Application of *Pleurotus ostreatus* SMC as soil conditioner for the growth of soybean (*Glycine max*)

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Abstract: Spent mushroom compost (SMC) of *Pleurotus ostreatus* (an edible fungus) was used as soil conditioner for the improvement of growth of a leguminous agricultural crop (Soya bean (*Glycine max*). The experiment was conducted in a screen house, located behind the Department of Botany and Microbiology, University of Ibadan. The experiment was laid out in a Complete Randomized Design and Replicated with six(6) treatments; 0%,10%,20%,30%,50% and 100% Results from this investigation shows that this dicot, soybean performed well at 10% (0.6kgSMS/6kg soil) treated level on most of its agronomic characters and yield (pod no, FW and DW) parameter. It was observed that, there were seed production on the control experiment plants, but were significantly lesser with the SMC treated seeds/plants($p \leq 0.05$),. However the control (soil) and Ref. Control (SMS only) produced less fruit (Pod) . It was also found that the substrate pH for growing these crops at all treatment level was increased from acidity to neutrality. The results from these findings were discussed in relation to the usage of SMC as a possible organic fertilizer for the improvement of this leguminous crop.

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1. Introduction

Soybean is a plant of the family Fabaceae, order Fabales . The height of the plant varies from below 20cm (7.9in) up to 2 metres (6.6 ft). It consists of approximately 8% seed coat 90% cotyledons and 2% hypocotyledons. The pods, stems and leaves are covered with fine brown or gray hairs. The leaves are trifoliate , having three to four leaflets per leaf, which are 6-15cm or 2.4-5.9in long and 2-7cm (0.79-2.8in) broad.Soya bean is among the new 40 varieties of five crops across targeted countries released by scientist working with the tropical legume II(TLII) project , funded by Bill and Melinda Gates Foundation in kwara state, Nigeria (Bulletin of Tropical Legume,2011)

SSA Soil fertility has been reported as one of the major constraints of tropical agriculture. This is due to the fact that Sub-saharan Africa's potential to grow food crops has been declining every year because of degradation in soil fertility (Yaker 1993; Brader 1995 as cited In (Oyetunji *et al* 2002).These SSA soils are very susceptible to nutrient depletion due to farming intensification, which is associated with low buffering capacity (kang and Wilson 1987; Tian *et al* .1994), and Coupled with the inability of farmers to procure the necessary imput to support the infertile soils

Edible mushrooms are a group of fungi species (Basidiomycetes) that grow naturally on tree trunks, leaves, root of trees as well as decaying woody materials (Stamets 2000; Lindequist *et al.*, 2005).They are achlorophylous organisms and thus can be grown in jars, bottles devoid of sunlight. From these mushrooms are deposits of lots of nutrients, very vital for the use as crop enhancer. Nutrients such as vitamins, minerals and micro and macro nutrient have been detected in most oyster mushrooms (Jonathan,*et al.,*2012a). Oyster mushrooms such as *Pleurotus ostreatus* is a fungus, that belongs to the class basidiomycetes. They are generally understood to be called white rot fungi, because of their ability to degrade lignocellulosic materials. *Pleurotus* species as primary rot fungi are able to colonize different agricultural wastes as substrates. *P. ostreatus* is among the edible mushrooms reported to be cultivated in Nigeria (Jonathan and Esho, 2010).*Spent Mushroom Compost* (SMC) is the remnant of substrates (sawdust any other agricultural substrates) used to cultivate a mushrooms (Jonathan *et al.,*2012b). It is a by-product of the mushroom industry after different flushes of mushroom fruitbodies have been harvested (Chang 1981; Fasidi *et al*., 2008).It is believed that the importance of industrial fertilizers in developing modern farming practices and provision of food for the world wide population has been acknowledged. However, some of these inorganic fertilizers could pose adverse effects on crops. Hoffman and Smith (1993), for instance reported that the application of potassium to citrus fruits could affect the shape of their fruit abnormally, and also increases their acidity. The susceptibility of pome fruit to physiological disorders, and the decrease in fruit color , when nitrogen fertilizer was applied (Shear and Faust,1980). Moreso, the flavor of apple fruit have also been affected when , high rate of nitrogen fertilizer , were applied to apple tree (Link 1980). In addition, the post harvest physiology of cucumber fruits, by affecting membrane lipid chemistry, membrane integrity and respiratory metabolism have been reported (Knowles *et al* ., 2001), when phosphorus nutrition was applied. These are all the negative effects of the excessive use of inorganic fertilizers. In any case these effect could be avoided when an alternative such as spent mushroom compost which is organic in nature can be employed. However SMS when allowed as waste in the environment can serve as environmental pollution source thereby causes nuisance to the environment which is harzadious. But this harzardous effect can be turned around for fortune when used as a substrate for growing agricultural crops. The use of spent mushroom compost in growing agricultural crops has been recognized in recent times as a possible means of enhancing sustainable agriculture or production of food crops(Olfati et al 2011).

