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CULTIVATION TECHNOLOGY OF VOVARIELLA VOLAVACEA (BULL. EX. FR.) SINGER FROM NIGERIA.

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Abstract

Volvariella volvacea, a mushroom which is commonly produced on oil palm pericarp waste mainly for subsistence interest, was cultivated using paddy straw and three different levels of sorghun grain spawn. Frult bodies were obtained between 10 and 35 days after spawning. Cumulative fresh mushroom yields of approximately 2.31 kg, 4.15kg and 4.97 kg were obtained for every 15.5kg of dry straw used at 6.0% 9.68% and 12.0% spawn rates, respectively. Approximately 74.0% of the total yiled was obtained during the first flush. There was a significant increase in yield as spawn rate increased. A general decrease in average weigth with increasing age of culture was observed.

INTRODUCTION

Edible wild muschrooms have been regarded by many Nigerians as a delicacy, flavour enhancer, and a health food since earliest times. The degree of deliciousness, however, varies with mushroom species. Mushrooms have been found to be relatively high in minerals (Alofe, 1985 unpublished; Alofe, 1991; Oei, 1991; Alofe et al. 1996). Vitamins (Tolentio, 1981; Oei , 1991; Alofe *et al.*, 1996). Several species of ebible mushrooms have been reported to have a definite effect on blood pressure (Suzuki and Ohshina, 1976; Mori, 1978), tumours and viruses (Oei, 1991). They have been found to be generally low in fat content (1-8%, with an average of 4%) (Oei, 1991; Alofe et al., 1996). Unsaturated fatty acid, of which linoleic acid is the most abundant, make up at least 72% of the total fat contgent in *V. volvacea* and other mushrooms unlike animal fat which is mostly made up of saturated fatty acids that are hazardous to health (Oei, 1991). Mushreooms are therefore, considered as health food. It is no wonder then that certain mushroom species that are indigenous to Nigeria are used in folk medicine in Nigeria (Oso 1972, Alofe, 1996) and others in oriental medicine (Oei. 1991).

Some edible wild mushrooms consumed locally grow in large numbers and are, therefore, readily available for sale. Others such as *V. volvacea* (Bull ex. Fr.) Singer which are very delicious, but produce only small number of mushrooms per flush, can only be

used to serve subsistence purpose. *Volvariella volvacea* naturally remains actively growing under its specific substrate, the oil palm pericarp waste, for many years in form of mycelial mats. Hence this species is usually stimulated to flush year after year on the same location during the rainy season particularly in Yewaland of Ogun State and in some rural areas of Kwara State.

Volvariella volvacea has been cultivated in many European and Asian countries for many years on a variety of substrates which are mostly agricultural wastes (Young and Graham, 1973; Chang, 1974; Bano , 1976; Chang, 1977; chang, 1978; Cheng and Tu, 1978; Chang, 1981; Tolentino, 1981). Nutritional status of mushrooms has also been extensively studied (Alofe, 1985 unpublished: Adewusi *et. al.* 1993). However, V. volvacea has never been reported to be cultivated in Nigeria before this work, though it is highly valued for its nutritional properties by many Nigerians. Characteristically, *V. volvacea* has been found growing naturally on oil palm pericarp waste. It has recently been found growing around the bases of banana plant butts and also on the hewed trunk of an unidentified wooden tree, both of which have reached an advanced stage of decay. The most natural substrate for the growth of this mushroom appears to be the oil palm pericarp waste which is usually deposited around palm oil procesisng pits called "ebu" or "eku" by the Nigerian Yorubas.

The objective of the present work is to describe the indigenous (local) method of *V. volvacea* mushroom production as practiced by some Yoruba farmers, and to design a simple and cheap but conventional method of cultivation that would be within the financial resources of the Nigerian farmers and which will make it possible to produce the mushroom all-year around. The work was also undertaken to determine the effect of spawn density on fresh mushroom yield.

Materials and Methods

V. volvacea mushrooms, commonly called the paddy straw mushroom, was obtained locally around Ile-Ife. Paddy straw was obtained from the International Institute of Tropical Agriculture (IITA), Ibadan. Light-coloured, red variety of sorghum (*Sorghum bicoler* L. Moench) was obtained from a market at Ile-Ife.

