



Efficacy of Different Cultural Parameters and Physical Factors on Mycelial Growth of Paddy Straw Mushroom (*Volvariella volvacea*)

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ABSTRACT

Background: Paddy straw mushroom (*Volvariella volvacea*) commonly known as the straw mushroom, in India, commonly referred to as "Pual mushrooms", "Pala chhatu" in odia belongs to Phylum-Basidiomycota, Class-Basidiomycetes, Order-Agaricales, Family-Pluteaceae and Genus Volvariella. The yield and flavor of the mushroom depends on several parameters including the mycelial density and growth. More vigorous is the mycelial growth, more will be the production. Hence to increase the mycelial run following treatments have been evaluated.

Methods: Fourteen different synthetic and semi-synthetic media were evaluated against mycelial growth of *V. volvacea*. Eight different pH levels and six different temperatures were tested against the test fungus along with different carbon and nitrogen sources.

Result: Out of the 14 media, Potato Dextrose Agar (90 mm), Yeast extract agar (90 mm) and oat meal agar (90 mm) were found suitable as they produce dense mycelium and cover the whole petriplate on 7th day of inoculation. From the various liquid media studied, it has been found that dense mycelium was obtained on Malt extract agar medium (449.25 mg). Maximum radial growth of the fungus was achieved at pH 7 and at 35°C temp (90 mm). Maltose, starch and sucrose recorded as best carbon sources for *V. volvacea* growth on solid base medium with an average diameter growth of 90mm and sucrose proved to produce dense mycelium on liquid media. Peptone was found to be best nitrogen source as it gives highest radial growth of the test fungus with an average diameter of 90 mm and also produced dense mycelial mat of the test fungus (400 mg).

Key words: Carbon, Media, Nitrogen, pH, Temperature, *Volvariella Volvacea*.

INTRODUCTION

Paddy straw mushroom (*Volvariella volvacea*), are generally known as the rice straw mushroom, or the Chinese mushroom. It belongs to the family Pluteaceae of the Basidiomycetes (Singer, 1961). Mushrooms are basically fungi, which have a fleshy and spore-bearing fruiting body. They have been in use not only for consumption purposes but also for medicinal purposes since ages (Verma and Vinita, 2017). Mushroom is a complete food suitable for all age groups, considering the health benefits, mushrooms are being considered as a future vegetable owing to their medicinal and nutritional properties and the mushroom consumption has significantly increased in recent years (Anusha *et al.*, 2023). Paddy straw mushroom is also known as warm mushroom because it grows in a fairly advanced temperature. It is an edible mushroom commonly grown in eastern regions of India, such as Odisha and West Bengal. Currently, countries with the highest mushroom production are China, USA, Poland, Netherlands, India, France, Spain, Canada, Mexico *etc* (Das *et al.*, 2024). Eastern Indian states, including the North Eastern region (Arunachal Pradesh, Meghalaya, Manipur, Mizoram, Tripura, Sikkim and Assam), as well as West Bengal, Bihar, Jharkhand and Odisha, have great potential and scope for paddy straw mushroom cultivation due to the wide vacuity of introductory substrate

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(paddy straw) and favorable temperature. The high temperature requirement of 26°C to 30°C for mycelium development and 34 to 37°C for fructification, along with relative humidity 70-90%, makes it a suitable choice for year-round mushroom cultivation (Biswas *et al.*, 2014).

MATERIALS AND METHODS

To test different artistic parameters and physical factors against mycelial product of *V. volvacea* this trial was conducted in SOA Mushroom Unit under Department of Plant Pathology of Siksha 'O' Anusandhan University,

Bhubaneswar in the year 2023-24 and the data were recorded.

