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Akansha Deora

Department of Plant Pathology, Rajasthan College of Agriculture, Maharana Pratap University of Agriculture and Technology, Udaipur, Rajasthan, India

Dr. SS Sharma

Department of Plant Pathology, Rajasthan College of Agriculture, Maharana Pratap University of Agriculture and Technology, Udaipur, Rajasthan, India

Abhitej Singh Shekhawat

Department of Soil Science and Agricultural Chemistry, Rajasthan College of Agriculture, Maharana Pratap University of Agriculture and Technology, Udaipur, Rajasthan, India

Suresh Kumar

Department of Plant Pathology, Rajasthan College of Agriculture, Maharana Pratap University of Agriculture and Technology, Udaipur, Rajasthan, India

Kalpana Yadav

Department of Plant Pathology, Rajasthan College of Agriculture, Maharana Pratap University of Agriculture and Technology, Udaipur, Rajasthan, India

Poonam Yadav

Department of Plant Pathology, Rajasthan College of Agriculture, Maharana Pratap University of Agriculture and Technology, Udaipur, Rajasthan, India

Corresponding Author Akansha Deora

Department of Plant Pathology, Rajasthan College of Agriculture, Maharana Pratap University of Agriculture and Technology, Udaipur, Rajasthan, India

Evaluation of the best time for spawning of *Pleurotus eryngii* mushroom to standardize the cultivation technology in Rajasthan, India

Akansha Deora, Dr. SS Sharma, Abhitej Singh Shekhawat, Suresh Kumar, Kalpana Yadav and Poonam Yadav

Abstract

The aim behind this experiment was to know the suitability of months that gives better Biological efficiency and its sustainability with respect to yield. This research work was accomplished at the project of AICRP on medicinal mushroom unit in Department of Plant Pathology, Rajasthan College of Agriculture, Udaipur during the months of November to April in the natural climatic conditions, where the temperature and the relative humidity of cropping room ranged between 9-24 °C and 60-80%, respectively. This work tells you about the best time for spawning to standardize the cultivation technology of *Pleurotus eryngii* in Zone IV a (Udaipur region) as well as in Rajasthan. Out of four different months (November, December, January and February) tested for spawning, the fastest mycelial run and pinhead initiation occurred in December and January months when the average temperature mean of maximum and minimum remains well under 20 ± 1 °C as per meteorological data whereas, maximum B.E.% was observed in the month of December followed by November. Hence, it clearly indicates that the best suited time for cultivating King Oyster (*Pleurotus eryngii*) Mushroom is during winter months of the year.

Keywords: Biological efficiency, king oyster mushroom, mycelial run, pinhead initiation, spawning

Introduction

The word "Mushroom" has been used in different forms at different times and in different nations. Commonly, mushroom refers to all macrofungi which possess stalks and caps or all the large and fleshy fungi (Chang and Miles 1992)^[1]. They are rich in protein, dietary fibre, vitamins and minerals (Alam *et al.* 2008, Sabaratnam *et al.* 2011, Singh 2017)^[2, 9, 11]. In India, due to suitable weather conditions and abundance of cheaper agro residues like straw, leaves and stalks of different field crops, mushroom cultivation has been adopted by progressive farmers, which not only benefitted them with an additional source of income but they are also recycling the farm products.

Since water resources are not satisfactory in Rajasthan and agriculture is mostly rainfed that is why mushroom cultivation has great potential in Rajasthan as this industry is labour intensive & requires less water and land as compared to commercial farming. Currently in Rajasthan, the subtropical and tropical varieties of mushroom like oyster and milky mushroom are rising and gaining popularity. Button mushroom is also being grown but due to unavailability of temperate climate throughout the year, it is being grown during very limited period of the year *i.e.* winter season only (Kumar 2005)^[4].

