# Evaluation of regenerating ability of mushroom (*Pleurotus tuber regium*) in Southern Nigeria

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## Abstract
*A screen house experiment was conducted at the Department of Crop Science, Akwa Ibom State University, ObioAkpa Campus to evaluate the effects of organic substrates (sawdust, poultry manure, horse dung and oil palm bunch refuse) and tuber regium weights (10g, 20g, 30g,40g and 50g) on performance of Pleurotus tuber regium. The experiment was a 4 x 5 factorial combination fitted into a completely randomized design (CRD) and replicated three times. Data collected were subjected to analysis of variance (ANOVA) at 5% probability level. The results of the study showed significant differences (p<0.05) on Pleurotus tuber regium yield and regenerating abilities. The oil palm bunch substrate had highest stripe height 4.90 cm, 6.58 cm cap diameter and highest mushroom yield of 339.45 grams per 2 kg substrate while sawdust substrate had the shortest stripe 3.09cm and poultry manure had the lowest cap diameter of 4.70cm and the least mushroom weight of 155.10g was obtained from poultry manure. The result indicated that oil palm bunch substrate resulted to 43-54% increases in mushroom yield while the least regenerating ability was observed from poultry manure substrate 50g tuber weight had 60-74% highest Pleurotus tuber regium yield as compared to others. The study therefore recommends oil palm bunch substrate with 50g tuber regium for high yield and regenerating ability of mushroom.*

**Keywords:** Regenerating ability, mushroom tuber regium, substrates

## Introduction
Mushrooms are increasingly becoming an important component of diets worldwide and are consumed for their medicinal values. Consumption of mushrooms is growing fast due to its balanced nutritional composition and will give benefits for health improvement because they are low in calories, sodium, fat, and cholesterol but rich in fiber, vitamins and minerals.

Mushroom cultivation is a profitable agricultural business. *Pleurotus tuber regium* is an edible mushroom having an excellent taste and flavour. It's usually consumed as soup, grilled or deep fried and it is almost having a great demand as local street food (Akpan, 2015). Mushroom is used in cooking many local cuisines and dishes. *Pleurotus tuber regium*, sclertia is used as soup thickener, raising agent and for fresh mushroom production and research purposes (Akpan, 2009). *Pleurotus* fungi are found both in the tropical and temperate climates throughout the world (Aman, 2002) and *Pleurotus tuber regium* produces underground sclerotia (Croan, 2004). Mushroom cultivation requires knowledge of the natural substrates of fungi. Though mushrooms are usually grown on beds of composite log, in bags, wood chip and sawdust but *Pleurotous tuber regium* can be grown on various agricultural wastes with the use of different technologies. *Pleurotus tuber regium* can be cultivated on a wide range of substrates. The substrates on which mushroom spawn is grown affects the mushroom production. Bhuyan in his Bhattacharyya DK *etal* (2015) opined that remarkable variations have been observed in the nutritional contents of Oyster mushroom grown on different substrates. This research work was limited to substrates such as decomposed sawdust (DS), battery cage poultry manure (BCPM), decomposed palm bunch (DPB) and horse dung (HD) and soil only. It is important to choose appropriate substrates in a given place to grow them for all year availability. The objectives of this study were to determine the regenerating ability of different sizes of tuber regium and assess its performance on different cultivated organic manure.

**Materials and methods**

The experiment was carried out in the screen house of the Department of Crop Science, Akwa Ibom State University, Obio-Akpa Campus. Obio-Akpa is located at latitude $04^0 30^0$ and $05^0 30^0$N and longitude $7^0 30^0$ and $08^0 30^0$E (Slus-Ak, 1989). ObioAkpa is located in the humid tropical rainforest zone of South Eastern Nigeria and has an annual mean rainfall of 2500 mm and monthly sunshine of 3.14hrs with a mean annual temperature of 200C. The relative humidity of the study area is 70% and evaporation rate of 2.6cm2 (Peters *et al,* 1989). The rainfall in ObioAkpa is bimodal. Rainfall usually starts in March and end in November with a short period of relative moisture stress in August, traditionally referred to as 'August Break' (Peters *et al*,; 1989). The temperature of the study area is generally high in the months of February through April (Enwezer *et al.,* 1990).

