Original Research Article

Assessment of grain extract media on mycelial growth of *Pleurotus* spp. (*P. sapidus* and *P. flabellatus*)

Abstract

The aim of present investigation pertains to study the impact of different culture media on the mycelial growth and dry weight of *Pleurotus* spp. Grains generally contains high nutritional content thereby decreases the harvesting period of mycelium and enhances dry weight. The study was conducted at Mushroom Laboratory Department Plant Pathology, S. V. P. University of Agriculture and Technology, Meerut, UP, India. Different crop grains were used as source of nutrition for fungus and added in PDA (media) in separate amount. Experiment was conducted in seven treatments with four replications. All the extracts observed varied mycelial growth of *Pleurotus* spp. After nine days maximum radial growth (90.00 mm) in both species (*P. sapidus* and *P. flabellatus*) was observed in barley extract media. The maximum growth rate (10.00 mm/day) of mycelium in *P. sapidus* and *P. flabellatus* was recorded in barley extract agar. Maximum dry mycelium weight (7.98 mg/100 ml & 8.35 mg/100 ml) of *P. sapidus* and *P. flabellatus* was observed in barley extract broth, respectively.

Key words: *Cereals, Media, Mycelial growth and Pleurotus.*

Introduction

Mushrooms are the macro fungi which possess fleshy, sub fleshy fruiting bodies of fungi. Mushroom has been defined as "a macrofungus with a distinctive fruiting body which can be either epigeous or hypogeous and large enough to be seen with naked eyes and picked by hand" (Chang and Miles, 1993). Mushroom is being widely used as food and food supplements from ancient times. The cultivated edible mushrooms are a group of large macroscopic fleshy fungi, generally belong to Basidiomycetes but some are Ascomycetes. Oyster mushroom (*Pleurotus* spp.*)* cultivation has increased tremendously throughout the world during the last few decades (Chang, 1999; Royse, 2002). The name *Pleurotus* has its origin from Greek word, 'Pleuro' that means formed laterally or lateral position of the stalk or stem. Oyster mushroom commonly referred as 'Dhingri' in India, is a Basidiomycetes and belongs to the genus *'Pleurotus'*. It is lignocellulolytic fungus that grows naturally in the temperate and tropical forests on dead, decaying wood logs, sometimes on drying trunks of deciduous or coniferous trees. It can also grow on decaying organic matter. The fruiting bodies of this mushroom are distinctly shell, fan or spatula shaped with different shades of white, cream, grey, yellow, pink or light brown depending upon the species. However, the colour of the sporophores is extremely variable character influenced by the temperature, light intensity and nutrients present in the substrate. The avocation of mushroom farming will become a very important cottage industry in the integrated rural development programmes, which will lead to the economic betterment of not only small farmers but also of landless labourers and other weak sections of communities. About 385 million tonnes of agricultural wastes are available annually in India and about half of this residue remains unused. If even 1% of this crop residue is used to produce mushroom, India will become a major mushroom

producing country in the world. Edible mushroom production represented an attractive method of improving the nutritional quality of ligno-celluloid wastes for use as an animal feed stock. Among the various physical, chemical and biological methods used for upgrading the digestibility and nutritive value of agricultural wastes, biodegradation by using white rot fungi including mushrooms have been found promising. Mushroom production represents one of the most commercially important steps towards diversification of agriculture based on microbial technology for large- scale recycling of agro-wastes in an agricultural country like India. Oyster mushroom (*Pleurotus* spp.) is a ubiquitous mushroom cultivating worldwide. Variations in environmental factors such as pH, temperature and different media have great influence on the growth (Bugarski, 2002). In view of this research was conducted to identify best media for growth of *Pleurotus* spp.

MATERIALS AND METHODS

Experimental site

For the present investigations, experiments were conducted at Mushroom Laboratory, Department of Plant Pathology, College of Agriculture, Sardar Vallabhbhai Patel University of Agriculture & Technology, Modipuram, Meerut, Uttar Pradesh- 250110 situated on the Western side of the Delhi - Dehradun high way NH-58 at a distance of 10.0 km away in the north of Meerut city. The district Meerut is situated between 29^{o} 01'N latitude and 77^{o} 45'E longitude at an altitude of 237 meters above the mean sea level.

Establishment and maintenance of pure culture

The culture of *P. sapidu*s and *P. flabellatus,* used in the present investigations were collected from Directorate of Mushroom Research Centre, Solan, Himanchal Pradesh and Mushroom Research and Training Centre, G. B. Pant University of Agriculture and Technology, Pantnagar. The cultures of *Pleurotus* species were further purified by single hyphal tip method. For this purpose, the cultures were grown in sterilized petri plates on potato dextrose agar (PDA) medium for 8 days. Single branched hyphae from the periphery of the growing colony were marked under low power (10x) of compound microscope and transferred to PDA slants for maintenance. These culture tubes were incubated at 24 ± 1^{0}C for about a week and again sub-cultured on PDA medium and then stored in a refrigerator at 05 ± 1^{0}C for further use.

