

## **Influence of Some Organic Supplements on the Growth and Yield of *Pleurotus pulmonarius* (Fries) *Quelet* on Sawdust Substrate**

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### **Abstract**

*This study investigated improved, cost effective organic wastes management and sustainable methods of *Pleurotus pulmonarius* cultivation. The cultivation of edible mushroom, *P. pulmonarius* was carried out using organic supplements such as corn bran (CB), wheat bran (WB), rice bran (RB), palm kernel cake (PKC), groundnut cake (GNC), soya meal (SM) and poultry manure (PM) mixed with the sawdust from *Gmelina aborea* wood species. These organic supplements- sawdust mixtures were made into compost heaps, and sawdust to which no supplement was added served as the control (CON). The primordial initiation was observed for the first time at approximately 30 days after spawning on CB and PKC. The biological and production efficiencies in all the supplemented substrates were increased over un-supplemented control. However, SM was found to be the best supplement producing 58.37% and 22.60% biological and production efficiencies respectively. PKC was the second best additive producing 56.92 and 22.43% biological and production efficiencies respectively. Shortest days to substrate colonization (21.86) and primordial initiation (30.86) were observed on PKC when compared with other supplements. These results showed that *P. pulmonarius* responded differently to organic nutrient sources and also indicated that the addition of these supplements to basal sawdust substrate necessitated greater supply of nutrients in the supplements which increased the mushroom yield, biological and production efficiencies.*

**Key words:** *Pleurotus pulmonarius*, supplements, growth and yield.

### **Introduction**

Food rich in essential nutrients like protein, vitamins and minerals obtainable from meat, fish, eggs, beans, fruits and vegetables are not always within the reach of the rural poor and the levels of the intake of these food values determine their health status.

In Nigeria, carbohydrate is culturally the major source of food intake, and it is one of the major reasons why malnutrition is rampant in the country (Iyangbe and Orewa, 2009). Majority of Nigerians do not consume the Food and Agriculture Organization (FAO) recommended 60g per person daily protein requirement for individuals (FAO, 1992) Nigeria's per capita daily protein intake is estimated to be 45.4g (ICN,1992) as against the FAO's minimum protein recommendation. Proximate analysis of mineral and energy content of edible mushrooms indicate their potentials for use as sources of good quality food and dietary supplements. The amount of crude protein, ash, and crude fibre found in

most edible mushroom species compare favourably with, and in some cases surpasses those of some fruits, vegetables and legumes except groundnut and soya beans (Ologhogbo, 1981; Aletor and Akdetimi, 1989; Mallavadhan, *et al*, 2006; Jegadesh *et al*, 2010). In view of the increased popularity of health foods, the dietary and medicinal importance of mushrooms cannot be over emphasized; it is therefore desirable to encourage the production and consumption of edible mushroom species.

In nature, *Pleurotus sp* mostly grow on the dead part of plants which are generally poor in nutrients and vitamins. It has been established that both mycelia growth and fruiting body development depend on lignocellulosic materials, particularly the carbon: nitrogen ratio (C:N) which has been reported to be 50:1 in various substrates (Balakrishnan and Nair, 1995). Substrates for mushroom growing have been supplemented with organic materials such as rice bran, earthworm cast, wheat bran, grain flour, wood shaving, humus and cocoa shell. (Madhab and Das, 1989; Peng *et al.*, 2000; Idowu *et al*, 2009). Incorporation of organic substances into the basic substrates for growing mushrooms is to increase or boost mushroom yields. This research was therefore aimed at determining the effect of the different organic supplements on the growth, yield and biological efficiency of the mushroom, *P. pulmonarius*.

### **Materials and methods**

The experiment was carried out at the mushroom production unit of National Horticultural Research Institute, Ibadan Nigeria. The primary inoculum was prepared from the fresh fruiting body of *P. pulmonarius* by tissue culture, which was maintained on potato dextrose agar slant incubated at room temperature for 14days after which they were stored at 4°C until needed.

