

## GROWTH AND LONGEVITY OF *Pleurotus tuberregium* (FR.) SING AS AFFECTED BY MIXTURES OF DECOMPOSED PALM BUNCH REFUSE AND TOPSOIL

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### ABSTRACT

Screen house study was conducted at the Rivers State University from April to July, 2006 to evaluate the effect of decomposed palm bunch and top soil mixture on the growth and longevity of *P. tuberregium*. The treatments consisted of top soil and decomposed palm bunch refuse in the following ratio T<sub>3</sub> (4:1), T<sub>4</sub> (3:2), T<sub>5</sub> (2:3), T<sub>6</sub> (1:4), T<sub>7</sub> (1:1). The treatments were laid out in a completely randomized design (CRD) with three replications. The treatments were bagged in 75x60cm perforated black polythene bag and watered daily. Growth was observed from 14 days after planting. No growth were observed on T<sub>2</sub> (decomposed palm bunch refuse) T<sub>3</sub> (4:1) and T<sub>4</sub> (3:2). Results of this study revealed that T<sub>6</sub> (1:4) gave the highest emergence 4.47%, highest numbers of shoots (47), regular harvesting per week and highest life span of 73 days. T<sub>6</sub> (1:4) is recommended amongst the various ratios for higher emergence percentage, number of shoots, frequency of harvesting and longevity. From the above result we can conclude that using proper ratios of decomposed palm bunch and top soil increases the production of mushroom. It should be used where emphasis is laid on increased production of fruiting bodies in mushroom cultivation. One part decomposed palm bunch refuse and 4 parts topsoil by weight is then recommended for increased production of *Pleurotus tuberregium*

**Key words:** Sclerotia, *P. tuberregium*, decomposed palm bunch refuse, topsoil, emergence, longevity.

### INTRODUCTION

Most mushroom species are classified under the Phyla Basidiomycota or Ascomycota in the fungi kingdom (Cho, 2004). *Pleurotus* mushroom commonly known as oyster mushroom belongs to the family pleurotaceae, order Agaricales and class Basidiomycetes. It is referred to as oyster mushroom because the pileus is shell-like in appearance. About 1000 species of oyster mushroom have been identified but only about 50 valid species are recognized in the genus *pleurotus* (Guzman, 2000). Commonly cultivated *pleurotus* species include; *P. sajor-caju* (fries) sing, *P. cystitiosus* (ok. Miller) *P. ostreatus*. *P. flabellatus*, *P. sapidous*, *P. tuberregium* etc.

*Pleurotus tuberregium* is a white rot fungus which grows on lignocellulic material

buried in the soil or from degradation of lignocellulic materials. Mushroom breed by spores, found under gills of the pileus (Oei, 2003). These spores, under favourable conditions germinate into hyphae, the germinated hyphae form primary mycelia and then secondary mycelia through plasmogamy. The mycelia absorbs nutrients from the substrate and colonize the substrates. It is the mycelia colony that forms pins, the pin differentiates into pileus, stipe forming the fruiting bodies. Once the substrate is fully colonized, fruiting bodies formed, life cycle completed, sclerotia will be formed to withstand dry season. Isikhuemhen and LeBauer (2004) revealed that sclerotia is an organ formed to withstand unfavourable condition until the return of rainy season at which production of sporophores begins.

Chen and Huang (2004) defined sclerotium as a non-fertile or sterile mycellial structure, tightly woven. Sclerotia are hard in nature and have a life span of about 2-5 years after harvest. The sclerotium of *P. tuberregium* is dark brown on the surface and white inside. The size of sclerotium could be as large as 30cm or more in diameter and over 5kg in weight. In Nigeria, sclerotia are collected from decaying log of wood or soil in thick virgin or secondary forest and consumed or sold at roadside or in the market. Chiejina and Olutokumbi (2010) confirmed that basidiocarps can be induced by burying the sclerotia in soil.