Oyster mushroom is classified under Phylum –*Basidiomycota* and belongs to Family -*Pleurotaceae. Pleurotus eryngii* (DC. ex FR.) Quel. is the largest species in the Oyster mushroom genus, *Pleurotus*, which also contains *P. ostreatus*, *P. sajor-caju* and *P. florida*. It has a thick, white and meaty stem or stalk called stipe, a large tan cap called pileus and gills are present underside of the pileus. *P. eryngii* is also called as Kabul Dhingri and King Oyster Mushroom (Moonmoon *et al.* 2010, Ryu *et al.* 2015) ^[6, 8]. Unlike other mushrooms species, this genus shows much diversity in their adoption to the varying agro climatic conditions (Zadrazil and Dube 1992) ^[13]. Different species of *Pleurotus* can be grown all around the year at various temperature ranges. In subtropical climatic conditions, some species of *Pleurotus* are suitable like *P. citirinopileatus*, *P. sajor-caju*, *P. florida* and *P. salmoneostramineus* (Pink oyster) that commonly require comparatively low temperatures making them suitable to cultivate during September to March months of the year in the state. The suitability of *P. eryngii* is found to be in winter months of the year. However, not much work had been done to standardize its cultivation technology in existing weather conditions; the study is aimed to evaluate the optimum parameters for getting higher biological efficiency.

Materials and Methods

Mother Culture: Pure cultures were procured from Directorate of Mushroom Research, Chambaghat, Solan, Mushroom Research and Training Centre, Haryana Agricultural University, Murthal, Haryana and Mushroom Research Laboratory, Indian Institute of Horticultural Research, Hesarghatta, Bengluru. Cultures were multiplied on malt extract agar and maintained in test tubes. We prepared six combinations from these three cultures through mycelial anastomosis. Out of them, one strain was found to be the most suitable under sub-tropical climate, which we later named as "Pratap King Oyster-1" (P.K.O-1) and this strain has been used in our experiment.

Preparation of master culture: Healthy wheat grains were soaked in water overnight and were boiled up to semi cooked conditions in the next morning. Excess water was drained off and seeds were left for surface drying. These seeds were then mixed with 1% CaCO₃ and were filled in milk glass bottles or polypropylene bags which we later sterilized at 20 lbs psi pressure (126.5 °C) for 2 hrs. After cooling, each bag was inoculated aseptically with two mycelial agar bits of P.K.O.-1 culture in Laminar Air Flow and incubated at $20\pm1^{\circ}$ C in incubator for 20-25 days till the grains were fully impregnated with the mycelium of inoculated mushroom culture (Monmoon *et al.* 2010)^[6] (Fig 1).

Preparation of substrate: The experiment was laid out by using straw of wheat crop, since; it is easily available and cheap. Straw was chopped into 3-5 cm pieces and chemically treated by soaking in mixture of Bavistin (8g), Formalin (300ml) and Nuvan (30ml) with water in drum of 200 litre

capacity and kept overnight. On the next day, after decanting excess water, it was used for spawning purpose (Peng 1996) ^[7] (Fig 2).

Spawning: The experiment was performed for the four winter months *viz.* November, December, January and February. For layer spawning, substrate filled in bag was pressed to a depth of 2-3 inches and broadcasted with spawn above it. Similarly, 2^{nd} and 3^{rd} layers of entire substrate were put simultaneously and after spawning, some amount of substrate is put over it, and then the bags tied up with rubber bands (Fig 3). Thus, five bags in each month were spawned and they were maintained under optimum conditions of mushroom growth. The date of spawning was also kept same in every month. Thus, four treatments with five replications of each were placed using Completely Randomized Design to reach the conclusion so that a consolidated recommendation can be made as to how the King Oyster can be grown with maximum BE.

Crop management and harvesting: After 20-22 days of spawning, when spawn run gets completed and bags turns white in colour due to white mycelial run, the polythene covers were teared off. Pin heads came out after 2 to 3 days of removing the poly covers which later developed into mature fruit bodies (Kirbag and Akyuz 2008)^[3] (Fig 4). At this stage, mushrooms were plucked off by twisting in clockwise direction at the substrate level, counted and weighed separately for each replication at every harvest. Yield was recorded and the Biological efficiency was also calculated.

Biological efficiency: The Biological efficiency was also calculated by following the given formula of Chang and Miles (1992)^[1].

