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EVALUATION OF ORGANIC SUBSTRATES FOR OUTDOOR CULTIVATION OF PLEUROTUS TUBER-REGIUM

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ABSTRACT

Pleurotus tuber-regium is a unique mushroom with a lot of benefits. Yield of the mushroom was evaluated when cultivated outdoor on agricultural beds different substrates (treatments) in order to assess the effect of the substrates. Four substrates used were topsoil, a mixture of topsoil and sawdust (2kg), a mixture of topsoil and poultry droppings (2kg), and a mixture of topsoil, poultry droppings (2kg) and sawdust (2kg). The design was Randomized Complete Block Design (RCBD) with four replicates. Results showed that mixture of topsoil and sawdust had the highest mean dry weight 3.54 ± 0.29 for first flush while the least was 2.57 ± 0.45gm for topsoil. For the second flush, the highest mean dry weight of 1.56 ± 0.37 was for mixture of topsoil and sawdust while the least, 0.58 ± 0.31gm was for topsoil, poultry droppings and sawdust. Results for dry weight of fruiting bodies, indicated that only the mixture of Topsoil & Poultry droppings and Topsoil, Poultry droppings & Sawdust had no significant difference ($P>0.05$), for first flush; there was significant difference between other treatments. Results for second flush had no significant difference ($P>0.05$) between Topsoil and Topsoil & Sawdust treatments; there was significant difference ($P<0.05$), between other treatments. There was significant difference ($P<0.05$) in the stipe girth, pileus diameter and wet weight for all the treatments for both flushes. Significant difference between treatments varied for stipe height. The mixture of topsoil and sawdust substrate is recommended for the outdoor cultivation of *Pleurotus tuber-regium*. These findings are discussed.

Contribution/Originality: This study contributes to the existing literature of *Pleurotus tuber-regium* and investigates its yield outdoor. This study is one of very few studies which have investigated outdoor cultivation of the mushroom. Its primary contribution is that *Pleurotus tuber-regium* could be cultivated outdoor on agricultural beds, using mixture of topsoil and some organic substrates.

1. INTRODUCTION

Mushroom is the fleshy, spore-bearing fruiting body of a fungus (macrofungus) typically produced above the ground on soil or on its food source. They are placed in a kingdom of their own apart from plants and animals, with a distinctive fruiting body which can either be epigeous (growing on or close to the ground) or hypogeous (growing under the ground) Chang (1991). The macrofungi have fruiting bodies large enough to be seen with the naked eye and to be picked up by hand. Ideally, the word mushroom refers only to the fruit body. Generally, mushroom has a stem (stipe), a cap (pileus), and gills (lamellae) on the underside of the cap. According to Boa (2004) "mushroom" can also be referred to as a wide variety of fungi, with or without stems and the terms used even more generally to describe both the fleshy fruiting bodies of some Ascomycota and the woody or leathery fruiting bodies of some Basidiomycota, depending upon the context of the word.

Pleurotus tuber-regium (Fr.) Sing. is a Tuberous wild species of white rot basidiomycete which produces fruit bodies from a unique globose sclerotium that is more like a giant truffle (Nwokolo, 1987). It is tropical and subtropical in distribution and found growing on many species of hard and soft woods like *Mangifera indica, Daniellia oliveri and Treculia africana. Pleurotus tuber-regium* is the only know *pleurotus* species which produces true sclerotium and also differs from all other *pleurotus* species in its non-pleurotoid habit (Isikhuemhen and Nerud, 1999). The sclerotia are usually of various sizes, ranging from a few centimeters to several centimeters in diameter. They are spherical to oval in shape, dark brown on the outside and whitish on the inside (Okhuoya and Okogbo, 1991). Several mushrooms are especially tasty and many are rich in nutrients. Many species are high in fiber and provide vitamins such as thiamine, cobalamin, niacin, riboflavin, ascorbic acid and biotin. Chang (1991) confirmed that mushrooms are also a source of some minerals, including iron, selenium, potassium and phosphorus. This important mushroom is often cultivated indoor mainly for research purposes. Indoor cultivation of mushrooms has deterred some farmers who showed interest in production. This study investigated the outdoor cultivation of *Pleurotus tuber-regium* in order to encourage its cultivation by farmers.

2. MATERIALS AND METHODS

2.1. Sources of Materials

The sclerotia used in this study were obtained from the Mushroom Farm, University of Port Harcourt Teaching and Demonstration Farm. The fresh sawdust was sourced from the timber shed at Rumuosi in Port Harcourt, Rivers State, Nigeria while the poultry droppings were from the University of Port Harcourt Teaching and Demonstration Farm Choba Campus.

2.2. Collection of Species

Pleurotus tuber-regium was selected for this study because it is particularly common in the South East of Nigeria, where both the sclerotia and the mushroom are commonly used in soup preparation (Akpaja *et al*., 2003). The four substrates selected were; topsoil (control), a mixture of topsoil and sawdust, a mixture of topsoil and poultry dropping and a mixture of topsoil, sawdust and poultry droppings. They are selected because they are readily and locally available.