Cultivation Media

For the effect of different media studies, seven media (*i.e.* wheat extract agar, rice extract agar, barley extract agar, oat extract agar, sorghum extract agar, pearl millet extract agar and potato dextrose agar) were used for the radial growth. The ingredients and methods of their preparation are given below:

Wheat Extract Agar (WEA) Medium

Wheat grain	200g
Agar-agar	20g
Distilled Water	1000ml

Two hundred gram grains were washed with water 2-3 times and then boiled with 500 ml distilled water for 20 minutes, allowed to cool, the grains were then separated and the liquid suspension passed through a muslin cloth. The volume of the extract so obtained was made up to 500 ml by adding distilled water. Twenty gram agar-agar was melted separately in 500 ml of distilled water and mixed with grains extract. The total volume was made up to 1000 ml by adding distilled water.

Rice extract agar, barley extract agar, oat extract agar, sorghum extract agar, pearl millet extract agar and potato dextrose agar were also prepared by same methods as described above for Wheat extract agar medium. All the seven prepared media were sterilized by autoclaving at 1.1 kg/cm^2 pressure (121^{0}C) for 20 minutes. The test media were poured to Petri plates and culture tubes then inoculated with culture of *Pleurotus* spp. under aseptic conditions. The plates (90 mm @ 20 ml/plate) were inoculated with culture of *Pleurotus* spp. centrally and incubated at 27±1°C. Radial growth and growth rate were determined at each 48 hrs till the colony covered the full plate.

Statistical Analysis

The suitable statistical design (CRD) was applied and the data thus obtained were analysed statistically. Analysis of variance (ANOVA) technique and critical difference (CD) was calculated at five per cent level of significance for comparison with other treatment.

Result and Discussion

This experiment was conducted for the study of effect of different cereal grains extract on mycelial growth of *P. sapidus* and *P. flabellatus in-vitro* condition. In the experiment six different types of cereals extract media *viz.* wheat extract agar, rice extract agar, barley extract agar, oat extract agar, sorghum extract agar, pearl millet extract agar were used, while potato dextrose agar media was use as control with four replications. The observations of mycelial growth were recorded on 3rd, 6th and 9th days after inoculation as shown in Table-.

On 3rd day, maximum radial growth (19.75 mm) of *P. sapidus* was recorded in barley extract agar followed by oat extract agar (17.00 mm) and minimum radial growth (11.25 mm) in potato dextrose agar. While in case of *P. flabellatus* maximum radial growth (29.00 mm) was recorded in barley extract agar followed by oat extract agar (28.50 mm) and minimum radial growth (20.75 mm) was observed in potato dextrose agar.

On 6th day in case of *P. sapidus* maximum radial growth (59.00 mm) was recorded in barley extract agar and statically similar with oat extract agar followed by pearl millet extract agar (57.75 mm). Minimum radial growth (49.50 mm) was recorded in potato dextrose agar which was statistically lower than all treatment. While in case of *P. flabellatus* maximum radial growth (72.50 mm) was recorded in barley extract agar followed by oat extract agar (65.50 mm) which was significantly higher than all treatments. Minimum radial growth (56.00 mm) was recorded in potato dextrose agar (control).

On 9th day in case *P. sapidus* maximum radial growth (90.00 mm) was recorded in barley extract agar followed by oat extract agar (88.00 mm), which was significantly higher than all treatments. Minimum radial growth (76.50 mm) was recorded in potato dextrose agar. While in case of *P. flabellatus* maximum radial growth (90.00 mm) was recorded in

barley extract agar followed by oat extract agar (87.75 mm). Minimum radial growth (75.50 mm) was recorded in potato dextrose agar (control).

On 9th day, growth rate (mm/day) was recorded in *P. sapidus* and *P. flabellatus.* Maximum growth rate (10.00 mm/day) of *P. sapidus* was recorded in barley extract agar followed by oat extract agar (9.77 mm/day) and minimum growth rate (8.50 mm/day) was recorded in potato dextrose agar. While in case of *P. flabellatus* maximum growth rate (10.00 mm/day) was recorded in barley extract agar followed by oat extract agar (9.75 mm/day) and minimum growth rate (8.38 mm/day) was recorded in potato dextrose agar which was significantly lower than all other treatments.