Soil rich in humus was obtained from the oil palm plantation in Akwa Ibom State University Teaching and Research Farm, Faculty of Agriculture, ObioAkpa Campus. The soil was air dried, crushed and sieved through 2.0mm mesh, for physico-chemical analysis.The experiment was a 4 x 5 factorial combinations laid out in a completely randomized design (CRD) and replicated three times. Factor A treatments were four organic substrates (poultry manure, horse dung, sawdust and oil palm bunch refuse) while five *tuber regium* weights (10, 20, 30, 40 and 50g) constitute the factor B treatment.

The poultry manure was obtained from ENI livestock farm, Uyo, Akwa Ibom State. The system of the livestock management was battery cage. The poultry manure was cured, sun dried and sieved into fine particles. Horse dung was obtained from the Polo club at Port Harcourt, Rivers State, Nigeria. The Horse dung was sun dried to reduce moist content. The lumps were broken and then sieved into fine particles. Decomposed sawdust was obtained from Uyo Timber Market at IfaAtai, Uyo Local Government Area. The sawdust sample was air dried for one week and sieved into fine particles. Decomposed palm bunch was obtained from the agro processing factory in the AkwaIbom State University, ObioAkpa Campus. All the organic substrates were sun dried and sieved into fine particles before analysis in the University Laboratory.

All the substrates were mixed with soil particles in the ratio of 3:1 (3/4 part of substrate and ¼ part of soil) and put in perforated polythene bags of 25 cm high and 48cm circumference. The substrates and soil mixture on treatment basis were filled at 75% field capacity. The *tuber regium* was planted into the substrate on treatment basis at depth of 10cm.The *tuber regium* was obtained from Urua Obo (Market) in EssienUdim Local Government Area of AkwaIbom State. The *tuber regium* were cut into five different sizes of varying weights based on the treatment basis (10g, 20g, 30g, 40g and 50g). The weights were determined with aid of sensitive electronic weighing balance (Meter Toledo, Made in Japan).

All the data collected were subjected to analysis of variance, using Gene stat discovering, 2012 version model. Significant means were compared with least significant difference (LSD) at 5% probability level.

## Results and discussion

*Results*

*Soil physico-chemical properties of the soil used for mushroom production*

The particle size analysis indicated that soil was sandy loam with 89.86% sand, 1.86% silt and 8.28% clay. The pH value was 6.42 The organic matter content was high (3.03%) while total nitrogen was low (0.11%). The available phosphorus content was also high (21.31mg/kg). the exchangeable bases such as calcium were high (3.10cmolkg) while K was moderate (Table 1). The exchange acidity, effective cation exchange capacity and base saturation were 2.41 cmol/kg, 7.75 cmol/kg and 69.03%, respectively. The result of the soil physico-chemical properties was within the recommended range for cultivation of mushroom as given by Chang and Miles (2004).