Growth rate of *P. tuberregium* depends on the vigor of the planting material, life cycle of the mushroom, species of the mushroom and number of flushes. One to three flushes can be harvested in *P. tuberregium* per year. In pleurotus, mushroom will be ready for harvesting 5-7 days after sprouting (Oei, 1991). In as much as the sclerotia is still white and firm, it can produce flushes of mushroom 2-3 crop per substrate. The sclerotia can produce fruiting bodies for harvesting every 3-5 days per week however, there may be 2-3 days interval when mushrooms are not available to harvest. It has been recorded that most farmers can harvest for 35-42 days but harvesting can go on as long as 150 days (Ayodele and Okhuoya, 2007; Stamet, 2001).

The sclerotia and fruiting bodies of *P. tuberregium* are sought after for its medicinal benefits and nutritious value (Oso, 1977, Olawuyi *et. al.* 2010 and Oluranti, *et. al.* 2012). Yongabi (2004) confirmed that use of sclerotia in natural water and waste water purification. Many studies have stated the use of pleurotus

species in bioremediation (Isikhuemhen *et al.*, 2003). With these benefits, there is need to research on the effect of the mixtures decomposed palm bunch refuse and topsoil on the emergence, number of shoots, frequency of harvesting, and longevity of the *P. tuberregium* sclerotia. Therefore the objective of the study was to determine the effect of decomposed palm bunch refuse and topsoil on the growth and longevity of *P. tuberregium* sclerotia.

## MATERIALS AND METHODS

### Experimental Site and Climatic Conditions

This study was carried out, at an ambient temperature and atmospheric pressure, in a screen house at the Rivers State University, Port Harcourt. Rivers State University lies within the Nigeria Rainforest zone with an altitude of 38m above sea level and an annual rainfall of 2000mm - 2484mm, relative humidity of about 79% and temperatures of 22.6°C - 31.2°C (FAO, 1984). Rainfall distribution is bimodal with peaks around the month of June and September. The dry season extends from November to March with a characteristic cold dry dust-laden wind interval (Harmattan) during the months of January to February.

### Sourcing of Planting Material

The planting materials (Sclerotia of *pleurotus tuberregium*) were sourced from a local market in Aba, Abia State and transported to the research site. The sclerotia were cut into sizes and weighed on the weighing balance (scale) in the Soil Science laboratory of Rivers State University, Port Harcourt to get an accurate weight of 20g of sclerotia (Ikeoba and Nwocha, 2001).

### Planting Media

The planting media were decomposed palm bunch refuse and top soil, mixed in various ratios. The decomposed palm bunch refuse was collected from an oil mill at Aluu in Ikwerre Local Government Area of Rivers State while topsoil was collected from the Teaching and Research farm of Rivers State University.

### Treatments

The treatments were made up of various mixtures of decomposed palm bunch refuse and top soil in the following ratios by weight; which were used for the cultivation of sclerotia.

**Table 1: Treatment Combinations**

Treatment	DPBR : TP	
T <sub>1</sub>	0 : 5	0kg decomposed palm bunch refuse : 2kg top soil
T <sub>2</sub>	5 : 0	2kg decomposed palm bunch refuse : 0kg top soil
T <sub>3</sub>	4 : 1	1.6kg decomposed palm bunch refuse : 0.4kg top soil
T <sub>4</sub>	3 : 2	1.2kg decomposed palm bunch refuse : 0.8kg top soil
T <sub>5</sub>	2 : 3	0.8kg decomposed palm bunch refuse : 1.2kg top soil
T <sub>6</sub>	1 : 4	0.4kg decomposed palm bunch refuse : 1.6kg top soil
T <sub>7</sub>	1 : 1	1kg decomposed palm bunch refuse : 1kg top soil

DPBR – Decomposed Palm Bunch Refuse, TP – Top Soil

All treatments were replicated three times and bagged in separate (75 x 60)cm<sup>2</sup> black polythene bag.