The results were in accordance with the findings of Hussain and Hussain, (2004) who reported that *Pleurotus* spp. showed fastest growth of mycelium on potato dextrose agar among different media used. Baliyan (2008) studied the mycelial growth of *Pleurotus* spp. (*i.e. P. florida. P. flabellatus, P. sajor-caju, P. fossulatus and P. sapidus*) rates which were higher on MEA (Malt Extract agar) medium than on PDA or WSEA. Zubair (2012) also observed maximum radial growth (9.00 cm) of *P. sapidus* in potato dextrose agar medium. Bhadana (2014) also reported maximum radial growth was found in oat extract agar medium and potato dextrose agar medium and pearl millet extract agar medium of *P. djamore*, *P. florida*, *P. eryngii* and *P. flabellatus* respectively. Yadav (2014) radial growth of *P. flabellatus* was found maximum in barley extract agar followed by oat agar medium and minimum radial growth recoded in potato dextrose agar medium. Sardar *et al*., (2015) also reported the effects of various growth conditions on growth and development of *Pleurotus* species six different *Pleurotus* strains were cultured on different agar media *viz*. PDA (Potato dextrose agar), MEA (Malt extract agar) and WEA (Wheat extract agar). Among these media Potato dextrose agar medium (PDA) was found to be the best medium than malt extract agar (MEA) and wheat extract agar (WEA) for the growth of mycelium of all *Pleurotus* species.

Dry mycelial weight of *Pleurotus* spp.

This experiment was conducted for the study of dry mycelial weight of *P. sapidus* and *P. flabellatus in-vitro* condition. In the experiment seven different types of broth media *viz.* wheat extract broth, rice extract broth, barley extract broth, oat extract broth, sorghum extract broth, pearl millet extract broth and potato dextrose broth were taken with four replications as shown in Table-2.

In case of *P. sapidus* maximum dry mycelium weight (7.98 mg/100ml) was observed in barley extract broth significantly higher than all treatments followed by oat extract broth (6.60 mg/100ml). Minimum dry weight of mycelium (5.06 m/100ml) was observed in potato dextrose broth. While in case of *P. flabellatus* maximum dry weight (8.35 mg/100ml) was observed in barley extract broth significantly higher than all treatments followed by oat extract broth (7.58 mg/100ml). Minimum dry weight (5.26 mg/100ml)) was observed in potato dextrose broth.

Dry matter growth rate (mg/day) of *P. sapidus*, maximum dried mycelial growth rate (0.53 mg/day) was observed in barley extract broth followed by oat extract broth (0.44 mg/day). The minimum dried mycelial growth rate (0.33 mg/day) was observed in potato dextrose broth. While in case of *P. flabellatus* maximum dried mycelial growth rate (0.55 mg/day) was observed in barley extract broth followed by oat extract broth (0.50 mg/day). The minimum dried mycelial growth rate (0.35 mg/day). The results were in accordance with

the findings of Potato Dextrose Broth has been reported to be supporting maximum mycelial growth by earlier workers Suharban and Nair (1991). Kumar (2015) also revealed that maximum radial growth was recorded on pigeon pea extract agar medium (90.00 mm) and minimum radial growth was found in control (69.50 mm) in *P. flabellatus*. Maximum radial growth rate was observed in pigeon pea extract agar medium (11.25 mm/day) in *P. flabellatus*. Maximum dried mycelial weight was in pigeon pea extract broth medium (5.86 mg/50ml) in *P. flabellatus*.

S. No.	**Media**	**Radial Growth (mm)**						**9th days Growth rate (mm/day)**	
		3rd day		**6th day**		**9th day**			
		P. sapidus	*P. flabellatus*	*P. sapidus*	*P. flabellatus*	*P. sapidus*	*P. flabellatus*	*P. sapidus*	*P. flabellatus*
1.	Wheat Extract media	11.50	23.50	50.50	59.50	80.50	80.00	8.94	8.88
2.	Rice Extract media	12.75	24.50	51.00	61.00	81.25	82.25	9.02	9.13
3.	Barley Extract media	19.75	29.00	59.00	72.50	90.00	90.00	10	10
4.	Oat Extract media	17.00	28.50	59.00	65.50	88.00	87.75	9.77	9.75
5.	Sorghum Extract media	15.75	26.25	56.50	60.25	83.75	84.25	9.30	9.36
6.	Pearl Millet Extract media	16.00	27.00	57.75	63.25	85.00	87.00	9.44	9.66

Table-01: Effect of different cereals grain extracts media on mycelial growth of *Pleurotus* spp. (*P. sapidus* and *flabellatus*).

7.	Potato Dextrose media (Control)	11.25	20.75	49.50	56.00	76.50	75.50	8.50	8.38
CD at 5%		**3.06**	**3.00**	**4.57**	**3.54**	**3.09**	**3.58**	**-**	**-**
SE(m)		**1.03**	**1.01**	**1.54**	**1.19**	**1.04**	**1.21**	**-**	**-**

Table-02: Effect of different cereals grain extracts broth media on dry mycelial weight of *Pleurotus* spp.

