

Full Length Research Paper

Growth of *Lentinus squarrosulus* (M.) Singer on sawdust of different tropical tree species

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***Lentinus squarrosulus* (Mont.) Singer was cultivated on the sawdust of five economic tropical tree species; *Chlorophora excelsa*, *Celtis zenkeri*, *Guera cedrata*, *Nesogordenia papaverifera* and *Brachystegia*, collected during processing at the sawmill. The highest yield of fruiting bodies (16.17 ± 1.25 g) was observed on *B. nigerica* sawdust supplemented with 1% CaCO₃, 1% sugar and 20% wheat bran. The lowest yield (4.56 ± 1.84 g), was observed on *N. papaverifera* sawdust without supplementation. The results of this study demonstrate that *L. squarrosulus* mushroom did not show any particular trend with increase in supplementation level. The study underscores the need for guided use of sawdust types along with the required appropriate supplements.**

Key words: Agro-industrial waste, bioconversion, cultivation, edible mushroom, sawmill waste.

INTRODUCTION

Production of edible mushrooms in Nigeria is almost at the level of commercialization. However, the practical field application of many laboratory results still leaves much to be desired. Many still depend largely on the mushrooms harvested from the wild.

In Nigeria, many mushrooms are highly prized, not only as food but also in traditional medicine (Oso, 1977; Alabi, 1990).

Lentinus squarrosulus (Mont.) Singer is one of the edible mushrooms very common in the southern part of Nigeria (Nicholson, 1989; Oso, 1975; Akpaja et al., 2003). The Ibo people of Nigeria have even devised a peculiar way of tenderizing the usually tough overripe fruit bodies by dressing it, robbing it with edible oil and thereafter wrapping it in leaves. The wrap will then be placed in a fire place. This mushroom is very popular in Nigeria and has been highly recommended for commercialization (Okhuoya, 1997).

A study by Wuyep et al. (2003) on this mushroom indicated its production of lignin and cellulose degrading enzymes. It is also reported to be widespread throughout equatorial Africa, South-East Asia, the Pacific Islands and Australia (Neda and Noi, 1998). Despite the widespread acceptance of this mushroom as an edible species in

Nigeria, there is lack of information about its cultivation. More importantly cultivation on sawdust being the most readily available and cheapest organic waste in Nigeria has not been investigated.

This study reports on the cultivation of this mushroom species using sawdust of different economic tree species.

MATERIALS AND METHODS

Spawn preparation

Pure *L. squarrosulus* (strain LS001UBNIG) was obtained from the Mushroom Biology Unit of the Department of Plant Biology and Biotechnology, University of Benin, Benin City, Nigeria. The spawn was prepared using Sorghum (*Sorghum bicolor*) grains as substrate.

Empty salad cream bottles (275 ml) (readily available as discs) were filled with parboiled sorghum grains (75% water) to three quarter full and covered with cotton wool. The bottles were autoclaved at a temperature of 121 °C and 1.05 Kgcm² pressure for 20 min, allowed to cool down overnight and were inoculated. The inoculated grains were incubated at room temperature (25 °C). After the grains have been fully colonized, they were kept in the refrigerator (5 °C) until used.

Collection of substrates

Sawdust from the following economic trees; *Chlorophora excelsa* (Welw.) Benth. (Moraceae), *Celtis zenkeri* Engl. (Ulmaceae), *Guera cedrata* (A.Chev.) Pellegr. (Moraceae), *Nesogordenia papaverifera* (A. Chev.) R. Capuron (Sterculaceae) and *Brachystegia nigerica*

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Table 1. Effect of sawdust type on the time of *Lentinus Squarrosulus* mycelial colonization.

Substrate mixture	Time of full mycelial colonization (days)			
	C1	C2	10% WB	20% WB
<i>Chlorophora excelsa</i>	13.80±2.35*	9.80±0.58	17.50±6.50	17.00±0.00
<i>Celtis zenkeri</i>	9.60±0.51	7.80±0.49	13.00±0.58	N.A
<i>Guera cedrata</i>	10.40±0.68	9.60±0.51	13.40±2.16	16.33±2.91
<i>Nesogordonia papaverifera</i>	10.20±0.37	10.25±0.25	12.75±1.93	15.00±0.00
<i>Brachystegia nigerica</i>	10.00±0.32	10.00±0.78	16.67±0.33	17.00±0.00

* = Mean of five replicates ± standard error.