B. E. $\% = \frac{\text{Weight of fresh fruiting bodies (g)}}{\text{Dry weight of substrate (g)}} \times 100$

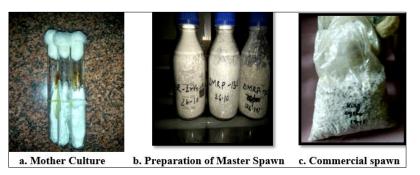


Fig 1: Steps for spawn preparation of *Pleurotus eryngii* from mother culture



Fig 2: Steps for substrate preparation \sim 1407 \sim



Fig 3: Steps for substrate inoculation or spawning

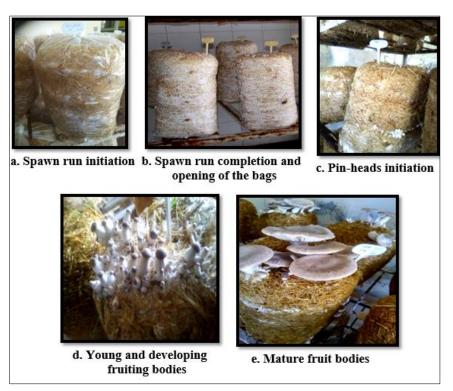


Fig 4: Photographs of different growth stages of *Pleurotus eryngii* from spawn run to mature fruit bodies development

Statistical analysis: Statistical Analysis Software (SAS software) developed by Indian Statistical Research Institute (ISRI), New Delhi was used for performing statistical analyses of data. The critical differences among treatment groups were determined using the ANOVA test at level of significance (p= 0.05). All the experiments were carried out in five replications in Completely Randomized Design (CRD).

The data analysis for this paper was generated using SAS software. Copyright [2014] SAS Institute Inc. SAS and all other SAS Institute Inc. product or service names are registered trademarks or trademarks of SAS Institute Inc., Cary, NC, USA.

Results and Discussion

Cultivation of *Pleurotus* mushroom is gaining popularity among small and marginal growers because of its simplicity of growing on wheat or paddy straw over other agro wastes. The cultivation of edible mushroom not only helps in recycling of agro wastes but also helps in filling up the protein gap prevalent among large population of the country. Data in the Table 1 indicates that, among the different months tested for the efficacy of *P. eryngii* production, the mycelial run was observed to be the fastest in the month of December (19 days) and January (19.4 days). Months of November and February gave same results (22.4 days) and the same trend was followed for the pinhead initiation time. This trend (Fig 5) exhibited due the fact that the temperature started declining slowly with the onset of November and reached minimum range at the end of November and continued to be in the same minimum range till mid January and further, started somewhat increasing at the end of January. So, the bags spawned on 1st December and 1st January gave significantly faster growth in comparison to the bags spawned on 1st November and 1st February. Whereas, the highest B.E. % (76.7%) and maximum number of fruit bodies (30.6) were obtained in the month of December (Table 2). November gave significantly higher BE% (72.1%) than January (65.5%) and February (56.8%). The fluctuation in this trend (Fig 6) in case of BE% is because of the higher temperature after mid January and lower temperature after mid November when the fruiting bodies of January and November months started developing, respectively. Average fruit body weight and fruit body measurements were highest in the months of January (93.5g) and February (93.1g) as shown in Table 3, Fig 7 and Fig 8. The experiment was statistically significant. Being the

temperate in nature, results shown by mushroom were accordingly perfect. According to the worldwide survey, different kinds of wastes have been proven to be useful for Oyster Mushroom growing in particular months. Around 20 °C and 20±1 °C, respectively are best suited temperatures for Pleurotus eryngii (Theochari et al. 2002, Kumar 2005)^[12, 4]. According to the findings of Kumar and Sarathi (2017)^[5], a minimum temperature of (13.4-24.2 °C) was found most appropriate for the cultivation of oyster mushroom. Saha (2017) ^[10] suggested winter season to be promising for obtaining maximum yields and B.E.s of different strains of Pleurotus species. This confirms our findings that the mycelial run and pin head initiation in our case was fastest in the month of December (19 days) and January (19.4 days), when the average temperature mean of maximum and minimum remains well under 20±1 °C as per meteorological data. Whereas, the highest B.E. % (76.7%) and maximum number of fruiting bodies (30.6) were obtained in the month of December.