Effect of different culture media on mycelial growth of *Volvariella volvacea*

In this experiment 14 different semisynthetic and synthetic media were taken. Potato dextrose agar, wheat extract agar, yeast extract agar, glucose peptone agar, sabouraud's dextrose agar, czapex dox agar, oat meal agar, richard's synthetic agar, malt extract agar, yeast malt agar, sweet potato dextrose agar, paddy straw extract agar, soil extract agar and compost extract agar media were prepared according to the standard literatures and the constituents are available easily in the locality. Ambystin 1 g per litre of media was added to prevent the bacterial infection. Liquid media were prepared by the same methods without agar. Final pH of the medium was adjusted to neutral 7 and the medium was autoclaved at 15 psi pressure for 20 minutes. Solid media were poured into presterilized petridishes and 40 ml of liquid media were kept in 100 ml conical flask. 5 mm discs of 10 days old pure culture of *V. volvacea* were cut with the help of cork borer and inoculated into each solid and liquid media and incubated $34\pm1^\circ\text{C}$ for 7 days. Four replications were maintained.

Effect of different pH on mycelial growth of *Volvariella volvacea*

For the study of different pH level effect on radial mycelial growth, PDA media of pH 2.0, 3.0, 4.0, 5.0, 6.0, 7.0, 8.0 and 9.0 were prepared by adding required amount of HCl and NaOH. 20 ml of PDA from each different pH was poured in four sterilized petridishes. 40 ml of each liquid media were prepared in 100 ml conical flask without adding agar. The medium was autoclaved at 15 psi pressure for 20 minutes.

After sterilization a small sample of the medium were taken and tested the pH level by pH meter and was observed a little deviation and accordingly the treatments were taken. Five mm discs of 10 days old pure culture of *V. volvacea* were cut with the help of cork borer and inoculated into each solid and liquid media and incubated at $34\pm1^\circ\text{C}$ for 7 days. Four replications were maintained.

Effect of different temperatures on mycelial growth of *Volvariella volvacea*

Five mm discs of 10 days old test mushroom was inoculated in the sterilized petridishes containing 20 ml of PDA and in the 100 ml conical flask containing 40 ml of potato dextrose broth. The medium was incubated at different temperatures viz, 20°C , 25°C , 30°C , 35°C , 40°C , 45°C for 7 days. Four replications were maintained. For the study of different temperature effect on mycelial growth of *V. volvacea*, this experiment was conducted in two stages adjusting three incubators at 20°C , 25°C and 30°C . After taking observation in 7 days second set of experiment was conducted by adjusting the incubators to 35°C , 40°C and 45°C . After period of experimentation both the data were combined to get the result.

Effect of different carbon sources on mycelial growth of *Volvariella volvacea*

Seven carbon sources namely glucose, fructose, maltose, starch, lactose, sucrose and mannitol were taken to optimize the growth of *Volvariella volvacea* keeping Sabouraud's dextrose medium as the basal medium. As we observed the sabouraud's medium supported good growth of fungi in comparison to other synthetic medium (Richard's and Czapex dox medium in growth study), we have taken Sabouraud's medium as basal medium. Twenty

Table 1: Growth of *Volvariella volvacea* on different solid and liquid media.

Treatments	Media	Radial growth of mycelium 7 DAI (mm)*	Dry weight of mycelium 10 DAI (mg)*	Quality of mycelia run
T ₁	Potato dextrose agar	90	106.25	Dense
T ₂	Wheat extract agar	68.7	159.50	Moderately dense
T ₃	Yeast extract agar	90	197.75	Dense
T ₄	Glucose peptone agar	90	303.75	Moderately dense
T ₅	Sabouraud's dextrose agar	90	203.50	Sparse
T ₆	Czapex dox agar	63.6	104.25	Dense
T ₇	Oat meal agar	90.0	328.25	Dense
T ₈	Richard's synthetic agar	35	100.00	Dense
T ₉	Malt extract agar	61.10	449.25	Sparse
T ₁₀	Yeast malt agar	90	307.00	sparse
T ₁₁	Sweet potato dextrose agar	80.70	243.75	Very sparse
T ₁₂	Paddy straw extract agar	52.50	122.50	Moderately sparse
T ₁₃	Soil extract agar	87.20	122.00	Very sparse
T ₁₄	Compost extract agar	90.00	103.50	Moderately sparse
	SE(m)±	0.95	2.78	
	CD (0.05)	2.73	7.96	

*Mean of 4 replications.

ml of media from each different carbon sources was poured in four sterilized petridishes. 40 ml of each liquid media was prepared in 100 ml conical flask without adding agar. The medium was autoclaved at 15 psi pressure for 20 minutes. 5 mm discs of 10 days old pure culture of *V. volvacea* were cut with the help of cork borer and inoculated into each solid and liquid media and incubated at $34\pm1^\circ\text{C}$ for 7 days. Four replications were maintained.

