

E-ISSN: 2278-4136 P-ISSN: 2349-8234 www.phytojournal.com JPP 2020; 9(6): 881-888

Received: 19-08-2020 Accepted: 30-09-2020

Phadke Monika Vilas Agricultural Microbiology Mahatma Phule Krishi Vidyapeeth, Rahuri, Maharashtra, India

Dr. Jadhav AC

Agricultural Microbiology Mahatma Phule Krishi Vidyapeeth, Rahuri, Maharashtra, India

Dhavale MC

Agricultural Microbiology Mahatma Phule Krishi Vidyapeeth, Rahuri, Maharashtra, India

Dr. Hasabnis SN

Agricultural Microbiology Mahatma Phule Krishi Vidyapeeth, Rahuri, Maharashtra, India

Dr. Gaikwad AP

Agricultural Microbiology Mahatma Phule Krishi Vidyapeeth, Rahuri, Maharashtra, India

Dr. Jadhav PR

Agricultural Microbiology Mahatma Phule Krishi Vidyapeeth, Rahuri, Maharashtra, India

Patil Savita Ajit Agricultural Microbiology Mahatma Phule Krishi Vidyapeeth, Rahuri, Maharashtra, India

Corresponding Author: Phadke Monika Vilas Agricultural Microbiology Mahatma Phule Krishi Vidyapeeth, Rahuri, Maharashtra, India

Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com



Effect of cultural variability on mycellial growth of eleven mushroom isolates of *Pleurotus* spp.

Phadke Monika Vilas, Dr. Jadhav AC, Dhavale MC, Dr. Hasabnis SN, Dr. Gaikwad AP, Dr. Jadhav PR and Patil Savita Ajit

Abstract

Mushroom is a macro fungus with a distinctive fruiting body which can be either epigeous or hypogenous and large enough to be seen with naked eye and to be picked by hand. The diverse climatic condition in India made this country a natural habitat for many mushrooms. The oyster mushroom (*Pleurotus* spp.) is highly suitable for commercial cultivation in subtropical regions of the world. They occur seasonally all over the world in various habitats such as humus rich soils, decaying plant litter and wood logs in forests as well as in meadows. The current study deals with "Molecular diversity among wild edible oyster mushroom (*Pleurotus* spp.) from western ghat of Maharashtra."

It includes the collection of isolates from AICRP on mushroom, college of Agriculture Pune. After collection its cultural variability on different media was studied different pH levels and different temperature regimes were studied. Three culture media (PDA, MEA and Richard's Agar) were tested for growth of different *Pleurotus* spp. Among them growth of PN-18-10 was significantly more on Malt extract agar (70.40 mm). Whereas, growth of PN-17-31 were significantly more on potato dextrose agar (44.12 mm) and growth of PN-17-23 significantly more on Richard's agar media (50.44 mm). All eleven isolates grown on 5,6,7,8, and 9 pH levels and among them pH 6 showed optimum growth rate for All eleven isolates. OnpH 6.0 growth was observed about 34.11 mm followed by pH 8.0. On pH 5.0 PN-18-of oyster mushroom were tested for effect of different range of temperature *viz.*, 20 °C, 25 °C, 30 °C, 35 °C, 40 °C and found that they were grown well at 30 °C temperature with maximum mycelial diameter 56.00 mm in *P. cystidiosus*, 87.00 mm in *P. populinus* and 48.00 mm in P. ostreatus.

Among the cultural variability variation due to different media different pH levels and different temperature regimes were studied. Among the tested eleven isolates of oyster mushroom all isolates showed significant growth on MEA media followed by PDA media. For the optimum pH level, pH6.0 found optimum for the growth of mycelia of most of the *Pleurotus* isolates.

Keywords: Mushroom, isolates, malt, growth, temperature

1. Introduction

Mushroom is a macro fungus with a distinctive fruiting body, which can be either epigeous or hypogeous and large enough to be seen with naked eye and to be picked by hand (Chang and Miles, 1992)^[6].

The word mushroom is derived from the Greek word for fungi and moulds. Mushrooms are non-timber forest products which are often found as saprophytes on soil, open fields, farm lands, woods and road sides. Around 1650, a melon grower near Paris discovered mushrooms growing on his melon field. Fungi were most likely cultivated for the first time around the year 600 in Asia. In Europe, the first cultivated fungi, the mushroom was introduced in the 17th century. Mushrooms were introduced in the Netherlands for the first time at the beginning of the 19th century. After 1900's, mushrooms were cultivated on a large-scale in the marl mines in Limburg.

