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Effect of Source of Pure Culture of *Volvariella volvacea* (Bull. Fries) Singer on its Growth and Yield

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A B S T R A C T

An experiment was conducted to study the effect of pure cultures of *Volvariella volvacea*, raised from tissue as well as mono and multi spore cultures, on the spawn quality and mushroom yield. It was revealed that spawn prepared from monospore culture sustained significantly higher yield (15.7 % BE) *vis-à-vis* tissue culture (14.6 % BE) and multispore spore cultures (13.0 % BE). The variations in the mushroom yield could be stabilized due to monospore culture. The cultures raised from tissue culture recorded highest radial growth in petriplates and maximum downward linear mycelial growth in spawn bottle(9.87 mm/day; 11.67 mm/day) as compared to monospore culture(9.38mm/day; 11.57 mm/day)) and multi-spore culture(8.65mm/day;11.33 mm/day). There was not much difference in the days taken for primordial initiation (9-10 days) and first harvest (14-15 days) irrespective of source of cultures. The average weight of sporophore was highest (12.8 g) in response to spawn raised from multispore cultures.

Introduction

Healthy and productive spawn plays a crucial role in successful cultivation in achieving high yield and quality mushroom crop. The yield potential of spawn not only depends on the selected strain but also on the techniques of its preparation. Odisha is the leading producer of straw mushroom in the country with an estimated annual mushroom production of 12364 MT (Anonymous, 2017-2018) and this has been due to the large number (265 numbers) of commercial spawn production units operating in the state. However, the quality of spawn of straw mushroom (*Volvariella volvacea*) has deteriorated over the years. It has been noticed that wide variations exist among spawn in terms of

viability, yield potential and some essential quality attributes. Large scale production due to high market demand as well as lack of adequate research to improve upon the quality parameters have probably compromised the spawn quality.

Most of the research work has been devoted to standardize the method of spawn production of different edible mushroom species. It is revealed that not much work has been done on quality improvement in spawn of straw mushroom excepting few. In Odisha, Pani and Das (1999) reported the performance of tissue culture taken from different stages and regions of sporophore of *V. volvacea*. Pani *et al.*, *(*2017) have done some pioneer work on improving the strainal purity of tissue cultures *vis-a-vis* the commonly practiced tissue culture techniques followed by the spawn production units to improve the spawn quality of straw mushroom. Kligman (1943) reported rejuvenation of strains by adopting tissue culture from cultivated crop of sporophores. Strainal purity in mushroom by tissue culture has also been suggested (Dhar *et al.*, 2002).

In Odisha and elsewhere in the country, the growers are often confronted with low and erratic yield of straw mushroom, even though they use good quality straw and other inputs and employ best of management practices in the most conducive climatic conditions. A thorough investigation revealed that the lack of good quality of spawn as influenced by various production parameters and inherent genetic characters has been the most limiting factor for the successful cultivation of straw mushroom. Kalra and Phutela (1991) have reported considerable variations in the biological efficiency and mycelia characters of straw mushroom. Therefore, the present study was undertaken to raise pure culture of straw mushroom (*Volvariella volvacea*) by tissue culture, multispore culture and single spore culture and use such cultures separately for spawn preparation with a view to evaluating them for improving the growth and yield.

Materials and Methods

A high yielding strain of straw mushroom (*Volvariella volvaceae*) of Odisha, namely OSM-11 maintained at Centre of Tropical Mushroom Research and Training, Department of Plant Pathology, Odisha University of Agriculture and Technology, Bhubaneswar was used in this study. The test fungus was sub-cultured at periodic interval of 14 days on PDA medium and 10 days old pure culture was used for physiological studies as well as spawn production.

Effect of method of raising pure culture on spawn quality

The method of raising pure culture plays an important role in deciding the quality of spawn. Tissue cultures and spore cultures have often been attempted for rejuvenation of mushroom culture (Kligman, 1943) which leads to stable and higher production. Keeping this in view, the present study was undertaken to raise pure culture of *V. volvacea* by tissue culture and spore culture (multispore and monospore) and study their effects on yield and yield attributing characters of straw mushroom spawn.

Preparation of tissue culture

For preparation of tissue culture, a fruiting body of straw mushroom at egg stage was selected from first flush from a bed with maximum fruiting and it was the largest from the lot. It was then surface sterilized by 70 % ethyl alcohol followed by rinsing 2-3 times in sterile water. With the help of a pre-sterilized knife, the fruiting body was cut longitudinally in to two equal halves and small pieces of tissues from the joint of stalk and pileus were transferred separately in to sterilized PDA

slants. From the single egg stage, 5 pieces of tissues were separately transferred to 5 slants. Selection of pure culture from tissue culture of was done as per the method suggested by Pani *et al.,* (2017).