Sample Description

V. volvacea is called "Olu-iha" by the Nigerian Yorubas because it is commonly found growing on oil palm bunches, and pericarp waste ("iha") which contains about 15 to 20% of partially de-oiled fibrous portion of the pericarp and mesocarp. The button stage of the mushroom is normally enclosed within a dark-grey membranous tissue called 'universal veil' which gives the young mushroom a greyish-black, egg-shaped appearance. As development advances, the stipe elongates, the veil ruptures just above the crown of the pileus (cap) and the pileus is eventually carried aloft, leaving the remains of the veil, called volva, at the base of the stipe, at the subtrate level. The volva is a charactristic feature of all *Volvariella* species.

The stipe of the mature mushroom is cream-coloured and stout. The pileus is thick and cream-coloured but it has a dark-grey crown. The mature mushroom is fleshy but slightly

fibrous to taste. The mushroom sample used for mycelial isolation through tissue culture technique was obtained from a decaying butt remains of a banana plant in November on the premises of one of the Senior Staff Quarters of the Obafemi Awolowo University, Ile-Ife.

Description of the Indigenous Technology for Stimulating Growth and Fruit Body Formation by *V. volvacea*

Processing of oil palm for extraction of palm oil generally takes place during the rainy season in Nigeria. Extraction of palm oil from boiled and mashed fruits is done within a water-filled, more or less circular pit of about 1 to $1^1/_4$ meters deep and 1 to $1^1/_2$ meters in diameter. When the oil portion of the pericap and mesocarp is deemed to have been completely extracted, the nuts and most of the fibrous portion are separated from the aqueous phase. The aqueous phase is then thrown over a shaded area that had been cleared of twigs and leaf lifter close to the extraction pit. The pericarp waste is allowed to be de-watered for 48 to 72 hours after which the semi-solid waste is covered with partially dry palm leaves to allow adequate ventilation for proper growth, mycelial mat and primordia formation, and fruit body production. All subsequent pericarp wastes are thrown over the palm leaf-covered initial layer. The pericarp waste bed is naturally inoculated by *V. volvacea* spores that are present both in the underlyng soil and air.

Cultivation of *V. volvacea* using the Conventional Method Preparation of Tissue Culture from V. volvacea using the

A healthy, young *V. volvacea* mushroom was broken into two parts and a small amount (2x2mm) of the exposed innner tissue was aseptically removed (Oei, 1991) with a sterile, new razor blade. The tissue was transferred immediately onto the surface of sterile solid potato dextrose agar. Three plates of such tissue isolation were made and the plates were incubated at 30°C until mycelial colonies were 2.5 to 3 cm in diameter.

PREPARATION OF *V. VOLVACEA* SPAWN ON SORGHUM GRAINS

Light-coloured, red variety of sorghum grains *(Sorghum bicolor)* (Oei, 1991) were manually cleaned by winnowing off and hand-picking the unwanted materials. Some of the cleaned grains (150g) were washed, rinsed and soaked in 400ml of tap water for 24 hours. The soaked grains were drained, rinsed with a 400ml portion of tap water and drained to dropwise water level. Gypsum (0.75g) was added to and mixed thoroughly with the grains in a 500ml conical flask to prevent clumping and also to provide calcium ions (Ca^{2+}) for the mycelium culture. The flask was plugged with cotton wool and then covered with aluminium foil, before it was autoclave-sterilized. The cooled sterile grains were inoculated with a piece (2x2mm) of mycelial tissue culture of *V. volvacea*. The flask was incubated at 22 to 25°C with occasional manual shaking to disperse the mycelium-ramified grains. Incubation was continued until the grains were completely covered by mycelium films. Such mycelium -ramified grains are referred to as mushroom seed or spawn.

Preparation of Soil Bed

A central portion (50 x 120cm) of a shaded area that had been cleared of weeds and leaf litter was delimited using a ditch (15 x 20 cm). The ditch was surrounded by another ditch (25 x 30 cm) which was about 25-40cm away from the inner ditch. A cavity (0.5 x 0.4 x 0.3m) was made in the centre of the central portion. The cavity was filled with sand to aid percolation of water and misting. The central portion together with the sand-filled cavity constituted the soil bed.