Environmental pollution with excessive fertilizers with heavy metal contents is a global concern being everywhere and has also been reported. Plants that are termed phytoaccumulative are those that have the ability to absorb and utilize heavy metals from the soil, displaying attributes of phytoremediation (Memon and Schroeder 2009). There have been records of harzadious effect of fertilizers on the environment and on plant at large which do not have the ability to utilize heavy metals present in fertilisers. For instance inorganic fertilizer is known to play a significant role in environmental pollution.Among which, nitrogen fertilizer increases denitrification , resulting in elevated emission of nitrous oxide (N_2O) to the atmosphere, and thus , contributing to global warming (Smith *et al* .,2008).The application of nitrogen fertilizer, has also been reported to possess the potential of depleting soil organic carbon with time (khan *et al.,* 2007).

In consideration, a need arises as a necessity to consider the use of biofertilizer, devoid of toxic metals, which has the ability to prevent the depletion of the soil organic matter, according to Jeyabal and Kuppuswamy(2001).And also which can be easily obtained, from mushroom farms at no cost or cheap rates. As the world drifts into practicing “Organic farming” in agriculture, the need to embrace the use of Biofertlizer, such as SMS should be encouraged. This study, is therefore aimed at, (i) Studying the responses of the aforementioned crop to the presence of Spent Mushroom Compost as a biofertilizer . (ii) To determine the quantity and quality of nutrients in SMS appropriate for growing agricultural crops such as cowpea, and how it affect its pH .Hence it was the objectives of the present investigation, to apply spent mushroom compost on Soya bean ,to find out its effect on growth and yield attributes of the crop. The Nutrient status of soil and Compost were evaluated , percentage of pods produced per six kilogramme soil were enumerated at harvest. Nutrient contents and uptake of whole crops were determined. Number of leaves, plant height and leaf length were the growth parameter sampled as data collected, together with the total yield produced.

2. Materials and Methods

2.1. Collection of and source of SMC and planting materials: The spent mushroom composts utilized were obtained from a Mushroom cultivating farm at odo-ona kekere Ibadan city, Nigeria. The compost/substrate was a remnant of the material (sawdust) used to cultivate *P.ostreatus* The seeds of *Glycine max*; TGX1740 were all collected from the Institute of Agriculture and Research Training (I.A.R& T) Moore Plantation ,Ibadan, Nigeria.

2.2. Preparation of the growth media. The spent mushroom composts substrates were weighed in different quantities (0.6kg, 1.2kg 1.8kg, 2.4kg and 3.0kg representing 10%, 20%, 30% 50% /6kg soil). Each of these quantities was properly mixed with 6kg of depleted garden soil, which was collected from a cultivated land behind the Department of botany at the screen house, at the University of Ibadan . Each mixture was packed into a 6kg bucket of soil and adequately watered (Iwase *et al* 2000). The treatment for each was replicated three times (Jonathan *et al*., 2012a).