Spore culture

Preparation of multispore culture

Under aseptic conditions, spores mass was scrapped from a spore print and suspended in 100 ml sterilized distilled water in flask and shaked to obtain uniform spore suspension. A few drops of this suspension was added to lukewarm culture medium and poured into oven sterilized petriplates. Petriplates were rotated to homogenize the spore suspension into culture medium. The culture medium was allowed to solidify and then petriplates were incubated at 35°C for 3 to 4 days. The spore germination was observed under microscope and germinating spores were transfer carefully to culture tubes along with a piece of agar containing a culture medium recommended for mushroom species isolated. The culture tubes were then incubated at 30°C for 7 to 10 days (Upadhyay, 2011). All the operations were carried out under strict aseptic conditions.

Preparation of monospore culture

Single spore or monospore cultures were prepared in same way as that in multispore cultures. Nevertheless, for monospore culture isolation, the spore suspension was serially diluted to obtain 10 to 12 spores/petriplate. Individual germinating spore was marked and lifted under aseptic conditions, transferred to culture tubes and incubated 25°C for 10 to 14 days. The productive among the monospore cultures was used in the evaluation process.

The radial mycelia growth (mm) of *V.volvacea* raised from tissue, multi and single spore cultures was determined from 5 to 9 days at 2 days intervals. The colony characteristics like colour of the colony and its morphology were also noted.

Spawn were prepared from tissue and spore cultures as per standard techniques (Pani *et al.,* 2017).Then the downward linear growth of the edible fungus in the spawn bottle was measured from 6 to 12 days at 3 days interval, so as to assess its growth rate(mm/day).Such spawn were used separately for mushroom cultivation as per the method suggested pani and Das(2001) to determine their relative yield potential. Mushrooms were harvested at egg stage from two flushes and fresh weight was immediately recorded. The days for primordial initiation and harvest as well as number and average weight of sporophores were noted. Data pertaining to yield were statistically analyzed.

Results and Discussion

A perusal of data presented in Table 1 revealed that the pure culture raised from tissue culture was cottony white, aerial thin and un-uniform in their colony characteristics. These cultures were also relatively fast growing as evident by their radial mycelia growth in petriplates and downward linear growth in spawn bottles as compared to other sources of pure cultures. The cultures raised by monospore culture were less aerial but having uniform growth characteristics but were slower in growth rate than tissue culture, but higher growth rate than multispore culture (Table 1).

The yield and yield attributing characters of spawn raised from tissue culture, multispore culture and monospore culture were presented in Table 2 and depicted in Figure 2. It was revealed that spawn quality of straw mushroom varied as per the source from which it was raised.

This finding can be co-related with the findings of Kalra and Phutela (1991) who have reported considerable variations in the biological efficiency and mycelia characters of straw mushroom. Significantly higher yield (1105.5 g) as well as highest biological efficiency (15.7 %) was recorded in the spawn raised by monospore culture compared to other sources Moreover, the biological efficiency of *V. volvacea* could be stabilized due to spawn raised from monospore culture. This finding corroborates earlier reports of Mandeep Kaur *et al.*, (2016).

Table.1 Effect of method of raising pure culture of *V.volvacea* on mycelia growth and colony characteristics

Method	Radial mycelia growth(mm)				Colony characteristics		Downward linear growth in spawn(mm)			
	5th day	7th day	9th day	Growth rate (mm/day)	Colour	Morphology	6th day	9th day	12th day	Growth rate (mm/day)
Tissue culture	40.2	74.5	88.9	9.87	Cottony white	Arial thin, less uniform growth	47.8	95.6	140.1	11.67
Multi-spore culture	31.6	65.6	77.9	8.65	Cottony white	Aerial thin, less uniform, little compact growth	42.3	87.8	136.6	11.33
Mono-spore culture	30.9	71.2	84.5	9.38	Cottony white	Less aerial, uniform growth	44.5	93.4	138.9	11.57

Each observation was the average of seven replications

Table.2 Effect of method of raising pure culture of *V. volvacea* on mushroom yield and yield attributing characters

Method	Primordial initiation (days)	1st harvest (days)	Sphorophore		Avg weight of sporophores (g)	Biological Efficiency (%)
			No.	Weight (g)		
Tissue culture	9	14	75.7	1022.0	12.1	14.6
Multispore culture	10	15	69.4	910.0	12.8	13.0
Monospore culture	9	14	95.6	1105.5	11.5	15.7
SE(m)				13.74		
CD(0.05)				42.82		

Each observation was the average of seven replications

Fig.1 Fruiting *V. volvacea* in response to spawn raised from different sources

A. Tissue culture

B. Multispore culture

C. Monospore culture

Spawn based on tissue culture sustained yield (14.6 % BE) which was statistically higher from that obtained with multispore culture (13.0% BE), but significantly lower compared to the yield recorded from monospore culture. This could have been due to the isolation of a single fertile spore giving rise to highly productive self-fertile progeny. As reported by Chang and Yau (1971), the basidiospores borne on a single basidium in straw mushroom hymenium were in the ratio of 3 sterile : 1 fertile spore. This indicated that the self sterile progeny were present in abundance in tissue and multispore cultures which might have been the possible reason for the comparatively lower yield of mushroom from such cultures.However, there was not much difference in primordial initiation (9 to10 days) and days for first harvest (14 to 15 days). The average weight of sporophore was highest in response to spawn raised from monospore cultures.

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