*Chemical properties of the organic manures used for mushroom cultivation*

The pH values of all the manure were above 7 indicating alkalinity. Poultry manure had the highest pH value of 9.40 while sawdust had the least (6.60). The organic carbon (%) contents in all the manures were high as it ranged between 32.29 % in poultry manure and 51.62 % in sawdust. Poultry manure had the highest total nitrogen value of 3.04%, followed by 2.50% from Horse dung while the least (0.52%) was from sawdust. Oil palm bunch refuse had the highest C/N ratio of 125.16, followed by sawdust 99.27 and the least, 10.56 was from poultry manure. The available phosphorus and exchangeable bases were also high (Table 2) based on Chang and Miles (2004) rating. Number of Mushroom Shoots per Substrates and Tuber regium weight. The number of shoots as influenced by substrates were significantly different ($p<0.05$) at 1, 3, 4 and5 weeks harvesting intervals (WHI). The oil palm bunch substrates had the highest number of shoots per substrate; 4.20, 5.70, 4.18, 2.33 and 2.02 at 1, 2, 3, 4 and 5 (WHU or WHI), respectively. Poultry manure substrates had the least number of mushroom shoots at 3, 4, and 5 WHI (Table 3). The result showed decrease in number of mushroom shoots at 5 WHI (Table 4). Poultry manure substrates had no mushroom shoot at 5WHI while oil palm bunch, sawdust and Horse dung had 2.02, 1.01 and 0.30, respectively at WHI. The result showed that oil palm bunch had the highest regenerating ability, followed by sawdust and Horse dung while poultry manure had the least. The effects of *tuber regium* weight on number of shoots were significantly different in all the harvesting weeks (Table 1.3). The result showed increase in number of shoots with increase in tuber regium weights, the 50 g weight had significantly higher number of shoots; 5.50, 6.21, 6.03, 2.12 and 0.98 at 1, 2, 3, 4, and 5 WHI with 20.84 total number of mushroom. The 40g *tuber regium* weight produced 5.30, 6.10, 5.90, 2.10 and 0.19 shoots, respectively with 20.31 total numbers of shoots. Generally, the least number of shoots was from the 10 g *tuber regium* weight. The result showed that increase in *tuber regium* weight increased regenerating ability. The 50g and 40g *tuber regium* weights had regenerating ability up to 5 weeks while 30, 20 and 10g weights stopped at 4 weeks. The interaction effect between substrates and tuber weights on number of shoots at all the harvesting periods were not statistically significant ($p<0.05$). Yield components of mushroom as affected by substrates and *tuber regium* weights. Stripe heights (cm). Stripe height as influenced by substrate of decomposing oil palm bunch varied significantly ($p<0.05$) (Table 4). The substrate of decomposing oil palm bunch had the longest stripe, 4.90 cm, followed by 4.50cm recorded from mushroom grown in horse dung. The shortest stripe; 3.04 was from poultry manure.

Stripe height as influenced by tuber regium was not significantly ($p>0.05$) different (Table 4). The stripe height ranged from 3.68cm in 10g weight to 4.30cm at 50g weight. The interaction between tuber regium weights and stripe height was not significantly different. Stripe Girth. Stripe girth as influenced by substrates and tuber regium weights were not significantly ($p>0.05$) different (Table 4). Decomposing oil palm bunch substrate had 4.92cm stripe girth, followed by 4.80 and 3.89cm from Horse dung and poultry manure, respectively, the smallest stripe girth; 3.77cm was from sawdust.Among, the tuber regium weights, the result showed no significant difference ($p>0.05$) . The interaction between substrates and tuber regium weights on stripe girth was not significantly ($p>0.05$) different (Table 4).Cap DiameterMushroom cap diameter as influenced by substrates and tuber regium weights indicated significant differences ($P < 0.05$) as presented in (Table 4). The decomposing oil palm bunch substrate had the highest cap diameter of 6.38cm which was significantly higher than those obtained from other substrates. The smallest cap diameter of 4.70 cm was recorded in poultry manure substrate.

Cap diameter as influenced by *tuber regium* showed significant differences ($P <0.05$) (Table 4.4). The result showed increase in *tuber regium* weight with increase in cap diameter. The 50g *tuber regium* had cap diameter of 6.32cm followed by 6.30cm from 40g weight. The smallest cap diameter; 4.81cm was from 10g weight. The interaction between substrates and *tuber regium* weights on cap diameter was not significantly different.

Mushroom yield and regenerating ability .
Mushroom yield as affected by different substrates varied significantly ($P <0.05$) at 1, 2, 3, 4 and 5 weeks of harvesting interval (Table 5). In all the substrates, the highest mushroom yield was observed at 2 weeks harvesting. The oil palm bunch had the highest mushroom yield; 50.18, 100.20, 81.25, 59.22 and 48.60g at 1, 2, 3, 4 and 5 weeks harvesting, respectively. The decomposing oil palm bunch substrate had a total of 339. 45g mushroom yield. This was followed by 24.80g, 86.81g from Horse dung at 1 and 2 weeks harvesting, seconded by Sawdust, 38.66 and 30.11g at 3 and 4 weeks harvesting. The decomposing oil palm bunch substrate had the highest total mushroom yield; 339.45, followed by 193.45g and 167.57g from Horse dung and

Sawdust, respectively. The least total mushroom yield; 155.10g was from poultry manure. The result showed that decomposing oil palm bunch substrate had the highest regenerating ability, followed by horse dung and sawdust. The least was from poultry manure substrate.