Pleurotus species constitute one of the choicest edible mushrooms, commonly known as 'Oyster Mushroom' and in India recognized by the name 'Dhingri Mushroom'. It grows naturally in the temperate and tropical forests on dead and decaying wooden logs or sometimes on dying trunks of deciduous or coniferous woods. It may also grow on decaying organic matter. The fruiting bodies of the mushrooms are distinctly shell or spatula shaped with different shades of white, cream, grey, yellow, pink or light brown depending upon the species. Species of *Pleurotus* are widely known as efficient decomposers of a large range of agricultural wastes and produce edible basidiomycota of high organoleptic qualities. These properties favored significantly the spread of their commercial cultivation which recently accounted for about a quarter of the total world mushroom production (Chang and Miles, 1991)^[5].

Oyster mushrooms are the third largest cultivated mushroom. The commercial production of oyster mushroom has increased 25 fold worldwide since 1981.

In 1981, 35000 tons of oyster mushrooms were produced, accounting for approximately 2.8% of the total world production of edible mushroom. By 1997, production reached 876000 tons accounting for 14.2% of the total world supply of cultivated edible mushrooms

Most cultivable mushrooms have specific requirements for growth in auxenic culture. The main factors affecting growth are nutrient sources and environmental factors such as temperature and pH. The media generally contain a carbon source, nitrogen source and vitamins. The carbon source is especially important and should be in greater quantities than other essential nutrients and generally in the range of 3-28%. Mushrooms can be grown on different carbon sources such as glucose, galactose, mannose, fructose, sucrose, cellulose, dextrin and starch. Nitrogen sources such as ammonium nitrate, calcium nitrate, yeast extract, soybean, arginine and glutamic acid have been used to promote mycelial growth. Mycelial growth is strongly influenced by in vitro conditions and they obtain their nutrients by absorbing soluble inorganic and organic materials from medium which assures the maximum and most vigorous germination. It has also been reported that healthy and active mycelial growth in medium plays a crucial role for protecting themselves against several stress factors.

Malt Extract Agar (MEA), Potato Dextrose Agar (PDA) and Richard's agar media support the mycelial growth of oyster mushroom. The optimum temperature of mycelial growth varies with the species of mushroom. For example, *Volvariella volvacea* grows well at 35 °C, *Pleurotuseryngii* at 25°C, *Pleurotus ostreatus* and *Pleurotuspulmonarius* at 30 °C, *Agrocybeaegerita* at 25 °C to 30 °C, *Lentinusstrigosus* at 35°C and *Lentinula edodes at* 20°C to 30°C

2. Material and Method

The present investigation "Molecular Diversity among wild Edible Oyster Mushroom (*Pleurotus* spp.)From western *Ghats* of Maharashtra" was conducted at All India Coordinated Research Project on Mushroom, College of Agriculture, Pune, Biotechnology Laboratory College of Agriculture, Pune and Department of Plant Pathology, College of Agriculture Pune. The details of the material used, methodology and statistical procedure followed during the course of investigation are described in this chapter.

2.1 Material

The experimental material for the study consisted of 11 isolates of Oyster mushroom (*Pleurotus* spp.) received from All India Co-ordinated Research Project on Mushroom, College of Agriculture, and Pune Maharashtra. The list of isolates is presented in Table No. 3.1.

2.1.1 Pure isolates

The present study was limited to only *Pleurotus* spp. The pure isolates were maintained on the slants of malt extract agar under aseptic condition which were collected in survey during monsoon of year 2017-18 from different forests of northern Maharashtra, including *Sahyadri* and *Satpura* valley regions.

2.1.2 Culture media

Potato Dextrose Agar (PDA) and Malt Extract Agar (MEA) media were used for sub culturing of different wild oyster isolates.

2.1.3 Surface disinfectants: Mercuric chloride: Stock Solution: Mercuric chloride 20g, concentrated hydrochloric

acid 100 ml. Working Solution: Mercuric chloride was dissolved in concentrated hydrochloric acid and maintained as a stock solution. At the time of use, 5 ml of the stock solution was diluted with 995 ml of distilled water. The disinfectant was used for surface sterilization.

2.1.4 Formalin

A four per cent of formalin solution was prepared by diluting 100ml of commercial formalin (40% strength) with 900 ml of distilled water.

2.1.5 Maintenance of aseptic condition

In order to maintain aseptic conditions in the laboratory, all operations were carried out inside a transfer chamber, provided with laminar air flow at All India Co-ordinated Research Project on Mushroom, College of Agriculture, Pune. After 14 days of inoculation, colony diameter was measured. Observations were recorded in three replications. Data were analyzed statistically using complete randomized design.

2.2 Study of Physiological Parameters like Different pH and Temperature Levels

These two experiments were conducted in the laboratory of Plant Pathology, Department of Plant Pathology, College of Agriculture, and Pune. Petri dishes (9 cm diameter) were used in experiments were cleaned with ordinary water after which they were dipped in a solution of potassium dichromate and sulphuric acid overnight. After treatment, these Petri plates were dried under shade, sterilized in hot air oven at 160 ^oC for two hours.