2.3. Preparation of the Substrates for Cultivation

The four substrates were prepared to ascertain the one that will be good enough in the outdoor production of *Pleurotus tuber-regium* in terms of qualities and quantities of the fruiting bodies. A 11m × 15m plot of land was mapped out and cleared properly. A bed of 2m × 2m was prepared and a furrow of 1m was made. The topsoil and sawdust(2kg) was mixed properly, the topsoil and poultry droppings(2kg) was also mixed, as well was the topsoil, sawdust and poultry droppings(2kg). The sclerotium was soaked in water for 8 hours and then planted in each bed containing the substrate. The sclerotium was planted with a spacing of 50cm and 1 seed was planted per hill which resulted in 4 sclerotia per bed.

2.4. Experimental Design

Experimental design was Randomized Complete Block Design (RCBD) with four treatments replicated four times.

2.5. Data Collection

Data were recorded from the different replicates and the mean of each set of data was calculated. The experiment was designed to determine the best substrate for outdoor cultivation of *Pleurotus tuber-regium*. The data collected were:

- Height of stipe: This was measured in centimeters using meter rule from the base to the stipe of the pileus.
- Stipe girth: This was measured using a rope. Using a meter rule, the actual girth was determined.
- Diameter of pileus: This was measured in centimeters using meter rule from one edge of the pileus to the other edge.
- Fresh Weight: The fruitbodies were weighed immediately after harvest using electronic balance.
- Dry Weight: After recording the fresh weight, the fruit bodies were than dried and weighed.

2.6. Statistical Analysis

The data obtained were statistically analysed using Statistical Package for the Social Sciences (SPSS) and significant means were separated using LSD @ 0.05 level of significance.

3. RESULTS

3.1. Performance of Pleurotus Tuber-regium (Stipe Height) on Different Treatments

The results of stipe height *of Pleurotus tuber-regium* on the different treatments showed that there was no significant difference ($P>0.05$) for mushrooms produced on mixture of Topsoil & Sawdust and mixture of Topsoil & Poultry droppings for the first flush whereas, there was significant difference ($P<0.05$) between other treatments. For the second flush, there was significant difference ($P<0.05$) in the stipe height between all the treatments Table 1. Topsoil (control) had the highest mean stipe height (5.36 cm ±0.36), while the least (4.19 cm ±0.67) was for the mixture of topsoil and poultry dropping. For the second flush, the mixture of topsoil and sawdust substrate had the highest mean stipe height (2.53 cm ±0.64), while the least was for the mixture of topsoil, poultry droppings and sawdust (1.00 cm ±0.54).

Table-1. Performance of *Pleurotus tuber-regium* (stipe height) on different treatments.

Substrate	First Flush (cm)	Second Flush (cm)
Topsoil (control)	5.36±0.36	2.31±0.63
Topsoil/Sawdust	4.23±0.51	2.53±0.64
Topsoil/Poultry droppings	4.19±0.67	1.51±0.59
Topsoil/Poultry droppings/Sawdust	5.03±0.44	1.00±0.54
LSD (0.05)	0.148	0.103

3.2. Performance of Pleurotus tuber-regium (Stipe Girth) on Different Treatments

There was significant difference ($P<0.05$) in the stipe girth between all the treatments for both flushes. In the first flush, the fruit bodies with the widest mean stipe girth (5.17 cm ±0.17) were those grown on topsoil (control), while the least (3.91 cm ±0.60) were from those grown in the mixture of topsoil and poultry droppings substrate. In the second flush topsoil (control) had the widest mean stipe girth (2.64 cm ±0.61), while the least (0.89 cm ±0.49) were from those grown in the mixture of topsoil, poultry droppings and sawdust substrate Table 2.

Table-2. Performance of *Pleurotus tuber-regium* (stipe girth) on different treatments.

Substrate	First Flush (cm)	Second Flush (cm)
Topsoil (control)	5.17±0.17	2.64±0.61
Topsoil & Sawdust	4.09±0.42	2.46±0.63
Topsoil & Poultry droppings	3.91±0.60	1.33±0.52
Topsoil & Poultry droppings & Sawdust	5.09±0.39	0.89±0.49
LSD (0.05)	0.060	0.050

3.3. Performance of Pleurotus Tuber-regium (Pileus Diameter) on Different Treatments

There was significant difference ($P<0.05$) in the pileus diameter between the treatments for the two flushes. The widest mean pileus diameter was in the mixture of topsoil, poultry droppings & sawdust treatment (8.55 cm ±0.83), while the mixture of topsoil and sawdust had the least (4.76 cm ±0.89). For the second flush, the mixture of topsoil and sawdust had the widest mean pileus diameter (3.08 cm ±0.88), while the least was from the mixture of topsoil and poultry droppings substrate (1.44 cm±0.79).

Table-3. Performance of *Pleurotus tuber-regium* (pileus diameter) on different treatments.