C1 = 100% sawdust

C2 = 98% sawdust, 1% CaCO₃, 1% sugar.

10% WB = 88% sawdust, 10% wheat bran, 1% CaCO₃, 1% sugar.

20% WB = 78% sawdust, 20% wheat bran, 1% CaCO₃, 1% sugar.

N.A = Not available.

Hoyle and A. P. D. Jones (Fabaceae) were obtained during timber processing from Uwasota sawmill, Benin City, Nigeria. Wheat bran was bought from an animal Feed Dealer at textile mill road stores in Benin City. Calcium carbonate (CaCO₃) was bought from a chemical dealer while sugar was bought at Uselu Market Benin City. Sawdust and wheat bran were sun dried on daily basis. It was done in such a way that the layer of material was not thicker than 4 cm allowing its proper and even drying. Both materials were mixed regularly during the process.

Substrate preparation

The experiment had two controls. In order to determine if the supplement addition, will affect the mushroom's growth and development. The first control (C1) was just the individual sawdust types measured into the cellophane bag on 300 g oven dry weight basis. The second control (C2) was set up by supplementing each sawdust type with 1% calcium carbonate and 1% sugar.

Two different treatments were prepared. The first treatment (10% wheat bran) was done by supplementing each sawdust with 1% CaCO₃, 1% sugar and 10% wheat bran. The second treatment (20% wheat bran) was supplemented with 1% CaCO₃, 1% sugar and 20% wheat bran. Moistened sawdust (300 g) oven dry weight and 75% water was loaded into a polythene bag (15 x 30 cm). Five replicate bags (15 x 30 cm) were prepared for each treatment. Thereafter, polyvinyl chloride 'neck' (5 wide x 3 cm long) was passed through the filled bag and the mouth of the bag covered with cotton wool. Bags were steamed for three hours on two consecutive days.

Substrate spawning and Incubation

After cooling bags were randomly picked up and inoculated with mushroom spawn at 5% spawn level (Quimio et al., 1990). Spawning bags were incubated for 30 days in a clean room well shaded from direct sunlight and protected from the infiltration of rodents and other pests.

Fruit body induction, harvesting and post harvest handling

Fruit body induction was achieved by reducing the temperature of the environment by spraying water into the room and opening of the mouth of the bag, followed by spraying water on the surface of the substrates. Fruiting bodies were harvested four days after primordial emergence. Fresh weight of each harvest/flush was recorded.

Mushrooms were dried at 105 °C for 48 h, cooled in desiccator and their weight determined.

Analysis of data

This was done by determining the mean and standard error for each parameter. Analysis of variance was calculated for each set of data. The experiment, comprising five types of sawdust and two supplement levels (10 and 20% WB) was replicated five times in a completely randomized design.

RESULTS AND DISCUSSION

L. squarrosulus mycelium was able to colonize the different sawdust types irrespective of the supplementation level. The only exception to this observation was found in the 20% WB sawdust of *C. zenkeri* (Table 1). This exception may have occurred due to an imbalance in the Carbon/Nitrogen ratio required for mycelial growth (Chang, 1978). The fastest mycelial colonization of (7.8 ± 0.49 days) was observed in (C2) *C. zenkeri* sawdust mixture, while it took as long as 17.50 ± 6.50 days for the mushroom to completely colonize the 10% WB sawdust of *C. excelsa* (Table 1). This result suggests *C. zenkeri* sawdust with minimal upgrading (1% CaCO₃ and 1% sugar) is more appropriate for mycelial growth of *L. squarrosulus*. The differences in the treatments as per total time of colonization were significant (p = 0.05).

Primordial emergence was first observed in the pure *B. nigerica* sawdust after 20.60 ± 0.06 days (Table 2) whereas the longest time of primordial emergence was found in *N. papaverifera* 20% WB. The differences in the mean time for first primordial emergence were significant (p = 0.05).

Mean fresh weight of dry fruiting bodies ranged from 16.17g ± 1.25 g in *B. nigerica* (20% WB) to 4.56g ± 1.84 g in *N. papaverifera* (C2). The differences in dry fruiting bodies yield (Table 3) on different sawdust and supplement combinations were significant (p = 0.05).

The highest organic matter loss due to the activity of the mushroom on the different sawdust substrates with their

Table 2. Effect of Substrate Supplementation on the first time of primordial emergence (days).