2.2.1 Effect of pH levels on growth of oyster mushroom

For studies of suitable pH, five different pH level *viz.* 5, 6, 7, 8 and 9 were used. Required pH of the culture media were adjusted with N/10 solutions of NaOH or HCl used before sterilization, it was measured by a digital pH meter. After sterilization at 121 $^{\circ}$ C (1.053 kg/cm²) for 20 minutes in autoclave, sterilized MEA media were poured into the Petri plates (90 mm @ 20 ml/plate). The plates were inoculated with mycelial disc centrally and incubated at $27\pm1^{\circ}$ C. The observations of radial growth and growth rate were taken at each 48 hrs till the colony covered the full plate.

2.2.2 Effect of Different temperature levels on growth of oyster mushroom

The mycelium disc consisted of mycelial disc of 5mm diameter was cut from margin of an actively growing colony of the test fungus. In solid media, the disc was carefully transferred to the center of each of the sterilized Petri plates. Strict aseptic conditions were maintained throughout the process of inoculation. To determine the optimum temperature for growth and multiplication of *Pleurotus* spp. the inoculated Petri plates were incubated at different temperatures *viz.*, 20, 22,24,26,28, and 30 °C under separate incubators. The observations of radial growth and growth rate were taken at each 48 hrs till the colony covered the full plate. Observations were recorded in three replications. Data were analyzed statistically using completely randomized design.

3. Results and Discussion

Molecular diversity studies among wild edible mushroom *Pleurotus* spp. was carried out by conducting experiments during the year 2018-19 in the Laboratory of Plant Pathology, AICRP on Mushroom and Laboratory of Agricultural Biotechnology at College of Agriculture, Pune. The

Journal of Pharmacognosy and Phytochemistry

experiments included collection of oyster mushroom isolates; study of variation effects due to different media, pH levels and temperature regimes on mycelial growth. The molecular characterization of eleven isolates of Pleurotus spp. has been accomplished. The results thus obtained are presented in different sections of this chapter and discussed with comparing earlier reported findings.

3.1 Effect of Different Media on Mycelia Growth of Oyster Mushroom

Based on cultural characterization it was observed that mycelium was hyaline, filamentous and whitish except in isolate PN-18-38. Its mycelium was blackish appeared on the inoculated bit. It was different in mycelial color characteristic than other ten isolates (Plate: 1). the result indicated that the initiation of mycelial growth of oyster mushroom in media was observed at different time. Mycelia growth was initiated after 24 hr of inoculation in PN-17-55 and growth rate was 5.34 mm day⁻¹ and in isolate PN-18-10 @ 5.66 mm day⁻¹. While, in the case of PN-18-15 (4.62 mm day⁻¹) initiation of mycelial growth was observed after 48 hr of inoculation. On 8th day of inoculation PN-18-10 was grown maximum (45.28 mm), which was followed by PN-17-55 (42.75 mm) and PN-17-23 (41.30 mm). Mycelium growth of PN-18-9 was 40.92 mm, followed by PN-17-30. Isolates PN-17-31 and PN-17-36 were statistically at par with each other with respect to mycelial growth. Lowest growth of mycelium as 36.11 mm was observed in PN-18-38 (Table: 1).

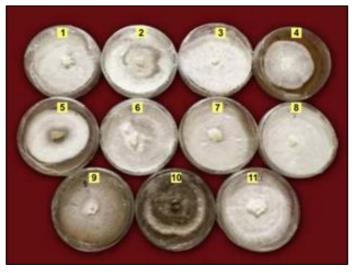


Plate 1a: Pure Cultures of isolates of Pleurotus Spp



Plate 1b: Cultural Variability among Pleurotus isolates on MEA medium (8 DAI)

 Table 1: List of 11 isolates of oyster mushroom (Pleurotus spp.)

Sr. No.	Isolates	DMR accession No.	Sr. No.	Isolates	DMR accession No.
1.	PN-17-16	DMRO-1026	2.	PN-17-23	DMRO-1027
3.	PN-17-30	DMRO-1029	4.	PN-17-31	DMRO-1030
5.	PN-17-36	DMRO-1033	6.	PN-17-55	DMRO-1037
7.	PN-18-9	DMRO-1113	8.	PN-18-10	DMRO-1114
9.	PN-18-15	DMRO-1116	10.	PN-18-38	DMRO-1119
11	PN-18-50	DMRO-1123			

Three different media including PDA, MEA and Richard's

Agar were tested for their preference toward the growth by different isolates of *Pleurotus* mushroom. The results on the mycelial growth are presented in Table-1 and Plate-.1. Among the various media tested, after 8th day of inoculation, the mycelial growth of all eleven isolates of oyster mushrooms was significantly more on malt extract agar (43.56 mm) and growth rate was 5.44 mm day⁻¹. This was followed by potato dextrose agar (37.78 mm); 4.72 mm day⁻¹. Slowest growth was observed on Richards' agar medium (37.12 mm) 4.64 mm day⁻¹ (Table: 2; Plate 2, 3, 4; Figure: 1).