### Physico-chemical properties of the planting media

The various treatment combinations were analyzed for the following physical and chemical properties: Particle size distribution was determined using the Bouyoucos hydrometer method as described by Gee and Bauder, (2002). Soil pH was determined in a water using 1:2:5 soil liquid ratio (Thomas, 1982). Organic carbon content was determined by the wet oxidation method (Nelson and Sommers, 1996). Total nitrogen was determined by the Kjeldahl digestion method (Bremner, 1996). Available phosphorous was determined by Bray 2 method (Olsen and Sommer, 1982). Exchangeable cation (K<sup>+</sup>, Ca, Mg, Na) was determined in ammonium acetate extraction procedure (Thomas, 1996).

### Mushroom Cultivation

The screen house prior to the onset of the research work, was swept, cleaned and fumigated with Rambo insecticide. 10 pieces of 20g of sclerotia was planted at a depth of 5cm per perforated black polythene bags, containing the various ratios of the decomposed palm bunch refuse and topsoil. The bags were placed on a raised platform in a screen house at room temperature. Each perforated bag containing the planted sclerotia was watered daily with 50cl of water until the sclerotia began to sprout. After sprouting the volume of water for watering was decreased to 25cl, to keep it moist.

### Data Collection

The experiment was observed daily and measurement were taken weekly from the 14 days after planting (DAP). All growth parameters were measured from 50% of the plant population per bag. Percentage emergence was collected by counting

number of sprouted sclerotia per bag divided by number of sclerotia planted per bag, multiplied by 100. Total numbers of shoot harvested per plant per bag were manually counted and recorded for each treatment.

The number of times mushroom was harvested per week from each bag per treatment was recorded and the mean from the number of replicate calculated to get frequency of harvesting.

The number of days from the time of planting up to when the sclerotia ceased to produce fruiting bodies per production unit, was used to estimate the longevity of each unit of production.

#### Experimental Design/Data Analysis

The experimental design used was completely randomization design (CRD) with 7 treatments replicated three times. Data collected were subjected to analysis of variance for a Completely Randomized Design and means were separated using the

least significant difference (LSD) at (5%) probability (SAS 1999).

#### RESULTS

##### Effect of the Various Mixtures of Decomposed Palm Bunch Refuse and Topsoil on Emergence of *Pleurotus tuberregium*

Effects of the various mixtures of decomposed palm bunch refuse and topsoil on the emergence of *P. tuberregium* are as shown in Fig. 1. Results show that there were significant differences among control T<sub>1</sub> (topsoil only) which gave 2.67% and control T<sub>2</sub> (decomposed palm bunch refuse only) where no growth was observed. Among the various ratios of the treatments, there were significant differences with T<sub>6</sub> (1:4) giving the highest percentage emergence of 4.67% while the lowest was given by T<sub>5</sub> (2:3) with 0.33% and T<sub>7</sub> (1:1) with 0.66%. No growth was observed in T<sub>3</sub> (4:1) and T<sub>4</sub> (3:2).