S. No.	**Broth Media**	***P. sapidus***		***P. flabellatus***	
		Dry matter weight (mg/100ml)	Dry matter growth rate (mg/day)	Dry matter weight (mg/100ml)	Dry matter growth rate (mg/day)
1.	Wheat Extract broth media	5.76	0.38	6.74	0.44
2.	Rice Extract broth media	5.88	0.39	6.80	0.45
3.	Barley Extract broth media	7.98	0.53	8.35	0.55
4.	Oat Extract broth media	6.60	0.44	7.58	0.50
5.	Sorghum Extract broth media	5.90	0.39	7.20	0.48
6.	Pearl Millet Extract broth media	6.41	0.42	7.33	0.48
7.	Potato Dextrose broth media (Control)	5.06	0.33	5.26	0.35
CD at 5%		**0.42**	**-**	**0.33**	**-**
SE(m)		**0.14**	**-**	**0.11**	**-**

Conclusion: In this article effect of grains was observed more or less effect on mycelial growth against *Pleurotus* spp. Radial growth was observed in Barley extract media. Maximum dry mycelium weight (7.98 mg/100ml & 8.35 mg/100ml) of *P. sapidus* and *P. flabellatus* was observed in barley extract broth respectively. This might enhance the mushroom production in the coming future.

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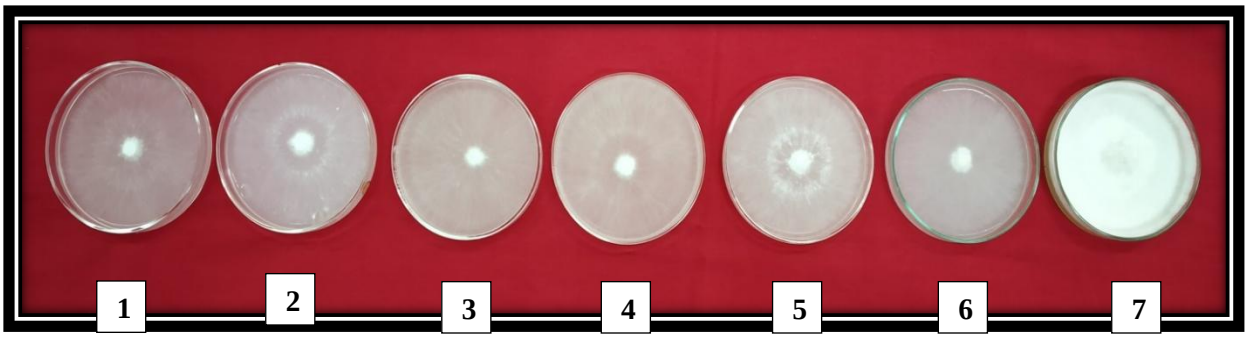

Fig 1 Pleurotus sapidus

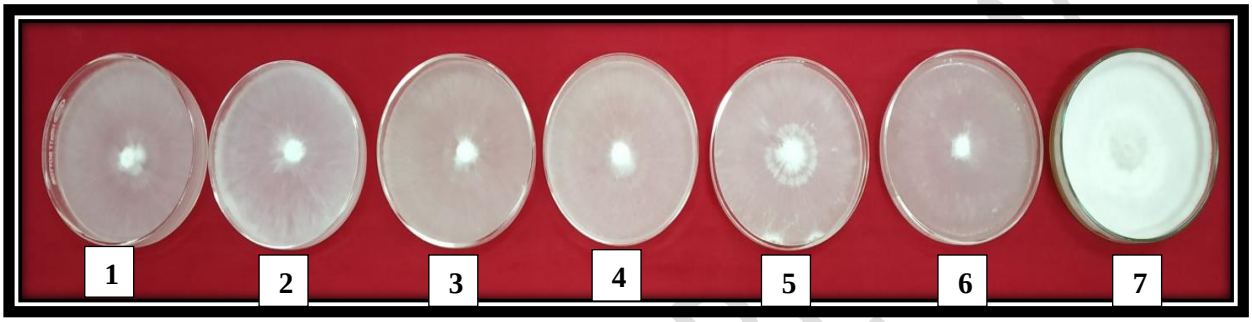

Fig 2 *Pleurotus flabellatus*

Plate-1: Effect of different cereal grains extract media on radial growth of *Pleurotus* species.

1. Wheat Extract media 2. Rice Extract media 3. Barley Extract media 4. Oat Extract media 5. Sorghum Extract media 6. Pearl Millet Extract media 7. PDA media (Control)

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