2.3 Soil treatment: To obtain fine sand, the soil was sieved (with a 2mm wire gauzed) metallic tray, in order to remove stones , plant debris and generally unwanted materials that could hinder plant growth. After this, the soils present in every bowl were treated (supplemented) with SMS except control bowls(0%) at the following varied concentrations; 10%, 20%, 30% and 50%. 10% SMS of 6kg soil require 0.6kg of SMS, while 20% ,30%,and 50% SMS all required 1.2kg,1.8kg and 3.0kg respectively. The thoroughly mixed substrates were allowed to decompose after wetting for about seven to ten days for biological and derivative activities of microbes to occur, which was expected to improve soil structure , composition, and more so its nutrient.

2.4. Experimental design and experimental set-up:

The experiment was designed in a Complete Randomized Design (CRD) which was set up in 3 Replicate, while the Treatment was done at six levels (0,10, 20, 30, 50 and 100) . Two controls, viz; SMS (100%) and SOIL (0%) were utilized.The CRD method of placing pots at the start of the experiment in the screen house;

Treatment In Percent, (0- 100%), Replicate Scattered.

Cowpea

A	B	C
0%	10%	20%
100%	30%	50%
10%	20%	0%
50%	100%	30%
0%	10%	50%
20%	30%	100%

2.5. Pre-Planting Analyses: The soil and SMC were tested for their physicochemical properties, prior to utilization for the study. 54 bowls were used for this experiment, and all were punctured underneath for drainage and proper aeration circulation. Soils were measured with a top weighing balance (Hana) in Kg. 6kg of soil were required and measured. 54 buckets were filled with soils of these masses. SMS were also measured and applied to the soil according to the treatment levels *viz*; 10%(0.6kg) through 50%(3kg).i.e

$SMS_{0\%/6kg\ soil}$; 0.0kg/6kg soil(No SMS)
$SMS_{10\%/6kg\ soil}$; 0.6kgSMS /6kg soil
$SMS_{20\%/6kg\ soil}$,; 1.2kg SMS/6kg soil
$SMS_{30\%/6kg\ soil}$; 1.8kg SMS/6kg soil
$SMS_{50\%/6kg\ soil}$; 3.0kgSMS/6kg soil
$SMS_{100\%/0kg\ soil}$; 6.0kg.SMS (No Soil)

2.6. Planting method: Direct planting method was employed; seeds were placed in the soil and treated soil at 2-3cm for all seeded crops(Jonathan *et al*.,2011). This experiment was carried out in the screen house, Department of Botany in the University of Ibadan. The was prepared according to the method described by Fasidi *et al*.(,2008).

2.7 Seed viability testing: All seeds of TGX1740 (Soya bean), were tested for viability, by planting in soil to study their germination. Result showed 100% viability, meaning the seed were all viable. The method employed was obtained from (www.Pulse-point-20seed viability testing pdf-Adobe Reader) Germinated seedlings/Total seedling X 100/1.

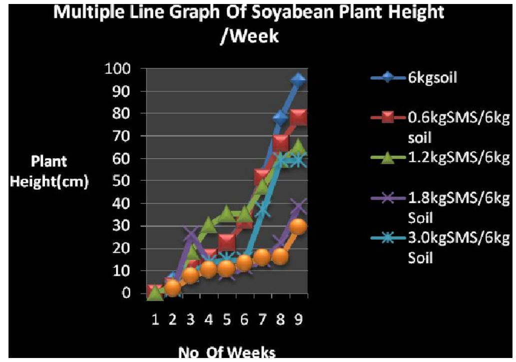

Figure 1. The plant height of soyabean

2.8. Chemical analyses The mineral element compositions of the SMC were determined before their application to the soil. This was done in order to know the various elements that were present in the spent mushroom substrate before usage. After the completion of the experiment the various substrate were also analyzed to know their utilization level. The mineral element analyses were carried out using the procedures of Association of Official Analytical Chemist (1990).