The effect of tuber regium weight on mushroom yield and regenerating ability is shown on Table 5. The result indicated increase in tuber regium weight with increase in mushroom yield and regenerating ability. In all the *tuber regium* weights, the highest mushroom yield was observed at 2 weeks harvesting. The 50g *tuber regium* weight had the highest regenerating ability of 480.65 g per 2 kg substrate weight, followed by the 40 g weight and then 30 g while the least was from the 10 g weight. The integration between substrates and *tuber regium* weights on mushroom yield and regenerating ability were not significantly different (P > 0.05) (Table 5).

**Discussion**

The result of the study showed that mushroom (*Pleurotus tuber-gium*) could be grown in poultry manure, horse dung, sawdust and oil palm bunch substrates. The ability of mushroom to grow in these substrates could be that the substrates were rich in chemical nutrients and also favourable for mycelia growth. This observation agrees with the findings of Chang and Miles (2009) that oyster mushroom could be cultivated on a wide range of organic substrates such as sawdust, paddy straw, bagasses, cornstalk, waste cotton stalk and banana leaves. Udo and Akpan (2012) reported that mushroom could be grown on locally available organic manures and crop residues. Despite the nutrients content of the substrates,the environment where the mushroom was cultivated could be a major factor enhancing mycelia growth. The temperature, moisture level, humidity and solar radiation were controlled in screen house (Akpan, 2015). The temperature range of the screen house was between $25^0$C to $30^0$C during the growing period, this temperature range could have enhanced mycelia growth and development of mushroom. This observation agrees with the report of Zadrazil (1978), Levy (1982), Zadrazi and Kurtzman (1982) who reported that; oyster mushroom is best strain for cultivation of mushroom in the tropical and subtropical areas where the environmental temperature is high.The result indicated significant increase in mushroom yield and regenerating ability from decomposing oil palm bunch substrate while low yield and regenerating ability was observed from poultry manure substrate. The high yield and regenerating ability observed from oil palm bunch could be due to moderate pH range (7.40), and significant higher C/N ratio (125.16) compared to 10.56 observed from poultry manure. This observation agrees with the report of Ridley (1996) that the best pH rang for growing mushroom is between 6.60 – 8.50. Peterson and Ridley (1996) also reported that C/N promotes mycelia growth and development. This also showed a significant increase in mushroom yield with increase in *tuber regium* weight. The 50g produced the highest mushroom while the least was from 10g. The following parameters were obtained from mushroom, stripe height, stripe diameter, cap diameter, number of mushroom shoots and mushroom yield. Data collected were subjected to analysis of variance. Significant difference (P<0.05) in all the parameters evaluated, among the substrates, decomposing oil palm bunch substrates was superior in both mushroom yield and regeneration ability. The least was observed from poultry manure.

The oil palm bunch substrate had 43-45% mushroom yield compared to the other substrates. The results of the study also indicated increase in *tuber regium* weight with increase in mushroom yield and regenerating ability. The 50g *tuber regium* weight had 6-74% greater mushroom yield when compared to the other *tuber regium* weights.

**Conclusion and recommendation**

Based on the study findings, it was concluded that;

- All the organic substrates used enhanced mushroom growth and the decomposing oil palm bunch substrate performed best in both mushroom yield and regenerating ability while the least was from poultry manure.
- Increase in *tuber regium* weight, resulted to significant increase in mushroom yield and regenerating ability.
- The 50g *tuber regium* had the highest mushroom yield and regenerating ability.

It is therefore recommended that:

- Oyster mushroom farmers should use decomposing oil palm bunch as substrates for high yield of mushroom and regenerating ability.
- Farmers should use 50g of *tuber regium* weight for high yield of mushroom and longer regenerating ability.
- *Pleurotus tuber regium* could be grown all year round using available and sustainable substrates such as sawdust, poultry dropping and decomposing palm lunch.
- All the organic substrates used are organic waste and in abundance.
- Farmers should avail themselves of the opportunity to transform waste to wealth.