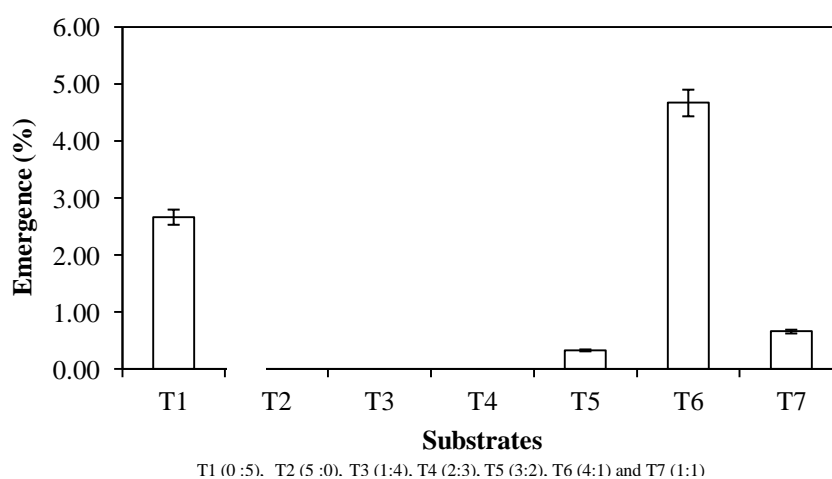


Figure 1: Effect of substrates on emergence of *P. tuberregium*

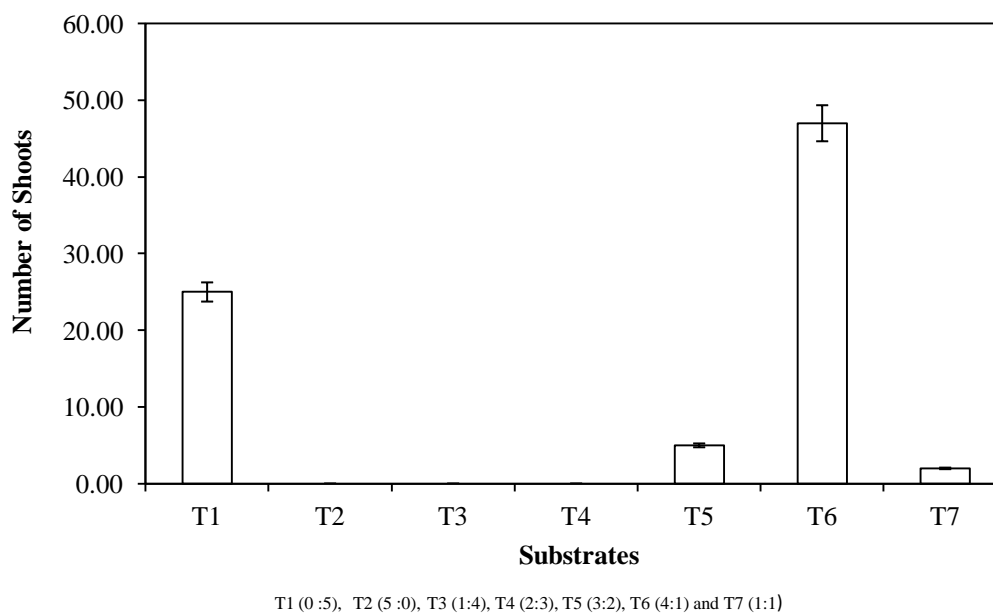
#### Number of Shoots Produced by *Pleurotus tuberregium*

The result in Fig. 2 shows that there was a significant difference ( $P < 0.05$ ) between the

controls and the various mixtures used, on the number of shoots of *Pleurotus*

*tuberregium* produced by each treatment. From the result, T<sub>6</sub> (1:4) recorded the highest number of shoot of 47, followed by control T<sub>1</sub> (topsoil only) of 25, T<sub>5</sub> (2:3) 5 while T<sub>7</sub> (1:1) gave 2 shoots. No growth were observed in control T<sub>2</sub> (decomposed

palm bunch refuse only), T<sub>3</sub> (4:1) and T<sub>4</sub> (3:2). The highest number of shoot were recorded in T<sub>6</sub> (1:4) with 47 which differed significantly ( $P < 0.05$ ) from other treatment/substrate used to grow *Pleurotus tuberregium*.