2.9. Planting method: Direct planting method was employed; seeds were placed in the soil and treated soil at 2-3cm.

2.10.Growth parameters and measurement: The growth parameters (Plant height, leaf length, leaf width, number of Leaves, girth size, and plant yield, fresh and dry weight and(total plant biomass) were monitored. Plant heights, leaf length, leaf width were determined using a meter rule (in cm). Girth sizes were read with a micrometer screw gauge with (x0.001mm) calibration, which were later multiplied by the circumference formula (π=3.142) while fresh and dry weight were measured with a sensitive weighing balance. Number of leaves were counted and recorded , on weekly basis, while the stem girths were taken once every two weeks, four times. The calculation of the individual plant species leaf area were effected with the formula A= (L x W x CF) proposed by Montgomery (1911), where L is the leaf length, W is the leaf width, and CF is the coefficient factor. The following coefficient factors were used for the calculations of leaf area for the different crops; 0.75 for maize (Montgomery 1911), as cited in McKee (1964), 2.7x LA for Cowpea (Jolaoso, 1988) and 0.65 for Soybean; (Wiersma and Bailey (1975), as cited in (Zur *et al* 1988).

2.11. Determination of exchange acidity in soil: 5g of air-dry soil was weighed into 50ml, centrifuge tube. 30ml of MKCl was added and the Centrifuge was tightly covered with a rubber stopper. it was shaken for 1 hour. The content was centrifuged at 2000rpm for 15 minutes and then decanted carefully the supernatant into a 100ml volumetric flask. Again, 30ml of MKCl was added to the soil sample, and shaken for 30 min.This was repeated, until the supernatant is clear. The clear supernatant was transferred into the same volumetric flask .The step was repeated again, until it was made up to 1 MKCl mark (AOAC, 1995).

2.12. Determination of soil micronutrient extraction: 5g of soil was weighed into a 100ml plastic bottle .50ml of HCl, was added, and Shaken for 30mins. The mixture was filtered through No. 42 filter paper. Then the presence of Cu, Zn, Fe, and Mn on atomic absorption spectrophotometer were then determined.(Baker and Amacher,1982).

3.Results and Discussion

3.1 Effect of SMC on soybean plant height

Soyabean plant had better plant height at 0% (untreated soil) , with values of 57.17cm; 27.3% and 44.9cm; 23.3% germination respectively, as compared to others. These values are larger than that on 100% treated sample. (SMS alone) (15.04cm) . These values suggested that, dicotyledonous plant tend to slow in increase in height , when they are supplied with nutrients as unfertilized soil, possess the highest , height , when compared with others.

3.2. Effect of SMS on soybean leaf numbers

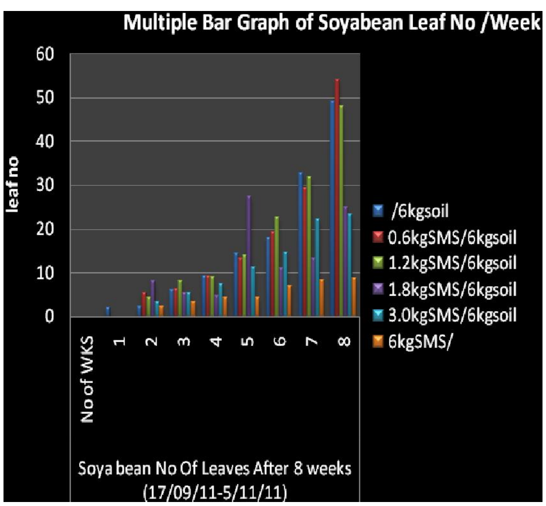

Fig. 2. Effects of SMC on Soybean leaf numbers.

3.3. SMC Effect on soybean stem girth. There were no significant difference between the stem girths of all the treated level of the soyabean plant(NS).

Table 1. Effect of SMC on Soyabean stem girths(cm) 8 wks of planting

SMS TRT	Glycine.max (Mean±SEM*)
0%	1.02± 0.88
10%	1.02± 0.74
20%	1.23± 0.62
30%	1.00± 0.91
50%	0.74± 1.01
100%	0.36± 0.27
LSD@5%	NS

*SEM = Standard Error of Mean, NS = Statistically Non significant.

Soyabean Leaf number is represented by a multiple bar graph (Figure 2), and it reveals that untreated soil had the first leaves to unfold, during the first week, followed by others. However the growth of the other varied treated soil aside control, remained almost constant till week 5, where 30% had a leaf number approximately 30(Figure2).This leaf dropped to a total number of 11, as a result of defoliation while that of 20%, 10% and 0% sprouted well. But finally 10% treated soil had the highest leaf numbers (Figure 2).

