushroom makes them easily digestible. The low-fat content obtained in this study agrees with the report of several authors with values ranging from 0.64 to 3.24 %^{43,45,46}. The low-fat content of *P. pulmonarius* is an indication that it is a food with a lower caloric value and suitable for patients with cardiac problems or at risk of lipid induced disorders. The most important function of lipids in human bodies has been to produce energy for muscle and body activities, to maintain body temperature, support digestion, and absorb food nutrients⁴⁷.

Minerals in diet are essential for metabolic reactions, healthy bone formation, transmission of nerve impulses, regulation of water and salt balance⁴⁸. The mineral content of *P. pulmonarius* varied with the different substrates used. In general, the results revealed that *P. pulmonarius* accumulated more phosphorus in comparison with other minerals, with potassium following closely and zinc being the least accumulated. This mineral uptake pattern indicates that *P. pulmonarius* may have a higher affinity or more efficient mechanisms for phosphorus absorption. This may be influenced by factors such as substrate composition, mycelial physiology, and environmental conditions. *P. pulmonarius* grown on GA substrate accumulated the highest concentrations of phosphorus, calcium, sodium, zinc and iron, while potassium and magnesium levels peaked in *P. pulmonarius* grown on PM substrate. Conversely, mushrooms grown on Plantain mid-rib substrate exhibited the lowest mineral concentrations across all assessed elements. This result aligns with that by Okwulehie IC *et al.* who reported that *P. pulmonarius* cultivated on Khaya ivorensis sawdust exhibited elevated mineral concentrations, including phosphorus and calcium, suggesting that lignocellulosic substrates can enhance mineral uptake in mushrooms¹⁴.

Assessing the heavy metal composition of *P. pulmonarius* grown on the different substrate is important due to the health risk associated with the consumption of foods with heavy metal contamination. The presence of heavy metals (Pb, Ni, Hg and Cd) in the fruiting bodies of *P. pulmonarius* grown on the different substrates is not surprising since they were also found in the different substrates used for this study. This is an indication that heavy metal accumulation in mushroom tissue is substrate dependent. Pb ranged from 0.05 – 0.14 mg/kg; Ni from 0.03 – 0.12 mg/kg; Hg from 0.00 – 0.04 mg/kg and Cd from 0.02-0.12mg/kg. However, the quantities of these heavy metals in the studied mushroom were very minute and below their permissible limit recommended by FAO/WHO. The range of Pb concentration obtained in this study was within the range of 0.04 – 0.35 mg/kg reported by Ihugba UA *et al.* in wild samples of *P. tuberregium* consumed within Imo State, Nigeria⁴⁹. Ihugba UA *et al.* also found mean concentration of 0.0001 mg/kg of nickel in wild *P. tuberregium*⁴⁹. Quarcoo and Adotey reported the range of 0.04 – 0.23 mg/kg of lead and 0.04 mg/kg of mercury in *P. ostreatus* and *Termitomyces clypeatus*⁵⁰. These results are comparable with the results obtained in this study.

The presence of Cd in food is of particular concern because Cd is known to be a toxic element as it inhibits many life processes. The range of Cd obtained in the present study is lower than the ranges of 0.35 – 0.57 mg/kg and 2.25 - 4.88 mg/kg reported by Quarcoo and Adotey and Udochukwu U *et al.*, respectively in wild edible mushrooms^{50,51}. Siric I *et al.* analyzed the heavy metal concentration of two wild oyster mushroom species, *P. ostreatus* and *P. djamor* and reported cadmium concentration of 0.10 and 0.08 mg/kg, iron conc. of 28.49 and 27.15 mg/kg, zinc conc. of 18.15 and 15.75 mg/kg in *P. ostreatus* and *P. djamor*, respectively⁵².

It is reported that exposures to lead, cadmium and mercury are the main threats to human health from heavy metals^{53,54}. Lead is toxic to the heart, bones, intestines, kidneys, reproductive and nervous systems. It interferes with the development of the nervous system and is therefore particularly toxic to children, causing potentially permanent learning and behavior disorders⁵⁴. However, the range of the heavy metals reported in this study may be considered too low to give any cause for concern. They were all below the health-risk threshold and therefore safe for consumption.

V. Conclusion

This study demonstrated that the substrates used contained appreciable amounts of nutrients, though in varying concentrations, which supported the growth and fructification of *Pleurotus pulmonarius*. Carbohydrates and fiber were the predominant constituents of the substrates. The growth and yield performance of *P. pulmonarius* was dependent on the substrate type, with non-wood agro-wastes outperforming sawdust-based substrates. Among them, the PM substrate produced the highest yield and was considered the most effective. *P. pulmonarius* cultivated on all substrates showed notable nutrient content, particularly high levels of fiber, protein, and carbohydrates, with low fat content. Phosphorus and potassium levels were also relatively high. Importantly, the concentrations of heavy metals were below safety thresholds, indicating that the mushrooms are safe for consumption.

Acknowledgement

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