**Figure 2:** Effect of Substrates on Number of Shoots *P. tuberregium*

### Frequency of Harvesting and Longevity of *Pleurotus tuberregium*

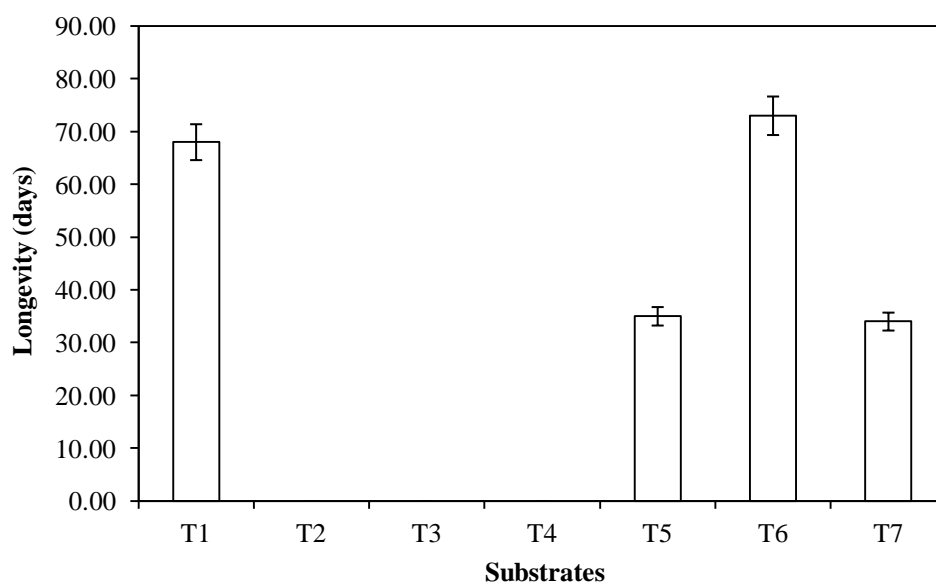
Results of the effect various mixtures of decomposed palm bunch refuse and topsoil on the frequency of harvesting and longevity of *P.tuberregium*, are shown on Table 2 and Fig. 3 respectively. There was irregularity in the frequency of harvesting mushroom per week from control T<sub>1</sub> (Topsoil) while frequency of harvesting per week from T<sub>6</sub> (1:4) was regular. Frequency of harvesting at 2 WAP for control T<sub>1</sub> (topsoil only) was 3 times while T<sub>6</sub> (1:4) recorded 2. From 4 WAP harvesting was recorded at once per week for T<sub>6</sub> (1:4) throughout the vegetative stage. Other

substrate gave significantly low rate of harvesting. Therefore T<sub>6</sub> (1:4) gave the highest frequency of harvesting throughout the vegetative stage.

From the result shown in Table 1, T<sub>6</sub> (1:4) produced mushroom for duration of 73 days before the sclerotia decomposed while control T<sub>1</sub> (topsoil) produced mushroom irregularly for 68 days. T<sub>5</sub> (3:2) gave a life span of 36 days while T<sub>7</sub> (1:1) gave a life span of 35 days. Therefore, T<sub>6</sub> (1:4) had a significantly ( $P < 0.05$ ) longer life span of 73 days than other substrates used in production of *P. tuberregium* at the vegetative stage.

**Table 2: Effect of Various Mixtures on the Frequency of Harvesting of Mushroom Per Week**

Substrates	2WAP	3WAP	4WAP	5WAP	6WAP	7WAP	8WAP	9WAP	10WAP
T <sub>1</sub> (0:5)	3	1	0	0	0	1	0	1	1
T <sub>2</sub> (5:0)	0	0	0	0	0	0	0	0	0
T <sub>3</sub> (4:1)	0	0	0	0	0	0	0	0	0
T <sub>4</sub> (3:2)	0	0	0	0	0	0	0	0	0
T <sub>5</sub> (2:3)	0	0	0	0	1	0	0	0	0
T <sub>6</sub> (1:4)	2	0	1	1	1	1	1	1	1
T <sub>7</sub> (1 : 1)	0	0	1	0	0	1	0	0	0



T<sub>1</sub> (0 :5), T<sub>2</sub> (5 :0), T<sub>3</sub> (1:4), T<sub>4</sub> (2:3), T<sub>5</sub> (3:2), T<sub>6</sub> (4:1) and T<sub>7</sub> (1:1)