For the reference control SMS(100%) , the total leaf number was 3, at week 1& 2 , which later defoliated to a total of 2, at week three , remaining the same through week 8. This clearly signifies that excessive nutrient is dangerous to a plant's health and should be discouraged hence, plant should not be planted directly in SMS, a claim supported and suggested in SMS brochure(2006) and Manahan (2010).

For the plant height of soyabean , the multiple line graph is used for representation and it shows that 0% (untreated soil) for soya bean plant had the best height, approximately 97cm (Figure 1). This is followed by 10%, while 20% treated soil increased slightly above that of 30% treated soil having values of 66.5cm. The present obvious SMS effect on the plants strongly denotes that nutrients should be applied to plant at moderate levels as excessive or abnormal applications could cause growth damages as observed by (Fry 1987).This plant had a poor growth and leaf discoloration, when planted in SMS directly (Plate not shown) but SMS supplemented soil had soyabean plants showing considerably plants with healthy nature(pictures not shown) . The discoloration and stunted growth on Soyabean plant clearly shows that planting directly into SMC/SMS could be highly toxic (Fry 1987; Wallace 1961) and this is why the use of direct planting into SMS has been discouraged (SMS Brochure, 2006)., shows Soyabean, growing on soil/ substrate treated with 0.6kgSMS (10%treated level) had the best growth based on fresh weight of pods (14.93 ±7.52).

3.4 Effect on Soyabean number of pods

Result from table 2, shows the number of pods produced on Soyabean plant were higher on treated soil/ substrate , when compared to untreated(0%), and highest level of treated substrate (100% SMS; reference control). Soyabean plant did best at 10% with a total number of pods of 45.33±4.096, with a biological efficiency of 24.9% ,while 50% and 20 % Supplemented soil had the same value (41.33);and the same B.E(Biological efficiency) of 22.7% respectively.

Table 2 : SMC effects on no of pods of *Glycine max*

SMS level	*Glycine max* Pod no (Mean±SEM*)	%Germination
0%	4.33c ± 4.333	2.4
10%	45.33a ±4.096	24.9
20%	41.33ab ± 4.096	22.7
30%	20.16 b ± 4.619	13.3
50%	41.33ab ± 8.951	22.7
100%	0.000c ± 0.000	0.0
LSD@ 5%	0.67	

*SEM=Standard error of Mean

Means having the same superscript letter(s) are not significantly different at the probability level (P<0.05), by LSD.

3.6. Effect of SMS on Soyabean fresh weight.

10% treated soil , produced the highest weight value (54.50± 7.77 g), having percent germination of 24.8% . Comparing these value of soyabean to that of control (0%)and the excess reference fertilizer(SMS alone) it will be noted that 10% treated soybean plant had a higher value.0%, and 100% had values of 41.86g, and 0.000g respectively.

Table 3:SMC Effect on fresh weight(kg) of *Glycine max* 8 weeks after planting.

SMC Level	*Glycine max* (Mean ±SEM*)	%Germination
0%	41.87 ± 9.07^{bc}	19.1
10%	54.50 ± 7.77^{a}	24.8
20%	38.56 ± 5.23^{cd}	17.6
30%	37.93 ± 5.62^{de}	17.3
50%	46.50 ± 6.50^{ab}	21.2
100%	0.00 ± 0.00^{e}	0.0
LSD @ 5%	0.94	

Means having the same superscript letter(s) are not significantly different at the probability level (P<0.05), by LSD,

*SEM =Standard error of Mean

3.7. *Effect on yield: Pod Fresh Weight*

Soya bean plant on 10% treated soil had the best pod fresh weight of 14.93g, which is slightly different from 30% treated soil, having 14.23g, and also from 20% having 13.87g. A remarkable difference is observed, when compared with both control levels (0% and 100%) with pod fresh weight of 1.667g, and 0.00g respectively, while 50% had a fresh weight of 1.33g

Table 4: SMC effect on fresh weight(kg) of *Glycine max* pods per plant 8 weeks after planting

SMS Level	*Glycine max* (Mean SEM)	Pod(g) %Germination
0%	1.67 ±1.67bc	3.6
10%	14.93±2.52ab	32.4
20%	13.87±2.14cd	30.1
30%	14.23± 1.76ab	30.9
50%	1.33± 0.67de	2.9
100%	0.00± 0.00e	0
LSD@ 5%	0.37	

*SEM =Standard error of Mean.