Preparation of substrate (the straw bed).

Grain-free, paddy straw pieces (50cm long) were tied into bundles, the butts of which were about 15cm in diameter. The straw bundles were completely submerged in boiling tap water in a covered metal drum throughout the sterilization procedure which lasted 4 to 6 hours. After cooling to about 60°C within the covered drum, the straw bundles were squeeze-drained to dropwise water level to obtain a moisture content of about 70 to 75%. The bundles were allowed to cool down to between 30° and 35°C before they were laid side by side on the soil bed until the entire soil bed was completely covered. Calcium carbonate($CaCo_3$) (20g) was sprinkled over the straw layer to provide a slightly acidic condition to prevent growth of *Trichoderma* and bacteria contaminants and to provide calcium ions which would enhance mycelial mat formation as well as fruit body production. Arrangement of straw bundles in layers was repeated until the straw bed was between 50 and 60cm high. The layers were arranged such that the butts of adjacent layers were opposite. Each layer was spawned thoroughly at the rate of either 6.0%, 9.68% or 12%.

Small pieces (2 to 3cm long) of semi-sterile straw were hipped on top of the spawned straw bed such that it was highest (25 to 30cm) at the centre. A transparent polythene sheet was spread over the bed before it was covered with a mosquito net cage which was placed on the strip of soil separating the two ditches. The mushroom culture bed was left in this condition for four days after which the cage and the polythene sheet were removed. The culture bed was heavily watered and then enclosed in a polythene sheet cage (Plate 1) to facilitate adequate misting that would provide a relative humidity of 80 to 90% which is essential for adequate mushroom flushing. The mosquito net cage was replaced and the culture bed was, there after, slightly watered at four-day interval. The procedure was repeated using varied amounts of spawn to determine the effect of spawn density on mushroom yield.

Fully expanded but still fresh fruit bodies were harvested manually by hand-picking (Plate 2) and weighed immediately after harversting. The experiment was done in three replicates. During the course of study, temperature ranged betewem 28° and 30°C.

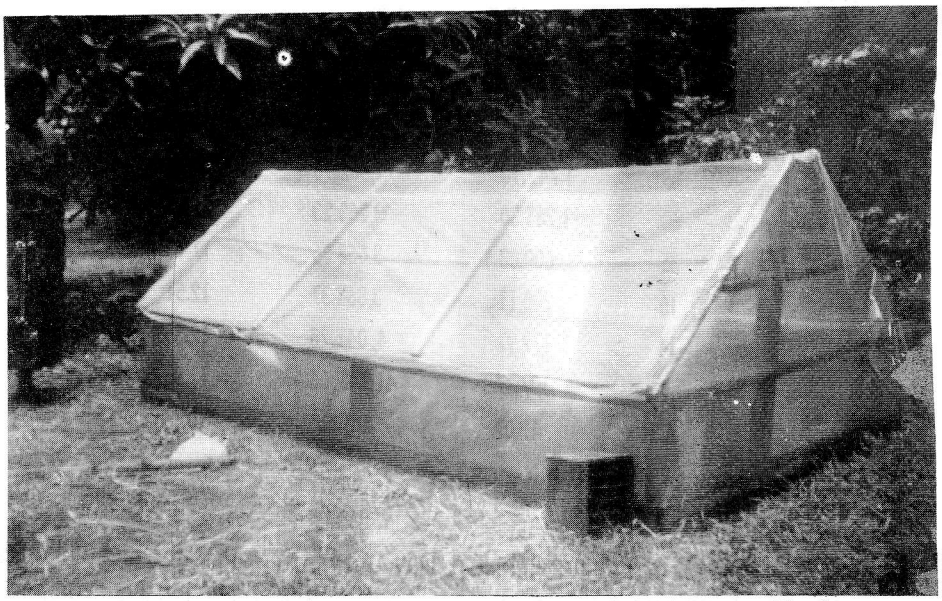

Plate 1. Mushroom house used for experimental mushroom cultivation.