**Figure 3: Effect of Substrates on the Longevity of the Sclerotia**

### Nutrient Composition and Particle Size of Substrates Used in Growing *Pleurotus tuberregium*

Table 3 shows the nutrient composition and particle size of substrates used in growing *Pleurotus tuberregium*. The result of the nutrient composition and particle sizes of all the treatment used to grow *Pleurotus tuberregium* reveals that all the treatments are alkaline in nature with pH value ranging from 8.5-9.6. The highest organic carbon was recorded in control T<sub>1</sub> (topsoil only)

and the least was by control T<sub>2</sub> (decomposed palm bunch only) which gave 1.11%. Nitrogen content of other substrates was same range of 0.02-0.04. The highest phosphorous (p) of 842.11 cmol/kg was recorded in T<sub>4</sub> (3:2) while the lowest of 29.74 cmol/kg was recorded in control T<sub>1</sub> (topsoil only). The highest potassium (k) value of 1400.00 cmol/kg was recorded in T<sub>6</sub> (1:4) while the lowest potassium (k) value of 29.74 was recorded in control T<sub>1</sub> (topsoil only). Control T<sub>2</sub> (decomposed

palm bunch refuse only) was highest in sodium (Na) 11.30 cmol/kg while the lowest was 0.33 cmol/kg in control T<sub>1</sub> (topsoil only).

Calcium (ca) content of T<sub>3</sub> (4:1), T<sub>4</sub> (3:2), T<sub>5</sub> (2:3) were 13.40 cmol/kg, 13.60 cmol/kg, and 13.60 cmol/kg respectively while T<sub>7</sub> (1:1) recorded 12.60 cmol/kg. Control T<sub>1</sub> (topsoil only) and T<sub>6</sub> (1:4) recorded calcium value of 9.60 cmol/kg and 9.20 cmol/kg respectively. Control T<sub>2</sub> (decomposed palm bunch refuse only) recorded 7.40 cmol/kg.

For magnesium, T<sub>6</sub> (1:4) gave the highest magnesium (mg) value of 9.40 cmol/kg while the lowest is control T<sub>1</sub> (topsoil only) 0.40 cmol/kg.

Particle size analysis as shown on Table 2 indicates that the textural class of control T<sub>1</sub> (topsoil only) to be sandy loam, control T<sub>2</sub> (decomposed palm bunch only) is sandy clay loam, T<sub>3</sub> (4:1) is sandy clay loam, T<sub>4</sub> (3:2) is sandy clay loam, T<sub>5</sub> (2:3) is sandy clay loam, T<sub>6</sub> (1:4) is sandy loam while T<sub>7</sub> (1:1) is sandy clay loam.

**Table 3: Nutrient Content and Particle Sizes of Substrate used in cultivation of *Pleurotus tuberregium***

S/N	Parameters	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>	T <sub>4</sub>	T <sub>5</sub>	T <sub>6</sub>	T <sub>7</sub>
1.	Sand (%)	80.00	58.00	62.00	68.00	72.00	78.00	72.00
2.	Silt (%)	3.40	7.40	9.40	5.40	5.40	3.40	7.40
3.	Clay (%)	16.60	34.60	28.60	26.60	22.60	18.60	20.60
4.	Textural Class	Sandy loam	Sandy clay loam	Sandy clay loam	Sandy clay loam	Sandy clay loam	Sandy loam	Sandy clay loam
5.	Soilp H <sub>w</sub> (1:2.5)	8.50	9.60	9.60	9.60	9.50	9.30	9.40
6.	Organic Carbon (%)	2.44	1.11	1.58	1.83	1.79	2.36	2.30
7.	Total Nitrogen (%)	0.02	0.04	0.04	0.02	0.02	0.04	0.02
8.	P (cmol/kg)	82.46	701.75	196.49	842.11	526.32	456.14	210.53
9.	K (cmol/kg)	29.74	323.08	230.77	174.36	35.90	1400.00	117.95
10.	Na (cmol/kg)	0.33	11.30	7.83	5.57	5.04	2.09	4.52
11.	Ca (cmol/kg)	9.60	7.40	13.40	13.60	13.40	9.20	12.60
12.	Mg (cmol/kg)	0.40	3.80	0.60	0.60	3.00	9.40	2.00