Table 2: Mycelial growth and growth rate of eleven isolates of Pleurotus spp. influenced by three different media	

	Mycelial growth (mm) and growth rate (mm / day) on ith day of inoculation									
Name of isolates.	2 DAI		4 DAI		6 DAI		8 DAI			
	mm	mm/day	mm	mm/day	mm	mm/day	mm	mm/day		
			Is	olates						
PN-17-16	21.30	10.65	25.00	6.25	30.96	5.16	37.73	4.72		
PN-17-23	23.19	11.60	27.41	6.85	33.13	5.52	41.30	5.16		
PN-17-30	22.96	11.48	27.19	6.80	34.00	5.67	39.50	4.94		
PN-17-31	24.19	12.10	27.68	6.92	34.30	5.72	39.19	4.90		
PN-17-36	22.52	11.26	26.99	6.75	34.97	5.83	38.11	4.76		
PN-17-55	24.35	12.18	29.10	7.28	33.99	5.67	42.75	5.34		
PN-18-9	21.13	10.57	26.18	6.55	31.00	5.17	40.92	5.12		
PN-18-10	24.97	12.49	32.95	8.24	39.46	6.58	45.28	5.66		
PN-18-15	18.91	9.46	22.70	5.68	28.68	4.78	36.95	4.62		
PN-18-38	20.99	10.50	23.97	5.99	30.21	5.04	36.11	4.51		
PN-18-50	21.37	10.69	26.17	6.54	29.43	4.91	38.07	4.76		
SEm ±	0.17		0.10		0.11		0.10			
C.D. (0.05)	0.49		0.28		0.31		0.29			
			Me	edia(b0)						
B1 (MEA)	25.13	12.56	24.70	6.18	36.19	6.03	43.56	5.44		
B2 (PDA)	21.43	10.71	30.03	7.52	31.42	5.2	37.78	4.72		
B3(Richard's Agar)	20.5	10.25	25.79	6.29	30.60	5.1	37.12	4.64		
SEm ±	0.09		0.05		0.06		0.05			
C.D. (0.05)	0.25		0.15		0.16		0.15			
	· ·		Interaction (Strain X Medi	a)					
S. Em. ±	0.30		0.17	0.19		0.18				
C. D. (0.05) 0.84		0.84	0.48	0.54		0.51				



Plate 2a: Cultural Variability among *Pleurotus* isolates on PDA medium (8 DAI)



Plate 2b: Cultural Variability among *Pleurotus* isolates on MEA medium (8 DAI)



Plate 3: Cultural Variability among Pleurotus isolates at pH 5.0 (8 DAI)

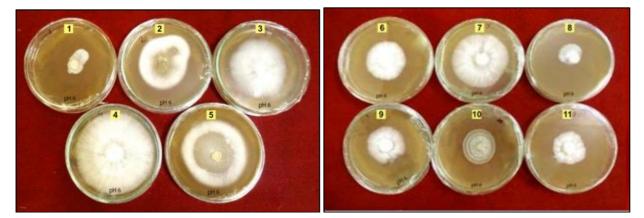


Plate 4: Cultural Variability among Pleurotus isolates at pH 6.0 (8 DAI)

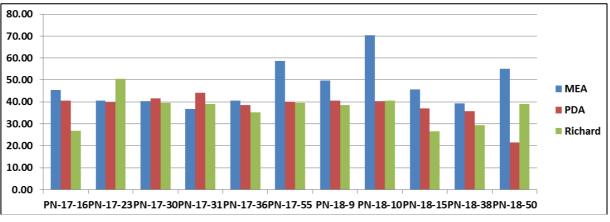


Fig 1: Effect of different media on mycelial growth of isolates of Pleurotus spp.

Among different media used in the present investigation i.e. PDA, MEA and Richard's Agar, malt extract agar proved to be the one of the best option for the mycelial growth of *Pleurotus*. The difference in mycelial growth on different media occurred due to availability of different carbon sources and required nutrients.

Mycelium growth was marginally better on a media containing dextrose than sucrose. However, Sardar *et al.* (2015) ^[19] reported that PDA exhibited higher carbon sources and nutrients for *P. oestreatus*, *P. sajorcaju*, *P. erynjii*, *P. columbine, and P. sapidus* mycelial growth in Petri plate. Chuku*et al.* (2015) ^[7] also reported that the effect of culture media on mycelial growth of *Pleurotusostreatus* on different media (MEA, PDA and SDA).