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**Table 1: Physico chemical properties of the soil used for mushroom production**

| Soil property | Value |
|---|---|
| Sand (%) | 89.86 |
| Silt (%) | 1.86 |
| Clay (%) | 8.28 |
| Textural Class | **LS** |
| Soil pH ($H_2O$) | 6.42 |
| EC (ds/m) | 0.132 |
| Organic Matter (%) | 3.03 |
| Total N (%) | 0.11 |
| Avail. Phosphorus (mg/kg) | 21.31 |
| Exchangeable bases: | |
| Calcium (cmol/kg) | 3.10 |
| Magnesium (cmol/kg | 1.80 |
| Sodium (cmol/kg) | 0.14 |
| Potassium (cmol/kg) | 0.31 |
| Exchange acidity (cmol/kg) | 2.41 |
| ECEC (cmol/kg) | 7.75 |
| Base Saturation (%) | 69.03 |

**Table 2: Chemical properties of the organic manures used for mushroom cultivation**

| Organic manure | pH | EC µs/M | Organic Carbon (%) | Total N (%) | C/N ratio | Ca | Mg | K | Na | P |
|---|---|---|---|---|---|---|---|---|---|---|
| Sawdust | 6.60 | 310.0 | 51.52 | 0.52 | 99.27 | 3310.0 | 2041.00 | 3100 | 886.60 | 197.25 |
| Battery Cage | 9.40 | 2590.0 | 32.09 | 3.04 | 10.56 | 144400.00 | 2880.0 | 4686 | 2484 | 749.55 |
| Horse manure | 8.10 | 3090.0 | 32.79 | 2.50 | 13.12 | 3360.00 | 800.00 | 6652 | 2500.20 | 789.00 |
| Oil Palm bunch | 7.40 | 2112.00 | 40.00 | 0.20 | 125.16 | 1440.0 | 10230.0 | 1398 | 688.80 | 157.80 |

Fig. 2: Suitability map of the study area for yam cultivation

**Table 3: Number of mushroom shoots per substrate and *tuber regium* weights**

| | Harvesting Intervals | | | | | |
|---|---|---|---|---|---|---|
| Substrates | 1 | 2 | 3 | 4 | 5 | Total |
| Poultry manure | 3.13 | 4.78 | 2.11 | 1.04 | 0.0 | 11.06 |
| Horse dung | 3.18 | 5.66 | 3.40 | 1.28 | 0.30 | 13.82 |
| Sawdust | 2.01 | 4.10 | 3.20 | 1.35 | 1.01 | 11.67 |
| Oil Palm bunch | 4.20 | 5.70 | 4.18 | 2.33 | 2.02 | 18.43 |
| LSD (p<0.05) | 1.15 | NS | 1.32 | 0.59 | 0.91 | 2.38 |
| ***tuber regium* Weight (g)** | | | | | | |
| 10 | 2.55 | 3.91 | 2.01 | 0.81 | 0.00 | 9.27 |
| 20 | 3.47 | 4.33 | 3.47 | 1.28 | 0.00 | 12.55 |
| 30 | 3.88 | 5.01 | 3.99 | 1.70 | 1.10 | 15.68 |
| 40 | 5.30 | 6.10 | 5.90 | 2.10 | 0.91 | 20.31 |
| 50 | 5.50 | 6.21 | 6.03 | 2.12 | 0.98 | 20.84 |
| LSD (P<0.05) | 1.82 | 1.40 | 2.10 | 1.83 | 0.09 | 3.49 |
| Interaction | NS | NS | NS | NS | NS | NS |

**Table 4: Yield component of mushroom as affected by substrate and *Tuber regium* weight**

| Substrates | Stripe (cm) | Heights | Stripe (cm) | Girth | Cap (cm) | Diameter |
|---|---|---|---|---|---|---|
| Poultry manure | 3.04 | | 3.89 | | 4.70 | |
| Horse dung | 4.50 | | 4.80 | | 5.58 | |
| Sawdust | 3.09 | | 3.77 | | 5.30 | |
| Oil Palm bunch | 4.90 | | 4.90 | | 6.58 | |
| LSD (P<0.05) | 0.48 | | NS | | 0.75 | |
| Tuber regium weight (g) | | | | | | |
| 10 | 3.68 | | 4.30 | | 4.81 | |
| 20 | 4.18 | | 4.33 | | 5.60 | |
| 30 | 4.25 | | 4.35 | | 5.81 | |
| 40 | 4.26 | | 4.37 | | 6.30 | |
| 50 | 4.30 | | 4.38 | | 6.32 | |
| LSD (P<0.05) | NS | | NS | | 1.03 | |