### **Substrate Preparation**

Sawdust of *Gmelina aborea* was collected from Forestry Research Institute of Nigeria, Ibadan and used as basal substrate. The sawdust were divided into portions to which 10% w/w of the following organic supplements were added, corn bran (CB), wheat bran (WB), rice bran (RB), palm kernel cake (PKC), groundnut cake (GNC), soya meal (SM) and poultry manure (PM) obtained from a local feed mill around Alakia-Isebo road, Ibadan, Nigeria. The sawdust to which no supplement was added served as the control (CON). Water was added to the various substrates to a moisture content of about 65% with calcium carbonate (1%) added to adjust the pH of the substrates to 7.0. These were used to investigate the growth and yield of the mushrooms.

### **Vegetative growth**

The different substrate mixtures above were filled into test tubes of 25x150mm plugged with cotton wool covered with aluminium foil before sterilized in an autoclave at 121°C for 15 minutes. After cooling down to room temperature, the tubes were inoculated with freshly prepared and actively growing mycelium of the mushroom, the vertical mycelium extension was taken every other day for two weeks. Each treatment was replicated eight times.

### **Fruiting body cultivation**

The basal medium of *Gmelina aborea* sawdust was used. Portions of the sawdust were added with the following supplements at the rate of 10% w/w each of rice bran (RB), wheat bran (WB), palm kernel cake, (PKC), corn bran (CB), soya bean meal (SOY), groundnut cake GNC), poultry manure (PM) and the control (CON) had no supplement added. These substrate mixtures were moistened with water to about 65% moisture content and left overnight (16 hours) for the water to permeate the entire substrate. They were dispensed into polyethylene bags held in place with polyvinyl chloride (PVC) pipe which was thereafter covered with cotton wool wrapped in aluminium foil and steamed and steamed for 6 hours at 100°C.

After cooling down to room temperature, the bags were seeded singly with freshly prepared spawn of *P. pulmonarius*. The inoculated bags were incubated at 25-27°C for 30 days in a dark room followed by cropping.

Days to full colonization and appearance of mushroom primordial were taken. The fruiting bodies were harvested as they appeared and growth data such as number of fruits, pileus width and length of stipe, average fruit weight, total fruit weight, were collected and the biological efficiency (Ratio of total fresh mushroom harvested to substrate dry weight x 100) and production efficiency (ratio of total fresh mushroom harvested to substrate weight before cropping x 100) calculated.

### **Proximate analysis**

Proximate analysis was carried out immediately on the mushroom fruiting bodies harvested from the various treatments after weighing. Harvested fresh fruiting bodies of mushrooms were weighed immediately after harvest. The samples were dried at 80°C for 2 days, weighed, powdered in a Moulinex blender and sieved through a 450µm sieve. The final powdered samples were stored in desiccators until needed. The dried and powdered fruiting bodies of *P. pulmonarius* from the various treatments were analysed for food and mineral composition according to the method of Official Analytical Chemists, (1995). The analyses included the determination of crude protein, fat, moisture, ash and mineral contents. The mineral elements included sodium, potassium, calcium, phosphorus, magnesium, iron, copper, zinc and manganese and the values determined using Atomic Absorption spectrophotometer.

### **Results**

The performance of the mushroom on the various substrate additives was studied. It was observed in Table 1 below that *P. pulmonarius* recorded the highest number of fruiting bodies on SOYA supplemented substrate with the lowest observed on PM. Total fruiting body yield was found to be highest on Soya, CB and WB and lowest on PM.