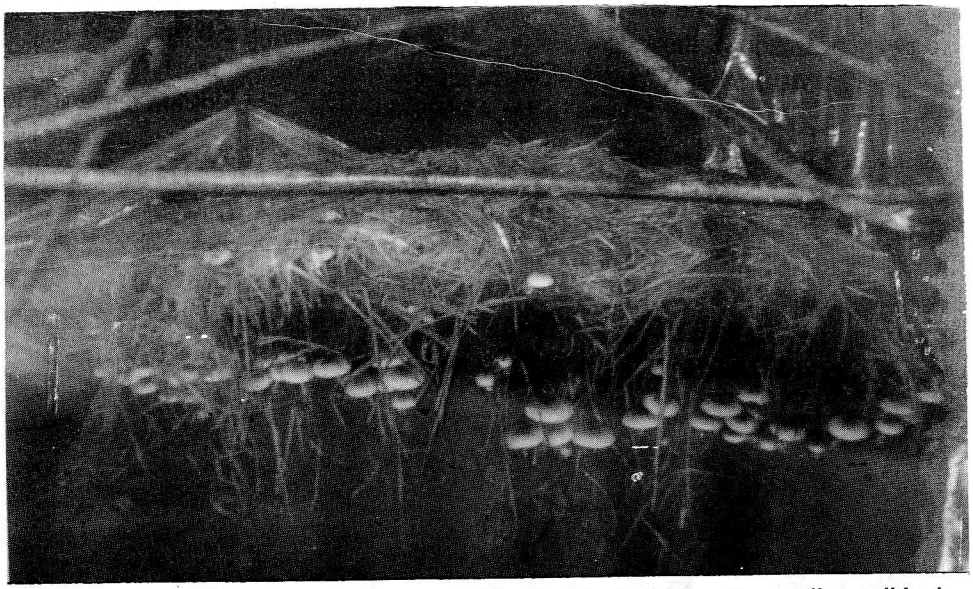

Plate 2. *Volvariella volvacea* fruiting on paddy straw bed and the surrounding soil bed.

Results and Dicussion.

The yield of fruit bodies was obtained between 10 and 35 days after spawning (Table 1, Plate 3). A cumulative yield of 4.15kg/15.5kg dry straw was obtained in 35 days with 9.68% spawning and 26.77% utilization of the substrate. The average weights of the fruit bodies ranged between 11.93g and 21.09g. Average weights of *Agaricus bisporus* strains have been reported to range from 4.57 to 70.00g (Tschierpe, 1981). The largest number of fruit bodies were obtained on "Day 11", which was the second day of the first mushroom flush.

Table 1. Fresh Yield of *Volvariella volvacea* Grown on Paddy Straw Bed
Yield* / 15.5kg dry straw

Time of picking (days)	No. of fruit bodies	Fesh weight (g)	Cumulative weight (g)	Average weight (g)	Subtrate utilization (%).
10	3	60.03	60.03	20.01	0.39
11	85	1,742.90	1,802.93	20.50	11.63
12	60	1,265.30	3,068.23	21.09	19.79
16	35	600.03	3,668.26	17.14	23.67
20	16	200.11	3,868.37	12.51	24.94
27	14	172.91	4,041.28	12.35	26.07
*35	9	107.37	41,148.65	11.93	26.77
LSD 0.05	6.34	12.07	10.11	0.15	———

*** Values are means of three replicates.**

The yield was 42.01% of the total yield. On "Day 12", which marked the end of the first flush, a 30.50% yield was obtained. Most of the fruit bodies obtained during this experiment were produced during the first flush and they constituted 73.69% of the total yield with only 19.709% utilization of the subtrate. It has been reported that the biggest and greatest numbers of fruit bodies were produced during the early stage of mushroom cultivation (Khan *et al.*, 1981; Khanna and Garcha, 1981; Tan, 1981). Each of the remianing four flushes occurred for only one day. A decrease in number of fruit bodies produced per flush, with increasing age of culture, was observed from "Day 12" to the end of the experiment. There was a decrease also in average weight of fruit bodies from "Day 20" to "Day 35" but it was very slight. A decrease in the numbers of fruit bodies with increasing age of culture has been reported (Khan *et al*, 1981; Khanna and Garcha, 1981; Tan, 1981). In 20 days, 93.24% of the total yield was obtained while further increase in yield in the remaining cropping was insignificant.