**NS = Not Significant**

**Table 5: Mushroom yield as influenced by substrates and *tuber regium* weight**

| | Harvesting Interval (WHI) | | | | | |
|---|---|---|---|---|---|---|
| **Substrates** | **1** | **2** | **3** | **4** | **5** | **Total weight (g)/ 2kg substrates** |
| Poultry manure | 23.90 | 80.90 | 29.90 | 20.40 | 0.00 | 155.10 |
| Horse dung | 24.80 | 86.81 | 32.40 | 28.70 | 20.74 | 193.45 |
| Sawdust | 23.70 | 61.80 | 38.66 | 30.11 | 13.30 | 167.57 |
| Oil Palm bunch | 50.18 | 100.20 | 81.25 | 59.22 | 48.60 | 339.45 |
| LSD (P<0.05) | 3.97 | 4.88 | 6.02 | 3.40 | 2.33 | 8.41 |
| **Tuber regium weight (g)** | | | | | | |
| 10 | 20.18 | 51.30 | 33.40 | 19.20 | 0.00 | 124.08 |
| 20 | 51.33 | 69.70 | 58.90 | 20.12 | 0.00 | 200.05 |
| 30 | 97.81 | 129.33 | 106.30 | 40.72 | 10.66 | 384.82 |
| 40 | 101.25 | 131.25 | 114.70 | 81.40 | 25.40 | 454.00 |
| 50 | 108.89 | 139.30 | 120.40 | 83.25 | 28.81 | 480.65 |
| LSD (P<0.05) | 5.50 | 7.67 | 11.12 | 6.12 | 2.12 | 9.23 |
| Interaction | NS | NS | NS | NS | NS | NS |

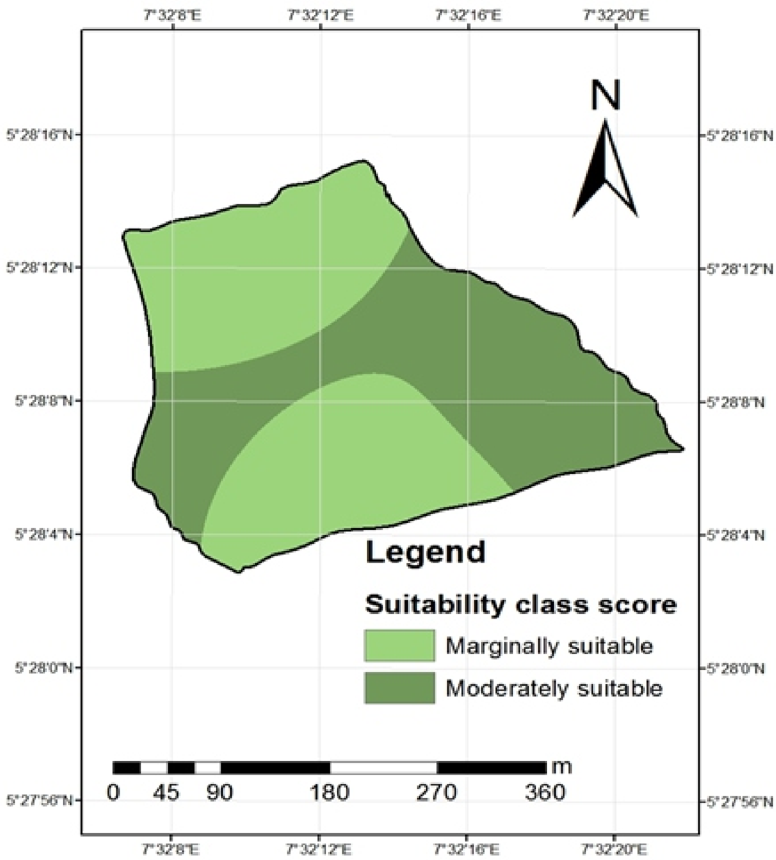


Fig. 2: Suitability map of the study area for yam cultivation