**Table 1: Effects of different organic supplement inclusions on the growth and yield of *P. pulmonarius***

Treatment	Mycelia extension	Number of fruits	Total fruiting body weight (g)	Width of pileus (cm)	Length of stipe (cm)	Mean fruiting body weight (g)	Production efficiency (%)
CB	11.49 <sup>a</sup>	8.00 <sup>b</sup>	67.03 <sup>b</sup>	7.63 <sup>bc</sup>	7.63 <sup>a</sup>	8.46 <sup>a</sup>	20.60 <sup>c</sup>
PKC	10.41 <sup>b</sup>	9.86 <sup>a</sup>	61.07 <sup>d</sup>	8.50 <sup>a</sup>	7.66 <sup>a</sup>	6.24 <sup>c</sup>	22.43 <sup>a</sup>
PM	3.33 <sup>f</sup>	5.14 <sup>c</sup>	18.11 <sup>g</sup>	5.70 <sup>f</sup>	6.11 <sup>d</sup>	3.62 <sup>e</sup>	5.68 <sup>f</sup>
WB	8.69 <sup>c</sup>	9.43 <sup>a</sup>	66.07 <sup>c</sup>	7.36 <sup>cd</sup>	6.87 <sup>c</sup>	7.05 <sup>b</sup>	21.73 <sup>a</sup>
RB	10.66 <sup>b</sup>	7.57 <sup>b</sup>	47.04 <sup>e</sup>	7.53 <sup>bc</sup>	7.27 <sup>b</sup>	6.24 <sup>c</sup>	16.25 <sup>d</sup>
GNC	7.71 <sup>d</sup>	9.43 <sup>a</sup>	61.10 <sup>d</sup>	7.73 <sup>b</sup>	6.96 <sup>c</sup>	6.50 <sup>bc</sup>	20.87 <sup>bc</sup>
SOYA	7.60 <sup>d</sup>	10.14 <sup>a</sup>	68.73 <sup>a</sup>	7.17 <sup>d</sup>	6.36 <sup>d</sup>	6.82 <sup>b</sup>	22.60 <sup>ab</sup>
CONTROL	6.13 <sup>e</sup>	7.29 <sup>b</sup>	39.98 <sup>f</sup>	6.89 <sup>e</sup>	6.19 <sup>d</sup>	5.54 <sup>d</sup>	13.42 <sup>e</sup>

Means followed by the same superscript letter(s) in each column are not significantly different ( $P > 0.05$ ) according to DMRT.

CB (corn bran), PKC (palm kernel cake), PM (poultry manure), WB (wheat bran), RB (rice bran), GNC (groundnut cake), SOYA (soy meal), and the control.

**Table 2: Effects of different organic supplement inclusions on the proximate contents of *P. pulmonarius***

Treatments	N (%)	Protein (ppm)	Fat (ppm)	Moisture (ppm)	Ash (ppm)
CB	2.84 <sup>g</sup>	17.75 <sup>f</sup>	2.05 <sup>cd</sup>	8.67 <sup>b</sup>	7.92 <sup>c</sup>
PKC	4.40 <sup>a</sup>	27.52 <sup>a</sup>	3.42 <sup>b</sup>	5.60 <sup>e</sup>	7.54 <sup>d</sup>
PM	4.20 <sup>b</sup>	26.27 <sup>b</sup>	2.12 <sup>d</sup>	9.30 <sup>a</sup>	6.77 <sup>f</sup>
WB	3.53 <sup>f</sup>	22.09 <sup>e</sup>	2.29 <sup>c</sup>	7.92 <sup>c</sup>	8.03 <sup>b</sup>
RB	4.00 <sup>c</sup>	25.02 <sup>c</sup>	1.90 <sup>d</sup>	8.64 <sup>b</sup>	7.13 <sup>e</sup>
GNC	3.91 <sup>d</sup>	24.45 <sup>d</sup>	3.27 <sup>b</sup>	5.94 <sup>de</sup>	6.35 <sup>g</sup>
SOYA	3.67 <sup>e</sup>	22.97 <sup>e</sup>	4.01 <sup>a</sup>	6.28 <sup>d</sup>	7.12 <sup>e</sup>
CONTROL	2.23 <sup>h</sup>	15.92 <sup>h</sup>	1.60 <sup>e</sup>	6.12 <sup>d</sup>	8.67 <sup>a</sup>

Means followed by the same superscript letter(s) in each column are not significantly different ( $P > 0.05$ ) according to DMRT.

CB (corn bran), PKC (palm kernel cake), PM (poultry manure), WB (wheat bran), RB (rice bran), GNC (groundnut cake), SOYA (soy meal), and the control.

The widest pileus was observed on PKC while the narrowest was on PM. The biggest fruit size was seen on CB and PKC and the smallest was on PM. Production efficiency was highest with WB and PKC but lowest on PM with the longest mycelia extension on CB and the shortest on PM. Nitrogen and protein contents were significantly higher in all the supplemented substrates compared to the control at  $p < 0.05$ , the highest of which were recorded on fruiting bodies harvested from PKC supplemented substrate. Fat content was generally low on all the treatments with the least on the control. Ash content was significantly highest on the control and least on others.