T<sub>1</sub> (0 :5)-Topsoil (TS), T<sub>2</sub> (5 :0)-Decomposed palm bunch refuse (DPBR) (Control T<sub>2</sub>), T<sub>3</sub> (1:4)-1 part DPBR + 4 part TS, T<sub>4</sub> (2:3)-2 part DPBR + 3 part TS, T<sub>5</sub> (3:2)-3 part DPBR + 2 part TS, T<sub>6</sub> (4:1)-4 part DPBR + 1 part TS, T<sub>7</sub> (1:1)-Equal part DPBR + Equal part TS

## DISCUSSION

### Percentage Emergence of *Pleurotus tuberregium*

Emergence of *Pleurotus tuberregium* from the different substrates occurred at different rates at 2 weeks after planting. This difference in emergence recorded in the different substrates may properly be as a result of the difference in their total carbon and Nitrogen as shown in table (2). This is because total carbon and total nitrogen of

substrates used to grow mushroom, have effects on mycelium growth, formation and development of fruiting bodies. This is in line with the report of Narain *et al.* (2011) that mycelium growth and primordial development of *pleurotus florida* were dependent on the lignocellulic materials especially C/N ratios; hence emergence rate recorded in control T<sub>1</sub> (topsoil only) 2.67%, T<sub>6</sub> (1:4) 4.67%, T<sub>5</sub> (2:3) 0.33% and T<sub>7</sub> (1:1) 0.66% has some relationship with their

respective C/N ratio found in their substrate. This is confirmed the finding of Albores *et al.* (2006) that there is a positive relationship between the C/N ratio of substrate and rate of pinhead formation.

### **Number of Shoots of *P. tuberregium* Produced**

There were significant difference in the number of shoots produced by each substrates. The ability of T<sub>6</sub> (1:4) to produce the highest number of shoots in comparism with other substrates may be due to the presence of a higher level of potassium (k) 1406.00 cmol/kg, phosphorous (p) 456.14 cmol/kg, magnesium (mg) 9.40 cmol/kg and a considerable higher amount of nitrogen 0.04 cmol/kg as shown in table (4.9). This observation agrees with the findings of Mangat *et al.* (2008) that phosphorous, potassium and nitrogen enhance mushroom production. Consequently, T<sub>6</sub> (1:4) produced more flushes with the resultant higher number of shoots.

### **Frequency of Harvesting *P. tuberregium* and Longevity**

Frequency of harvesting mushroom from the different substrates is directly proportional to the number of flushes produced by each substrate throughout the lifespan of the sclerotia.

This ability of T<sub>6</sub> (1:4) to produce more flushes, higher frequency of harvesting and longer life span is properly due to its higher nutrient content. The longevity of *P. tuberregium* in this experiment confirms the reports Ayodele and Okhuoya (2007); Stamets (2001) that most farmers harvest oyster mushroom for 35-42 days but harvest can go on as long as 150 days.

This study has revealed that decomposed palm bunch refuse and topsoil can be mixed

to enhance the productivity of *Pleurotus tuberregium*, however the appropriate ratio is required. Result shows that 1part decomposed palm bunch and 4 parts topsoil can promote high percentage emergence, increase in number of shoots and longevity of production unit.

Results showed decomposed palm bunch refuse to be richer in phosphorous; which is required for cell division and elongation in plants and differentiation of secondary mycelia to form pin head and fruiting bodies. It is therefore, recommended that it should be used for mushroom cultivation where the emphasis is on increased production of fruiting bodies.

One-part decomposed palm bunch refuse and 4 parts topsoil by weight has been shown to be the best ratio for increased production of *Pleurotus tuberregium*; and therefore, recommended.

More research is suggested on mixing of different substrates with topsoil; with particular emphasis on particle size and pH, so that the nutrient in the formulation can be easily absorbed by sclerotia planted in the formulation. Also on the effect of acidity of soil in the production of *P. tuberregium*.

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