The result indicated that MEA and PDA exhibited abundant growth of *P. ostreatus* mycelium while SDA (Sabouraud Dextrose Agar) recorded scanty mycelial growth. Hoa and Wang (2015) ^[10] carried out an investigation on the mycelial growth of oyster mushroom *P. ostreatus* and *P. cystidiosus*. They reported that PDA and YDA (yam dextrose agar) were the most suitable media for the mycelium growth of oyster mushroom (*P. ostreatus*) while four media PDA, YDA, MEA, and SPDA (sweet potato dextrose agar) were supporting mycelium growth of oyster mushroom (*P. cystidiosus*).

Gibriel, *et al.* (1996) ^[9] had also reported Potato Dextrose Agar medium, as liquid or solid are the best medium tested for both rate and amount of fungal growth. Nasim *et al.* (2001) ^[16] studied the effect of culture media for mycelial growth of *P. ostreatus, P. sajorcaju, P. cistydious* and *Volvariella volvacea* on the fresh media plates of MEA, MS and PDA. The results indicated that the mycelial growth of *P. ostreatus, P. sajorcaju, P. cystidiosus and Volvariella. Volvacea* were maximum in MEA medium. Mohd (2012) ^[15] had evaluated six media, maximum radial growth was observed on MEA medium followed by PDA and minimum in water agar medium. Suharban and Nair (1994) ^[22] studied different media on growth of *Pleurotus* spp. and reported oat meal and potato dextrose agar to be superior for mycelial growth. Similar studies were conducted by Kapoor *et al.* (1997) and reported potato dextrose agar to be most suitable medium for growth of *P. fossulatus*.

It was observed on 8th day, that PN-17-30 and PN-17-31 showed highest growth on PDA rather than MEA. PN-18-10 showed highest growth on MEA i.e.70.40 mm. On Richard's agar medium it was observed that PN-17-23 and PN-18-10 showed good growth.

However, slowest growth was observed by PN-17-31 and PN-18-38 on MEA medium. On PDA, PN-18-38 and PN-18-50 showed slowest growth. Furthermore PN-17-16 and PN-18-15 showed slow growth on Richard's agar medium. This clearly states that different isolates of *Pleurotus* sp. had shown difference in preferences toward media.

3.1.1 Effect of Different pH levels On Mycelial Growth of Oyster Mushroom

Results of optimum pH required for mycelial growth of oyster mushroom after 8 days of inoculation were presented in Table-3, Table-4, Figure-2 and Plates-3, 4, 5, 6, 7. The eleven isolates of *Pleurotus* spp. were inoculated at five different pH regimes on MEA.

The observations on growth of mycelium were noted from 2nd day after inoculation till 8th day of inoculation. On 8th day after inoculation it was observed that PN-17-30 had maximum mycelial growth (35.49 mm) and growth rate 4.44 mm day⁻¹. The growth of PN-17-23 isolate was next highest and it was 34.85 mm @ 4.36 mm day⁻¹. This was followed by PN-17-31

which showed growth 31.98 mm in diameter; 4.00 mm day^{-1} . The isolates PN-17-36 and PN-18-9 were statistically at par with each other.

Lowest growth of mycelium was observed in isolate PN-18-15 (19.36 mm) 2.42 mm day⁻¹. Mycelium growth in PN-18-10 and PN-17-16 was statistically at par with each other as 20.86 and 20.69 mm, respectively. PN-17-55 showed intermediate growth (25.29 mm); 3.16 mm day⁻¹.

On pH 5 it was observed that isolate PN-17-23 and PN-17-30 grown maximum with 34.85 and 35.49 mm with growth rate 4.36 and 4.44 mm day⁻¹, respectively (Plate:3). On pH 6 all isolates showed good growth but isolate PN-17-30, PN-17-31, PN-17-23 and PN-17-36 showed more profuse growth. (Plate: 4). Media with pH 6 was observed superior in comparison to other levels studied as 5, 7, 8 and 9. Mycelium growth on pH 6 was observed 34.11 mm diameter; 4.26 mm day⁻¹. This was followed by media with pH 8 (28.07 mm), pH 7 (27.87 mm), pH 9 (24.76 mm) while the minimum colony diameter was measured on media with pH 5 (20.33 mm). PN-17-30 showed highest growth on pH 6 and pH 7. At pH 6, PN-17-31 showed profuse growth. PN-17-31 also showed good growth on pH 6. Slowest growth was observed by PN-18-15 and PN-18-38 at pH 6 and pH 7, respectively.