Table 3 shows the macro-element content of fruit bodies harvested from the different supplemented substrates. Potassium was the most abundant of all the macro-elements evaluated. Sodium, magnesium, calcium and phosphorus were also present in appreciable amount.

**Table 3: Effects of different organic supplement inclusions on the major element contents of *P. pulmonarius***

Treatments	K (%)	Na (ppm)	Mg (%)	Ca (%)	P (ppm)
CB	1.15 <sup>f</sup>	81.77 <sup>b</sup>	0.19 <sup>c</sup>	0.12 <sup>e</sup>	0.86 <sup>e</sup>
PKC	1.36 <sup>d</sup>	70.29 <sup>e</sup>	0.20 <sup>b</sup>	0.17 <sup>a</sup>	1.01 <sup>c</sup>
PM	1.14 <sup>f</sup>	76.57 <sup>d</sup>	0.18 <sup>d</sup>	0.12 <sup>e</sup>	0.90 <sup>d</sup>
WB	1.45 <sup>b</sup>	57.50 <sup>h</sup>	0.18 <sup>d</sup>	0.15 <sup>c</sup>	1.04 <sup>b</sup>
RB	1.24 <sup>e</sup>	60.20 <sup>g</sup>	0.16 <sup>e</sup>	0.16 <sup>b</sup>	0.92 <sup>d</sup>
GNC	1.41 <sup>c</sup>	100.35 <sup>a</sup>	0.21 <sup>a</sup>	0.13 <sup>d</sup>	1.09 <sup>a</sup>
SOYA	1.26 <sup>e</sup>	78.34 <sup>c</sup>	0.18 <sup>d</sup>	0.12 <sup>e</sup>	0.67 <sup>f</sup>
CONTROL	1.55 <sup>a</sup>	66.92 <sup>f</sup>	0.16 <sup>e</sup>	0.11 <sup>f</sup>	0.49 <sup>g</sup>

Means followed by the same superscript letter(s) in each column are not significantly different ( $P>0.05$ ) according to DMRT. CB (corn bran), PKC (palm kernel cake,) PM (poultry manure),WB (wheat bran), RB (rice bran), GNC (ground nut cake), SOYA (soy meal), and the control.

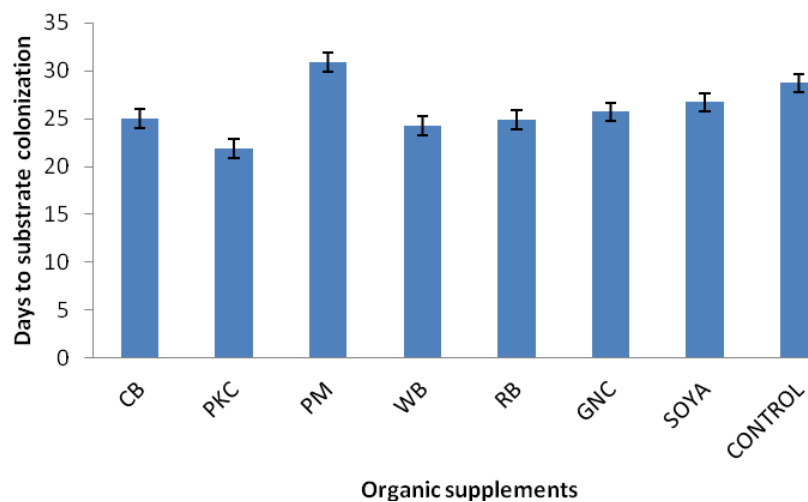
The results in Table 4 shows the values of micro-mineral elements in fruiting bodies from all the treatments. The fruiting bodies harvested from the control treatment recorded significantly highest amounts of all the micro-elements assayed with least occurring in the other treatments.