Means having the same superscript letter(s) are not significantly different at the probability level (P<0.05), by LSD.

3.8. Effect of SMS on the growth parameters*:*

Plant height(cm), Number of leaves, stem girth(cm) and leaf Area(cm^2) are represented on Fig1, 2 and tables 1 and 8. The highest plant height grew on untreated soil (0%). But both the stem girth and number of leaves had better values at 20% treated soil /substrate, with the following values; 1.23cm, and 24.5cm respectively .while the leaf area, was best at 10% (19.51cm^2).Table , also shows that soyabean plant grew tallest in height on the 0%soil(44.87±10.15) compared to others.

Table 5: SMC Effect on Soyabean Leaf Area(cm^2) after 8 weeks

SMS TRT	Glycine.max
0%	17.81ab± 2.80
10%	19.51a± 2.96
20%	16.24bc± 1.78
30%	11.39de± 1.04
50%	15.79cd± 3.02
100%	4.78e± 0.87
LSD@5%	0.98

3.9 SMC Effect on substrate pH

Table shows the range of increase in pH values. There is a considerable constant increase in the pH of the substrate from the fourth week, which is slightly acidic to the end of the experiment, near neutrality It is known that most crops grow best within a soil pH of 6-7, in which crop nutrient exist in an available form that can be taken up by plant roots(Anonymous authors, 2002), unlike that observed in the substrates of this work (Table 1) . In this study, the pH of the growing medium , containing the various(treated) crops ,were taken after the fourth week , and generally it was observed that the minimum mean pH value was . However , on the Glycine max(soyabean) soil/ substrate, the range of pH falls between5.42 on 50% treated soil to 5.95 and 5.99 on both 20% and 30% soil. It is known that pH influences plant growth indirectly by affecting nutrients availability.

Table.6 Effect of SMS on Soil pH

Sample/Week	0	1	2	3	4	5	6	Mean
Soil(in KCl)	6	0	0	0	(inH2O)		6.7	
SMS		7.5						
0%(G.max)		0	0	0	5.55	5.61	5.66	5.61
10%(G.max)		0	0	0	5.13	5.17	5.22	5.2
20%(G.max)		0	0	0	5.97	5.99	6	5.99
30%(G.max)		0	0	0	5.77	5.95	6	5.91
50%(G.max)		0	0	0	5.42	5.52	5.63	5.5

The increase in soyabean yield is in line with that of Steward(1995) and Steward et al(1997) who jointly observed in their work that SMS application increases yield of potato. Similarly Maynards (1994) and Wang *et al* (1994) reported that vegetable production could be sustained with the application of SMS. Additionally, Chang and Yau (1981) and Iwase *et al* 2000 reported its use in increasing production of tomatoes sevenfolds. The Dry weigh and Fresh weight matter of Soybean plant were significantly increased and this is also in line with that of Mullen and Mcmahon (2001). Again this work reveals the fresh weight, dry weight, fruit fresh weight of, soybean pod no , plant height, leaf number and leaf area, have also been significantly influenced by SMS at (P< 0.05).

The loss of organic matter is believed to be as a result of conventional agricultural practices ,therefore it is advisable to employ organic agriculture which employs closed cycles of energy and materials, by maximixing reuse, and exercising the use of nutrients of organic origin. This work clearly reveals that organic fertilizer type such as SMS can significantly affect at the probability level (P<0.05), plant total yield, number of pods per plant, pod dry weight, pod fresh weight, number of leaves, leaf area, and fruit (Pod) fresh weight , it is therefore advisable to use SMS as a substitute for cow manure as a soil conditioner(J.A Olfati *et al* 2011), at moderate levels.

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