Singh *et al.* (2000) ^[20] studied effect of different initial pH value of nutrient medium of *P. flabelltus, P. oesterous,* and found pH 5 to 8 was most suitable for the growth of mycelium of *Pleurotus.* Ram and Pant (2001) ^[24] recorded the best mycelial colonization of both *P. flabellatus* and *P. sajorcaju* at pH 6.0 on all the three substrate media. Hao H.T.*et al.* (2015) ^[10] reported that the most suitable pH value for *P. flabellatus* ranged from 4.0 to 7.0 with an optimum pH of 5.5 to 6.0 and is in conformity with present findings. Shukla *et al.* (2003) ^[21] had observed mycelium growth on different pH, wherein investigated the suitable pH (3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 6.5.7.0, 8.0 and 8.5) for the growth of button mushrooms. The best pH level for the growth was 6.0 followed by 5.5.

3.1.2 Effect of different temperature levels on growth of oyster mushroom: Results of optimum temperature required

for mycelial growth of oyster mushroom after 8 days of inoculation were presented in Table-5, Figure-3 and Plate-8, 9, 10. Among the various temperature levels studied, the mycelial growth and growth rate of eleven *Pleurotus* isolates were significantly more at 30° C temperature (57.35 mm and 7.17 mm day⁻¹, respectively). The mycelial growth was 42.72 mm at 28°C and growth rate was 5.34 mm day⁻¹. Mycelium growth and growth rate was comparatively declined at 24°C as 33.7 mm and 4.22 mm day⁻¹, respectively. Furthermore, at temperature level 26°C it was observed 30.71 mm and growth rate was 3.84 mm day⁻¹ and at 20°C growth rate declined to 3.69 mm day⁻¹, respectively.

Overall lowest mycelial growth was 25.30 mm at 22°C temperature. Moreover, at same temperature the isolate PN-17-36 grew well with maximum mycelial diameter 47.52 mm and growth rate 5.94 mm day⁻¹ which was followed by 44.69 mm mycelium growth and growth rate 5.59 mm day⁻¹ by PN-17-23. The next best in the order was 42.47 mm; 5.31 mm day-¹ by PN-17-31. The isolate PN-17-30 exhibited growth of 40.53 mm and rate as 5.07 mm day⁻¹. Isolates PN-17-16 and PN-18-10 were statistically at par with each other. Isolates of oyster mushroom grew well at 30°C temperature with maximum mycelial diameter. The results are in support with the earlier findings of Ragupathi et al. (2016) [23] who observed that the mycelium of oyster mushroom grew at an average temperature of 28° to 30 °C. Nayak et al. (2015) reported that the *Pleurotus* spp. showed maximum mycelial growth at 25°C. Rout et al. (2015) ^[25] also carried out an investigation on the influence of incubation temperature on the linear mycelial growth of oyster species and showed that the mycelial growth of oyster mushroom was better at 25°C.In present investigation isolates of Pleurotus PN-17-23 and PN-17-36 showed maximum mycelium growth at 20 °C temperatures. These findings are in agreement with work of Ram and Pant (2001) ^[24] who observed that higher mycelial growth of P. sajorcaju and P. flabellatus at the temperature of 20 °C as compared to 25, 30, 35°C.

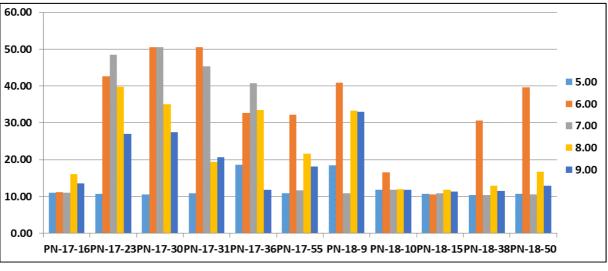


Fig 2: Effect of different pH levels on mycelium growth of isolates of Pleurotus spp.

At temperature 22° C the isolates PN-17-23 and PN-17-36 grew maximum and the pattern was followed by PN-17-31 and PN-18-9. The isolates PN-17-36 showed maximum growth rate of 5.94 mm day⁻¹ at 26° C. However, at temperature 28° C, PN-17-30 and PN-17-31 showed maximum

mycelium growth rate as 5.07 and 5.31 mm day⁻¹, respectively as compared to other tested nine isolates. Dung *et al.* (2012) ^[8] studied the growth parameter of two oyster mushrooms namely *P. cystidiosus* and *P. floridanus*. The superior mycelial growth was found at temperature of 27 °C. Kumla *et* *al.* (2013) ^[13] studied that the edible oyster mushroom genus *Pleurotus* was able to grow at a temperature ranging from 15 to 35°C with an optimal growth at temperature of 25 °C. Klomklung *et al.* (2014) ^[12] studied the growth parameter of three wild mushrooms namely *Lentinusconnatus*, *L. roseus* and *Pleurotus giganteus*. Mycelial growth occurred at a temperature ranging from 20 to 30 °C with an optimal growth at temperature of 30 °C and 25 °C for *Lentinus* and *Pleurotus* species, respectively. These findings are in conformity with work of Neelam *et al.* (2013) ^[17], who reported optimum temperature for *P. ostreatus* is in a range of 25 °C to 30 °C. Nwokoye *et al.* (2010) ^[18] described three different isolates of *P. ostreatus*. The high temperature strain (25 °C to 30 °C), medium temperature strain (16 °C to 22 °C) and low temperature strain (12 °C to 15 °C).