Substrate mixture	C1	C2	10% WB	20% WB
<i>Chlorophora excelsa</i>	31.00±4.04*	27.30±2.85	34.00±0.00	35.33±2.40
<i>Celtis zenkeri</i>	25.40±0.51	27.40±1.91	30.50±3.18	N.A
<i>Guera cedrata</i>	31.25±0.85	28.20±0.74	39.00±2.00	33.67±0.33
<i>Nesogordenia papaverifera</i>	23.50±2.18	21.67±1.20	34.75±0.75	41.00±1.00
<i>Brachystegia nigerica</i>	20.60±0.16	22.25±1.60	39.00±0.71	42.00±1.00

*= Mean of five replicates ± standard error.

C1 -100% sawdust

C2- 98% sawdust, 1% CaCO₃, 1% sugar.

10% WB-88% sawdust, 10% wheat bran, 1% CaCO₃, 1% sugar.

20% WB-78% sawdust, 20% wheat bran, 1% CaCO₃, 1% sugar.

N.A = Not available.

Table 3. Sporophore yield of *Lentinus squarrosulus* cultivated at different supplementation levels.

Substrate mixture	Supplementation with wheat bran (%)			
	C1	C2	10% WB	20% WB
<i>Chlorophora excelsa</i>	15.93±0.98*	6.72±2.21	12.70±0.00	9.69±1.78
<i>Celtis zenkeri</i>	15.97±2.17	16.02±2.21	11.17±2.97	N.A
<i>Guera cedrata</i>	6.60±2.23	6.36±1.30	12.13±0.00	13.01±1.44
<i>Nesogordenia papaverifera</i>	4.56±1.84	7.16±3.17	14.85±0.65	14.70±0.60
<i>Brachystegia nigerica</i>	12.65±2.42	12.41±2.53	12.56±0.70	16.17±1.25

* = Mean of five replicates in grams.

C1 -100% sawdust

C2- 98% sawdust, 1% CaCO₃, 1% sugar.

10% WB-88% sawdust, 10% wheat bran, 1% CaCO₃, 1% sugar.

20% WB-78% sawdust, 20% wheat bran, 1% CaCO₃, 1% sugar.

N.A = Not available.

Table 4. Percent organic matter loss due to the production of *Lentinus squarrosulus* on sawdust of selected economic trees.

Substrate mixture	Level of wheat supplementation (%)			
	C1	C2	10% WB	20% WB
<i>Chlorophora excelsa</i>	19.93*	33.96	13.88	10.86
<i>Celtis zenkeri</i>	29.23	21.61	25.53	13.07
<i>Guera cedrata</i>	21.51	32.59	33.51	12.66
<i>Nesogordenia papaverifera</i>	28.86	24.99	15.06	15.85
<i>Brachystegia nigerica</i>	25.19	16.33	29.19	15.96

*= Mean of five replicates.

C1 -100% sawdust

C2 - 98% sawdust, 1% CaCO₃, 1% sugar.

10% WB-88% sawdust, 10% wheat bran, 1% CaCO₃, 1% sugar.

20% WB-78% sawdust, 20% wheat bran, 1% CaCO₃, 1% sugar.

N.A = Not available.

their respective supplement ranged between 33.97% in *C. excelsa* (C2) and 12.66% in *G. cedrata* 20% WB mixture (Table 4).

L. squarrosulus (Mont) singer grew successfully with sporophore production on the various sawdust types. Different treatments and controls gave different results. This may be due to effective substrate utilization (Can-

non, 1986; Laborde, 1992) in several substrate mixtures.

The results of this study indicate that even though sawdust is readily available and cheap, there is the need to be guided regarding its sources. This is necessary because sawdust type strongly influences mushroom yield. Also, as shown by Royse et al. (1990), amendment of synthetic substrates may enhance mushroom yield.

Also, an ethno-knowledge obtained from some individuals in sawmills suggests that there is the need to be cautious in the use of *Berlinia* sp. sawdust. It is believed that this tree imparts toxicity to edible mushrooms (personal communication). At least, two individuals claim they had seen persons affected with some ailments eating mushrooms grown on this type of wood. A study on the empirical confirmation of this ethno-knowledge is ongoing and will be reported in the nearest future.

It may be necessary to conduct further research into the use of local sawdusts for cultivation of this mushroom specie so as to increase the yield.

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