Substrate	First Flush (cm)	Second Flush (cm)
Topsoil (control)	7.49±0.94	2.69±0.85
Topsoil & Sawdust	4.76±0.89	3.08±0.88
Topsoil & Poultry droppings	5.95±1.09	1.92±0.86
Topsoil & Poultry droppings & Sawdust	8.55±0.83	1.44±0.79
LSD (0.05)	0.069	0.218

3.4. Performance of Pleurotus tuber-regium (Fresh and Dry Weight) on Different Treatments

3.4.1. Fresh Weight

There was significant difference ($P<0.05$) in wet weight for all treatments in first and second flushes. In the first flush, the highest mean fresh weight (24.29g±1.55) was from the mixture of topsoil and sawdust treatment and the least (17.48 g ±2.95) was from topsoil (control). In the second flush, the mixture of topsoil and sawdust had the highest mean fresh weight (10.41 g ±2.72), while the least (4.29 g ±2.36) was from the mixture of topsoil, poultry droppings & sawdust treatment Table 4.

3.4.2. Dry Weight

Results for dry weight showed that only the mixture of Topsoil & Poultry droppings and Topsoil, Poultry droppings & Sawdust had no significant difference ($P>0.05$), for first flush; there was significant difference between other treatments. Results for second flush had no significant difference ($P>0.05$) between Topsoil (control) and Topsoil & Sawdust treatments; there was significant difference ($P<0.05$), between other treatments. In the first flush, the highest mean dry weight (3.54g ±0.29) was from the mixture of topsoil and sawdust substrate while the least (2.57g ±0.45) was from topsoil. Topsoil and Sawdust and mixture of Topsoil, Poultry droppings & Sawdust had (1.56g±0.37), highest dry weight and (0.58g±0.31), least dry weight respectively Table 4.

Table-4. Performance of different substrate on *Pleurotus tuber-regium* fresh weight and dry weight.

Weight	Substrate	First Flush (g)	Second Flush (g)
Wet	Topsoil (control)	17.48±2.95	9.04±2.55
	Topsoil & Sawdust	24.29±1.55	10.41±2.72
	Topsoil & Poultry droppings	20.42±2.53	6.03±2.55
	Topsoil & Poultry droppings & Sawdust	19.78±2.53	4.29±2.36
	LSD (P≤0.05)	0.076	0.127
Dry	Topsoil (control)	2.57±0.45	1.41±0.35
	Topsoil & Sawdust	3.54±0.29	1.56±0.37
	Topsoil & Poultry droppings	2.96±0.44	0.91±0.36
	Topsoil & Poultry droppings & Sawdust	2.95±0.47	0.58±0.31
	LSD (0.05)	0.137	0.181

4. DISCUSSION

The results obtained in the experiment reveal that the mixture of topsoil and sawdust produced mushroom with highest fresh and dry weights. This result could probably be due to the topsoil providing high concentration of organic matter and microorganisms for the germination and fructification of the mushrooms and sawdust being a lingo-cellulosic agricultural waste. The combination of the qualities of topsoil and sawdust might have been responsible for the highest yield recorded in this treatment. Sawdust has been reported as the best substrate for mycelial growth and fructification (Kadiri and Fasidi, 1990). Furthermore, researchers have reported that sclerotia of *Pleurotus tuber-regium* can be grown outdoor in lignocellulosic agricultural wastes as substrates, which is much faster, economical and easier than growing it from the spawn raised from the spores (Isikhuemhen and LeBauer, 2004; Olufokunbi and Chiejina, 2010). Topsoil treatment produced good stipe height and stipe girth yield of the mushroom. Okhuoya and Etugo (1993) reported loam soil as the best for planting sclerotia, and that might be due to high water holding capacity. Soil supplies water for the growth of the mycelium and fruitbodies, it buffers weather conditions in the environment, protects the mycelium from drying and provides an environment suitable for the stimulation of fruit bodies and their development (Oei, 1996; 2016). The mixture of topsoil, poultry droppings and sawdust substrate gave the widest pileus diameter yield. This is probably due to the combination of the properties of topsoil, poultry droppings and sawdust substrates. The least yield was from topsoil. The observation could probably be due to the nutrient content of the soil. Other inhibitory effects of pathogens present in the soil might have also contributed to the lower yield. This observation agrees with the result of Okhuoya and Okogbo (1991).

As the flushes increase, the quantity of mushroom produced also reduced. This occurred because the nutrients in the substrates were used up to produce the earlier flushes. The fruit bodies from the second flushes were less because the nutrient left within the substrate has been reduced hence, the lower yield.

5. CONCLUSION/RECOMMENDATION

The study concludes that outdoor cultivation of *Pleurotus tuber-regium* is feasible. The mixture of topsoil & sawdust as well as the mixture of topsoil & poultry droppings treatments produced larger number of fruit bodies when compared with those from topsoil and mixture of topsoil, poultry droppings & sawdust substrates.

Considering all the parameters investigated; mixture of topsoil and sawdust treatment is recommended for outdoor production of *Pleurotus tuber-regium fruiting bodies.* Further investigations are encouraged using other agricultural organic substrates for outdoor cultivation of *P. tuber-regium* to find out if they would also support *P. tuber-regium* fruitbodies. This will encourage the use of these substrates in mushroom production; assist in the natural recycling for food and medicine thereby resulting in healthier people and environment.

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