**Table 4: Effects of different organic supplement inclusions on the trace element contents of *P. pulmonarius***

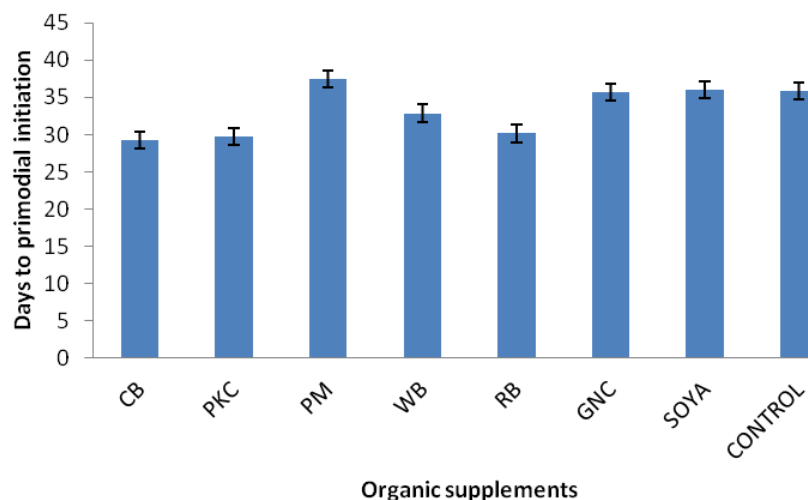
Treatments	Mn (ppm)	Zn (ppm)	Cu (ppm)	Fe (ppm)
CB	6.40 <sup>d</sup>	53.23 <sup>e</sup>	0.24 <sup>f</sup>	88.70 <sup>b</sup>
PKC	12.55 <sup>c</sup>	59.39 <sup>c</sup>	0.10 <sup>h</sup>	74.36 <sup>d</sup>
PM	5.28 <sup>e</sup>	61.67 <sup>b</sup>	0.28 <sup>e</sup>	64.80 <sup>f</sup>
WB	4.15 <sup>f</sup>	52.62 <sup>d</sup>	0.13 <sup>g</sup>	70.41 <sup>e</sup>
RB	3.43 <sup>g</sup>	59.27 <sup>c</sup>	1.07 <sup>b</sup>	59.14 <sup>g</sup>
GNC	13.79 <sup>b</sup>	58.95 <sup>c</sup>	0.39 <sup>d</sup>	83.56 <sup>c</sup>
SOYA	2.11 <sup>h</sup>	57.60 <sup>d</sup>	0.90 <sup>s</sup>	69.82 <sup>e</sup>
CONTROL	21.06 <sup>a</sup>	63.80 <sup>a</sup>	2.97 <sup>a</sup>	165.62 <sup>a</sup>

Means followed by the same superscript letter(s) in each column are not significantly different ( $P>0.05$ ) according to DMRT. CB (corn bran), PKC (palm kernel cake,) PM (poultry manure),WB (wheat bran), RB (rice bran), GNC (groundnut cake), SOYA (soy meal), and the control.

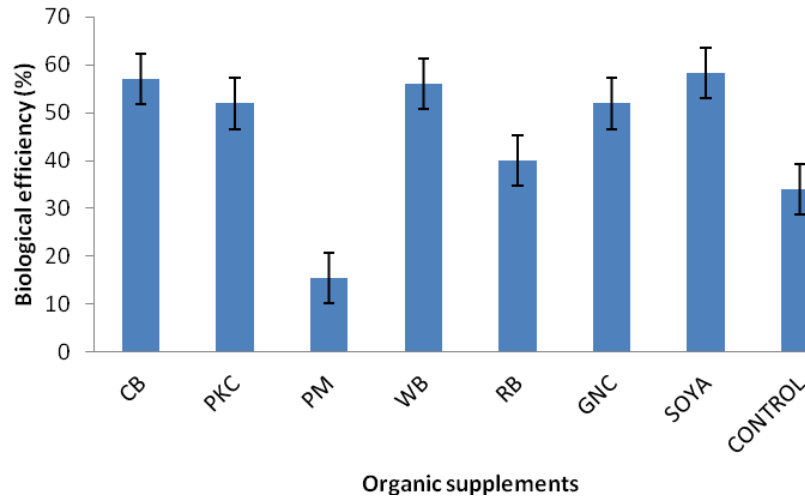
Days to substrate were shortest on PKC and longest on PM while days to primodial initiation (Days to first appearance of mushroom initials) were longest on PM and shortest on both PKC and CB (Figs 1 and 2). Highest biological efficiency was observed on soya supplemented substrates.



