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A Modified Approach in Substrate Preparation Technique for Small Scale Oyster Mushroom Farming

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Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

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Short Communication

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ABSTRACT

Aims: Cultivation of oyster mushrooms has increased vastly in a global scale during last few decades. Contaminants and indigenous microflora present in the substrate may led to the low productivity of mushrooms. Keeping this in mind, the present study was under-taken with slight modification in substrate preparation technique to eliminate dust particles from the substrate and to assure contamination free mushroom production.
Study Design: Comparative evaluation between modified approach and control beds.
Place and Duration: The study was carried out in Shri Laxman Prasad Pyare Lal Agro Products, Ayodhya during 2017.
Methodology: The wheat straw substrate was immersed in the drum containing tap water, mixing properly and allowed to stand for 10-15 min to settle down the dust particle in the bottom of the drum. After that, the floating substrate was transferred to the slant surface so that the extra water was decanted off. This substrate was put in to the steam sterilised gunny bag and steeped in the chemical solution consisting of Formaldehyde (500 ppm) and Bovistin (75 ppm) for 18 h. For the

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control, fresh wheat straw was put in to the steam sterilised gunny bag and steeped directly in the chemical solution as suggested above. After removal of excess water, the substrate was used for spawning with inoculums of *Pleurotus sajor-caju*, Strain- Malaysia.
Results: Beds with modified approach showed a complete absence of contaminants during entire crop cycle which showed the effectiveness of modified method while beds treated as control showed little incidence of various contaminants viz., *Alternaria alternata, Aspergillus fumigatus*, *Curvularia* sp., *Penicillium* sp., and *Rhizopus stolonifer* with 6.67-20.00 percent incidence.
Conclusion: The results revealed that modified method should be considered to contamination free mushroom production.

Keywords: Contaminants; mushroom; substrate.

1. INTRODUCTION

Oyster mushroom (*Pleurotus sajor-caju*) can be grown on a wide range of agricultural waste such as straw, saw dust, rice hull etc. due to its strong enzymatic features [1]. These substrates are accompanying with indigenous microflora. The antagonistic interaction between these microflora and desired fungus contributed to the low productivity of mushroom [2,3]. Therefore, proper substrate disinfection is pre requisite to eliminate weed and obtain good yield. Substrate disinfection can be achieved by various means such as using material boiled with hot water [4], steaming [5], autoclaving [6], radiation and solarisation [7], composting [8] and chemical pasteurisation [9] etc. Among these methods, the chemical pasteurisation is the most popular method for small scale growers due to its easiness, effectiveness and moreover minimising time, labour and expenditure on appliances and fire wood. In this method, substrates are steeped in chemical solutions like 1% formaldehyde [10], 0.2% Bavistin [11], 0.5% commercial Lime [12] and 0.01% Derosal [4] or their combinations viz., 500 ppm Formaldehyde +75 ppm Bovistin [9,13] for different time intervals. After removal of excess water, the substrate becomes ready for inoculation. Several researchers have been reported in their work regarding the appearance of contamination from chemically pasteurised substrates during the production span [9,13]. The present study was under-taken with slight modification in substrate preparation technique to eliminate dust particles from the substrate and to assure contamination free mushroom production.

2. EXPERIMENTAL DETAILS

2.1 Mushroom Culture

The pure culture of *Pleurotus sajor-caju* Strain-Malaysia was obtained from the Mushroom Section of Plant Pathology Department, Chandra Shekhar Azad University of Agriculture and Technology, Kanpur (U.P.) India. It was maintained on Potato Dextrose Agar medium (peeled, sliced and boiled potato, 200g; dextrose, 20g; agar, 20gl^{-1}) by using serial subculture method [14].

2.2 Spawn Strategy

2.2.1 Spawn substrate

Wheat grain (*Triticum aestivum*) was used as a spawn substrate which was purchased from the seed market of Faizabad. The spawn was prepared by the conventional method [14].

2.2.2 Spawn dose

Inoculation of substrate was made with spawn of *P. sajor-caju* Strain-Malaysia at 15% w/w on dry weight basis under aseptic conditions.

2.3 Substrate for Mushroom Cultivation

2.3.1 Substrate

Wheat straw was used as a substrate due to its easy availability in this area. It was collected at the time of threshing of harvested crop from village Madarahia.

2.3.2 Substrate preparation (Pasteurisation)

The wheat straw was immersed in the drum containing tap water and mixing properly and allowed to stand for 10-15 min to settle down the dust particle in the bottom of drum. After that, the floating substrate was transferred to the slant surface so that the extra water was decanted off. This substrate was put in to the steam sterilised gunny bag and steeped in the chemical solution consisting of Formaldehyde (500 ppm) and Bovistin (75 ppm) for 18 h as suggested by Vijay and Sohi [9]. For the control, fresh wheat straw was put in to the steam sterilised gunny bag and

steeped directly in the chemical solution as suggested above. After removal of excess water, the substrate was used for spawning. A total of 30 bags (15 each for treatment and control) were prepared with 4.5 kg wet weight (=1.5 kg dry weight) of the substrate in each replica.

2.4 Method of Cultivation

Plastic bag technology was employed in this experiment. The beds were prepared from pasteurised substrate by multilayered spawning following the procedure adopted by Bano [15]. These were incubated in cultivation room at 22-30°C temperature for spawn run. When mycelia had completely covered the beds, the polythene covering were turned off and relative humidity was maintained 85-95% with the help of humidifier.