All tested eleven isolates of *Pleurotus* strain profusely grown at 30 $^{\circ}$ C temperature. Among eleven isolates PN-17-23 and PN-17-36 were able to grow profusely at lower temperature as 20 $^{\circ}$ C.

The PN-17-30 showed maximum growth at 28 and 30 ^oC which was followed by PN-17-31 showed growth at 26, 28 and 30 ^oC. PN-17-36 and PN-17-23 showed maximum growth at 20, 22 and 24 ^oC. Slowest growth rate was observed on PN-18-15 at all tested temperature levels except for 20^oC. Here also it showed variability towards growth by isolates with respect temperature regims.

4. Summary and Conclusion

Mushrooms are macro fungi. Macro fungi are those fungi that form large fructifications visible without the help of a microscope and have easily observable spore-bearing structures. Mushroom is a food of high quality flavor and nutritional value and has high content of protein, low content of fat, vitamins, minerals and high content of fibers. Oyster mushroom (*Pleurotus* spp.) is popularly known as '*dhingri*' in India and grows naturally in the temperate and tropical forests on dead and decaying wooden logs or sometimes on dying trunks of deciduous or coniferous woods. It may also grow on decaying organic matter. The fruiting bodies of this mushrooms are distinctly shell or spatula shaped with different shades of white, cream, grey, yellow, pink or light brown depending upon the species.

Investigation on "Molecular characterization of oyster mushroom (*Pleurotus* spp.) from western *Ghats* of Maharashtra" is carried out. The study included collection of isolates from AICRP on Mushroom, College of Agriculture Pune; effects of culture media, pH of medium and temperature on mycelial growth of oyster mushroom and molecular characterization of eleven isolates.

The study on effect of three media on growth of oyster mushrooms indicated that the growth of all eleven isolates was significantly more on Malt extract agar medium as 43.56 mm and growth rate was 5.44 mm day⁻¹. This was followed by potato dextrose agar (37.78 mm); 4.72 mm day⁻¹. Slowest growth was observed on Richards' agar medium (37.12 mm) 4.64 mm day⁻¹. Isolate PN-18-10 was grown maximum (45.28 mm), which was followed by PN-17-55 (42.75 mm) and PN-17-23 (41.30 mm). Mycelium growth of PN-18-9 was 40.92 mm, followed by PN-17-30. Isolates PN-17-31 and PN-17-36 were statistically at par with each other with respect to mycelial growth. Lowest growth of mycelium as 36.11 mm was observed in PN-18-38.

The study on effect of pH on mycelial growth of oyster mushroom indicated that on pH 5 it was observed that isolate PN-17-23 and PN-17-30 grown maximum with 34.85 and

35.49 mm with growth rate 4.36 and 4.44 mm day⁻¹, respectively. On pH 6, all isolates showed good growth but isolate PN-17-30, PN-17-31, PN-17-23 and PN-17-36 showed comparatively more profuse growth. Isolates PN-17-23 and PN-17-30 showed distinct characteristic of growth that they exhibited growth on all pH regimes.

Among the various temperature levels studied, the mycelial growth and growth rate of eleven *Pleurotus* isolates were significantly more at 30 °C temperature (57.35 mm and 7.17 mm day⁻¹, respectively). The mycelial growth was 42.72 mm at 28 °C and growth rate was 5.34 mm day⁻¹. Mycelium growth and growth rate was comparatively declined at 24 °C as 33.7 mm and 4.22 mm day⁻¹, respectively. Furthermore, at temperature level 26 °C it was observed 30.71 mm and growth rate was 3.84 mm day⁻¹ and at 20 °C growth rate further declined to 3.69 mm day⁻¹, respectively.