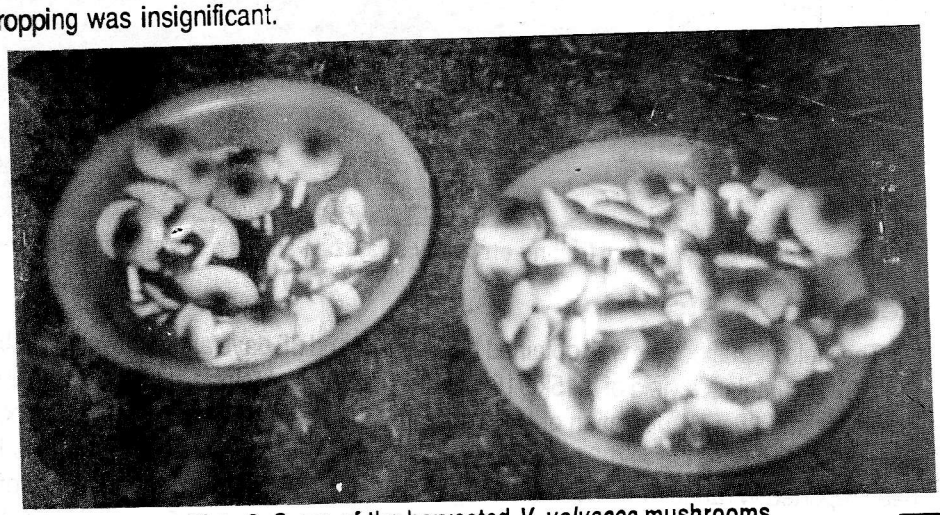

Plate 3. Some of the harvested *V. volvacea* mushrooms

The results obtained on the effect of spawn density on yield of *V. volvacea* are shown in Table 2. There was a significant increase in fresh mushroom yield with increase in spawn rate, with a corresponding increase in the utilization of subtrate. However, the largest number of fruit bodies were obtained on "Day 11" at the different spawn rates used. Only 2.31kg of fresh mushrooms were obtained in 35 days when 6.0% spawn rate was used. When 9.68% spawn rate was used 4.15kg was obtained, while 4.97kg was obtained when 12.0% spawn rate was used. Only 46.50% and 83.51% of the total yield obtained with 12.0% spawn rate were obtained with 6.0% and 9.68% spawn rate, respectively. The results obtained in this work are in agreement with those obtained for the same mushroom species (V. volvacea), (Tolentino, 1981) and other mushroom species (Cheng and Tu, 1978; khan et al, 1981; Khanna and Garcha, 1981).

Table 2: Effect of Spawn Density on the Yield *of Volvariella volvacea

Time of Picking (days)	Spawn rate (%) 6.0			9.68			12.0		
	A	B	C	A	B	C	A	B	C
10	62.34	62.34	0.40	60.03	60.03	0.39	83.11	83.11	0.54
11	967.15	1,029.49	6.64	1,742.90	1,802.93	11.63	2,050.02	2,133.13	13.76
12	756.97	1,786.46	11.53	1,265.30	3,068.23	19.79	1,415.73	3,548.86	22.90
16	363.65	2150.11	13.87	600.03	3,668.26	23.67	710.11	4,258.97	29.41
20	102.70	2, 252.81	14.53	200.11	3,868.37	24.94	460.04	4,719.01	30.45
27	41.85	2,294.66	14.80	172.91	4,041.28	26.07	133.28	4,852.29	31.31
35	15.06	2,309.72	14.90	107.37	4,148.65	26.77	115.29	4.967.58	32.05
LSD 0.05	16.34	72.07	____	9.63	30.14	____	8.83	41.66	____

A: daily yield (g, fresh weight basis)
B: cumulative yield (g, fresh weight basis)
C: substrate utilization (%) per 15.5kg of dry straw.
*** Values are means of three replicates.**

Conclusion

It is apparent from these results that local wood-inhabiting edible wild mushrooms such as *Volvariella volvacea* can be produced commercially throughout the year on agricultural wastes. The technology described in this report is simple and can be easily adopted by the local farmers. The masses will thus have access to the essential nutrients that are present in our edible wild mushrooms but which are only sparingly available during the rainy season.

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