2.5 Concerning Data

The growth and development of mushrooms were monitored daily. Data on days require for mycelia colonisation, initiation and maturation of fruit bodies, yield and biological efficiency were recorded. The biological efficiency (%) was calculated as follows:

$$\%BE = FWm/DWs * 100\%;$$

Where, BE is Biological Efficiency (%); FWm is total fresh weight (g) of mushroom yield across all flushes, and DWs is substrate dry weight (g).

Although the yield data of mushroom are given in present communication, the observation regarding encountered microorganism was prime concern to evaluate the efficiency of the modified method to minimise the appearance of weed. Daily inspection was made to trace any growth of unwanted microflora on the beds till harvesting of the mushrooms. The incidence of competitor microorganism were recorded as the number of infected beds with particular microorganism out of the total beds per treatment and expressed in percentage. The competitors encountered on the mushroom beds were examined under a compound microscope and mycological features like cultural character, hyphal characteristics, septations, branching pattern of hyphae, differentiation and development of sporophore, measurements etc. were studied and compared with the relevant literature for identification of the microflora isolated from contaminated mushroom beds.

$$\text{Incidence of contamination} = \frac{\text{number of infected beds with particular microorganism}}{\text{Total number of beds per treatment}} \text{ X } 100$$

2.6 Statistical Analysis

Completely randomized design (CRD) was followed for the experiment. The yield data was statistically analysed by using the analysis of variance (Anova) at P=5%. The critical difference (CD) was worked out at five per cent probability level.

3. RESULTS AND DISCUSSION

Beds with modified approach showed a complete absence of contaminants during the entire crop cycle which shows the effectiveness of the modified method (Fig. 1) while control showed little incidence of contaminants during spawn run period. *Aspergillus fumigatus* was predominant mycoflora appearing on the control beds with incidence of 20%. The incidence of other contaminants viz., *Penicillium* sp*., Rhizopus* sp., *Alternaria alternata* and *Curvularia* sp., were in the range of 6.67 to 13.34% (Fig. 2). Later, the growth of these mycoflora was suppressed by mushroom mycelia. It might be due to combined effect of both higher proportion of inoculums of *Pleurotus* against contaminants and effectiveness of pasteurisation practice. It is well established when contaminants are scarce in the substrate they do not offer a competence for the mycelium of *Pleurotus* sp which quickly colonised straw substrate [16]. During the course of study, five fungal species viz., *Aspergillus fumigatus*, *Alternaria alternata, Curvularia* sp., *Penicillium* sp., and *Rhizopus stolonifer* were encountered on control. The yield data shows that there was no significant difference among treatments which reflects no adverse effect of these contaminants on mushroom yield (Table 1). It might be due to the suppression of growth of competitors by mushroom mycelia. The susceptibility of control against the contaminants was possibly due to the presence of dust particles in the wheat straw substrate that provides inoculum for the development of competitors [17]. According to the Chhetri et al. [18] contaminants like *Aspergillus* sp. is the major contaminant of mushroom beds which could cover up the beds very rapidly and restricted the mycelia run in the pasteurised straw and caused highest yield loss. Although, the appearance of lesser incidence of competitors in our investigation revealed the efficiency of chemical treatment.

Fig. 1. Beds with modified approach showing complete absence of contaminants

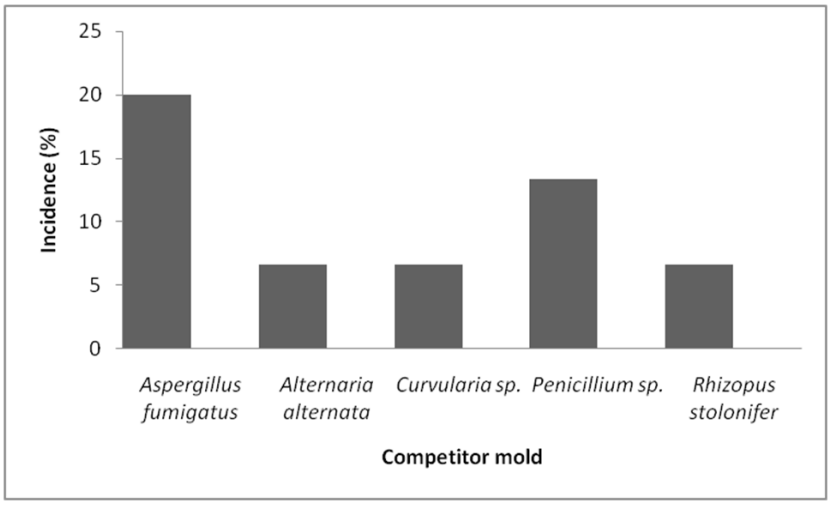

Fig. 2. Prevalence of competitor moulds in control beds

Table 1. Effect of modified treatment on various parameters of *Pleurotus sajor-caju*

Treatment	Spawn run (Days)	Fruit body initiation (Days)	First harvest (Days)	Total yield from three flushes (gm/4.5kg wet substrate)	Biological efficiency (%)
Modified treatment	14.8	20.9	23.2	1404.8	91.96
Control	15.2	21.3	24.8	1379.4	93.65
SE	**-**	**-**	**-**	**34.14**	**2.27**
CD (P=0.05)	**-**	**-**	**-**	**69.98**	**4.67**

Average of fifteen replications

4. CONCLUSION

The *Pleurotus* sp. can be cultivated on a wide range of agro-industrial wastes which are attacked by several competitors of fungal and bacterial origin. The antagonistic relationship between these microflora and desired fungus contributes a low productivity of mushrooms. In severe cases, complete failure of the mushroom crop may take place. Despite of no significant difference in yield, modified method to eliminate dust particles from the substrate should be considered to contamination free mushroom production.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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