**Fig 1: Effects of organic supplements on number of days to substrate colonization of *P. pulmonarius*,**  
CB (corn bran), PKC (palm kernel cake), PM (poultry manure), WB (wheat bran), RB (rice bran), GNC (groundnut cake), SOYA (soy meal), and the control.



**Fig 2: Effects of organic supplements on number of days to appearance of *P. pulmonarius*,**  
CB (corn bran), PKC (palm kernel cake), PM (poultry manure), WB (wheat bran), RB (rice bran), GNC (groundnut cake), SOYA (soy meal), and the control.



**Fig 3: Effects of different organic supplements on the biological efficiency of *P. pulmonarius*,**  
**CB (corn bran), PKC (palm kernel cake,) PM (poultry manure),WB (wheat bran),**  
**RB (rice bran), GNC (ground nut cake), SOYA (soy meal), and the control.**

### Discussion

In this present study, the supplementation of the basal mushroom substrate (sawdust) with the various organic additives enhanced the biological efficiency (Percentage ratio of total fresh mushroom harvested to substrate dry weight), production efficiency (Percentage ratio of total fresh mushroom harvested to substrate weight before cropping) and mycelia growth of *P. pulmonarius* when compared with the control. Substrates for mushroom cultivation have been reported to have nitrogen content ranging between 0.5 and 0.8%, hence the addition of nitrogen sources to the substrates resulted in higher mushroom yield (Azizi *et al*, 1990; Gupta and Vijay, 1991). The addition of soya meal (SOYA) increased significantly the biological and production efficiencies and all the growth characters evaluated for *P. pulmonarius* which agreed with the findings of Naraian *et al* (2009) that during the cultivation of *P. florida* on maize cobs, the addition of soya meal as supplement increased the biological efficiency by 93%. Oseni *et al*,(2012) also reported Inclusion of these organic supplements especially PKC which reduced days to substrate colonization and primodial initiation which resulted in shortening the production circle of the mushroom and overall reduction in running cost of production (Hanai *et al*, 2005). Royse, (2004) earlier demonstrated that shortening of production period of *P. comucopiae* was achieved by inclusion of some additives into its basal substrate while Oseni *et al*,2012, obtained the same result with *P. ostreatus* when its substrate was supplemented with wheat bran.

The addition of the various organic supplements employed in this study also brought about a significant change in the nutritional quality and chemical composition of the mushroom fruiting bodies. This results agree with the findings of bank and Nandi, (2004) who reported a significant change in the nutrient composition of *P. sajor-caju* grown on spent rice straw compost supplemented with cotton seed hulls. The results of the nutritional analysis of the harvested fruiting bodies from the various treatments revealed higher content of protein and low fat contents in the mushroom fruiting bodies from the

supplemented substrate compared with the control which could be indicative of additional protein from the organic supplements added to the basal substrate. This result is in agreement with work of Jegadesh *et al*, (2010) who reported high protein and low fat contents in the fruiting bodies of *Volvariella bombycina*.

Macro and micro-elements are present in appreciable amounts, sufficient quantities of sodium in fruiting bodies from both supplemented and unsupplemented substrate is essential in the regulation of volume flow and blood pressure while potassium is essential in the synthesis of amino acids (Malik and Srivastava, 1982; Saïqa *et al*, 2008). Zinc and magnesium are required in enzyme synthesis (Chaney, 1982), calcium is important in bone formation while manganese and copper are required in protein synthesis (Ayaz *et al*, 2006)

### **Conclusion**

In conclusion, the results of this present study indicated that addition of organic supplements to the basal mushroom fruiting substrates will increase fruiting body yield, biological efficiency, production efficiency and nutritional quality of the mushroom. It can therefore be reasonably concluded that mushroom fruiting bodies from supplemented substrate hold tremendous promise in complementing protein and mineral needs of the teeming populace in the developing countries of which Nigeria is one.

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