5. References

- 1. Ahmad I, Fuad I, Khan ZK. Mycelial growth of pink oyster (*Pleurotusdjamor*) mushroom in different culture media & environmental factors. Agr Food Sci 2015;2:6-11.
- Bresinsky A, Hilber O, Molitoris HP. The genus *Pleurotus* as an aid for understanding the concept of species in Basidiomycetes. In: The species concept in Hymenomycetes. (ed. Clemenson H) Cramer, Vadiz 1976, pp. 229-258.
- 3. Bao D, Ishihara H, Mori N, Kitamoto Y. Phylogenetic analysis of oyster mushrooms (*Pleurotus* spp.) based on restriction fragment length polymorphisms of the 5' portion of 26S rDNA. "J.Wood Sci 2004;50:169-176.
- 4. Chang ST, Buswell JA. Mushroom nutriceuticals. World J. Microbiol. Biotechnol 1996;12:473-476.
- 5. Chang ST, Miles PG. Recent trends in wood production of edible mushrooms. The Mushroom J 1991;503:15-18.
- 6. Chang ST, Miles PG. Mushroom biology a new discipline. The Mycologist 1992;6:64-65.
- Chuku SB, Nwachukwu EO, Stanley HO. Effects of selected culture media on mycellial growth of oyster mushroom (*Pleurotus*ostreatus). Afr J Biotechnol 2015;14:1471-1474.
- 8. Dung NTP, Tuyen DB, Quang PH. Morphological and genetic characteristics of oyster mushrooms and conditions effecting on its spawn growing. Int.Food Res J 2012;19:347-35.
- Gabriel AY, Ahmed M, Rasmy N, Rizk I, Abdel Rehem, NS, et al. Cultivation of Oyster Mushrooms (*Pleurotus* spp.): Evaluations of different media and organic substrates. Mushroom Biology and Mushroom products, Royse (ed.) 1996. ISBN 1-883956-01-3.
- 10. Hoa HT, Wang C. The effects of temperature and nutritional conditions on mycelium growth of two oyster mushrooms (*Pleurotus ostreatus* and *Pleurotus cystidiosus*). J Mycobiology 2015;43:14-23.
- 11. Kashyap V, Mishra R, Raghubansi BPS, Kumar P, Mishra YD. Effect of culture media and dose of substrate on growth parameter of oyster mushroom. Environment and Ecology 2015;34:686-688.
- 12. Klomklung N, Karunarathna SC, Hyde KD, Chukeatirote E. Optimal conditions of mycelial growth of three wild edible mushrooms from northern Thailand. Acta Biology Szeged 2014;58:39-43.
- 13. Kumla J, Suwannarach N, Jaiyasen N, Bussa Ban B, Lumyong S. Development of an edible wild strain of the

oyster mushroom for economic mushroom production. Chiang Mai J Sci 2013;0:161-172.

- 14. Kumar A, Thakore BB, Rathore VRS. Effect of growth parameter on mycelial culture and fructification of Plerotuseryngii. J Mycol. Pl. Path 2009;39:19-22.
- 15. Mohd Z. Studies on morphological variability of oyster mushroom. SVPUA&T, Meerut 2012, 40-68.
- Nasim G, Malik SH, Bajwa Afza lM, Mianm SW. Effect of three different culture media on mycelial growth of oyster and chinese mushrooms. J.Bio.1 Sci 2001;1:1130-1133.
- 17. Neelam S, Chennupati S, Singh S. Comparative studies on growth parameters and physio-chemical analysis of Pleurotus Ostreatus and Pleurotus Florida. Asian J Plant Sci Res 2013;3(1):163-164
- Nwokoye AI, Kuforiji OO, Oni PI. Studies on mycelial growth requirements of *Pleurotus ostreatus* (fr.) Singer. Int. J. Basic Applied Sci 2010;10:47-53.
- Sardar H, Muhammad AA, Chaudhary MA, Rashid A. Effects of different culture media, temperature and pH levels on the growth of wild and exotic Pleurotus species. Pakistan J. Phytopathology 2015;27:139-145.
- 20. Singh SK, Rana MK, Verma RN. Amplified DNApolymorphisms of cultivated mushrooms. Mushroom Res 2000;9(1):19-25.
- 21. Shukla S, Jaitly AK. Morphological and biochemical characterization of differentoyster mushroom (*Pleurotus* spp.). J Phytol 2011;3:18-20.
- 22. Suharban M, Nair MC. Physiological studies on *Pleurotus* spp. Mushroom Res 1994;3:87-104.
- 23. Ragupathi V, Kumerasan S, Selvaraju S, Karthikeyan S. Optimizing the growth conditions and adopting new methods growing oyster and milky mushrooms insame conditions. Int. J Herb Med 2016;4:1-4.
- 24. Ram RC, Pant DC. Effect of temperature and pH on mycelial growth of *Pleurotus* species. Indian J. Pl. Path 2001;19:58-60.
- 25. Rout MK, Mohapatra KB, Mohanty P, Chandan SS. Studies on effect of incubation temperature and light intensity on mycelial growth of oysters pecies. Jcrop weed 2015;11:44-46.
- Zardazil F, Kurtzman Jr RH. The biology of *Pleurotus* cultivation in the tropics. In Tropical Mushrooms, (Eds. S. T. Chang and T.H. Quimio), The Chinese University Press, Hong Kong 1984, pp. 227-298.
- 27. Zervakis G, Balis C. A pluralistic approach in the study of *Pleurotus* species with emphasis on compatibility and physiology of the European Morphotaxa. Mycological Research 1996;100:717-731.