According to the results of our study, it clearly indicates that the best suited time for cultivating King Oyster (*Pleurotus eryngii*) Mushroom is during winter months of the year starting from October to February and if temperature remains suitable, it can be up to March also.

Table 1: Effect of spawning in different months on spawn run and
pinhead initiation of Pleurotus eryngii

Treatment	Days taken in spawn run	Days taken in Pin head initiation
November	22.4	28.0
December	19.0	25.0
January	19.4	26.0
February	22.4	28.0
SEm±	0.3	0.4
CD(p=0.05)	0.9	1.1

S.Em \pm : Standard error of the mean. CD (p = 0.05): Critical difference, where p = level of significance. All the observations are average of five replications.

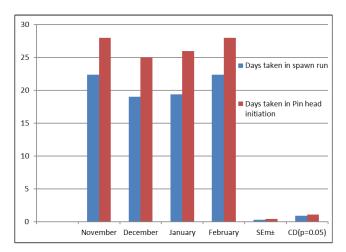


Fig 5: Average number of days taken in spawn run completion and pin head initiation in different months

Table 2: Effect of spawning in different months on yield and BE% of Pleurotus eryngii

Treatment	Number of fruiting bodies	Yield (g)	Biological efficiency %
November	17.0	720.8	72.1
December	30.6	767.4	76.7
January	8.2	655.2	65.5
February	8.8	568.2	56.8
S.Em±	2.0	33.2	3.3
CD(p=0.05)	6.0	99.4	9.9

S.Em \pm : Standard error of the mean. CD(p = 0.05): Critical difference, where p = level of significance. All the observations are average of five replications.

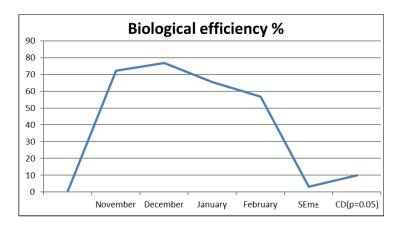


Fig 6: BE% obtained from different months.

The state of the second	Average fruit body	Fruit Body Measurement		
Treatment	weight (g)	Pileus (cm)	Stipe length (cm)	Stipe diameter (cm)
November	45.1	7.9	5.0	3.0
December	26.3	6.5	4.6	2.0
January	93.5	10.0	5.7	4.5
February	93.1	9.0	5.5	4.0
S.Em±	16.3	0.1	0.1	0.1
CD(p=0.05)	48.8	0.4	0.4	0.3

Table 3: Effect of spawning in different months on morphology of *Pleurotus eryngii*.

S.Em \pm : Standard error of the mean. CD(p = 0.05): Critical difference, where p = level of significance. All the observations are average of five replications.

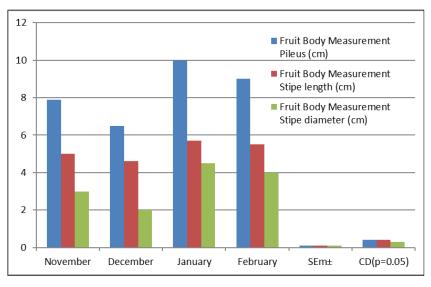


Fig 7: Effect of spawning in different months on morphology of Pleurotus eryngii.

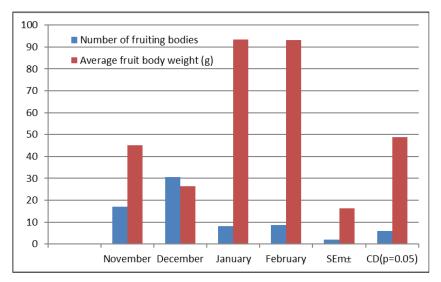


Fig 8: Number of fruiting bodies and average weight of Pleurotus eryngii in different months.

Disclaimer

All photographs are original and copy right of the author.

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