Effect of different nitrogen sources on mycelial growth of *Volvariella volvacea*

Seven nitrogen sources namely sodium nitrate, potassium nitrate, ammonium phosphate, ammonium ferrous sulphate, ammonium chloride, glycine and peptone were taken to optimize the growth of *Volvariella volvacea* keeping Sabouraud's dextrose medium as the basal medium. As Sabouraud's medium found superior than other synthetic medium, we have taken Sabouraud's medium as basal medium. 20 ml of media from each different nitrogen sources was poured in four sterilized petridishes. 40 ml of each liquid media was prepared in 100 ml conical flask without adding agar. The medium was autoclaved at 15 psi pressure for 20 minutes. 5 mm discs of 10 days old pure culture of *V. volvacea* were cut with the help of cork borer and inoculated into each solid and liquid media and incubated at $34\pm1^\circ\text{C}$ for 7 days. Four replications were maintained.

RESULTS AND DISCUSSION

Effect of different culture media on mycelial growth of *Volvariella volvacea*

From the diameter growth it has been revealed that Potato dextrose agar (90 mm), Yeast extract agar (90 mm) and oat meal agar (90 mm) were found suitable as they produce dense mycelium and cover the whole petriplate on 7th day of inoculation. Maximum dry weight of mycelium was obtained on Malt extract agar medium (449.25 mg) followed by Oat meal agar (328.25 mg), Yeast malt agar (307 mg) and Glucose peptone agar (303.75) (Table 1).

In support of our observations, Sivani Sharma *et al.* (2019) while studying the growth of fungal mycelium in

different liquid media reported that on an average malt extract broth was recorded to support significantly maximum dry weight of the test fungus followed by potato dextrose broth and sweet potato dextrose broth while, average minimum dry weight of the fungus was observed in Czapek's dox broth followed by oat meal broth irrespective of the different strains used and days after inoculation. Similarly, observation on solid media reported by Shivani

Table 2: Growth of *Volvariella volvacea* under different pH levels.

Treatments	pH	Radial growth of mycelium 7 DAI (mm)*	Dry weight of mycelium 10 DAI (mg)*
T ₁	2.0	10.55	165.25
T ₂	3.0	32.15	180.75
T ₃	4.0	40.85	191.75
T ₄	5.0	84.53	227.25
T ₅	6.0	85.03	235.00
T ₆	7.0	90.00	284.75
T ₇	8.0	85.13	236.00
T ₈	9.0	63.23	212.50
SE(m)±		0.36	2.20
CD (0.05)		0.88	5.35

*Mean of 4 replications.

Table 3: Growth of *Volvariella volvacea* under different temperatures.

Treatments	Temperature (°C)	Radial growth of mycelium 7 DAI (mm)*	Dry weight of mycelium 10 DAI (mg)*
T ₁	20	22.50	61.00
T ₂	25	37.00	178.00
T ₃	30	71.00	266.50
T ₄	35	90.00	555.75
T ₅	40	55.00	143.75
T ₆	45	0.00	0.00
SE(m)±		0.84	2.53
CD (0.05)		2.09	6.27

*Mean of 4 replications.

Table 4: Growth of *Volvariella volvacea* using different carbon sources.

Treatments	Carbon sources	Radial growth of mycelium 5 DAI (mm)*	Dry weight of mycelium 10 DAI (mg)*	Density of mycelium
T ₁	Glucose	66.63	273.75	Dense
T ₂	Fructose	38.13	28.00	Dense
T ₃	Maltose	90.00	292.50	Dense
T ₄	Starch	90.00	295.00	Dense
T ₅	Lactose	36.88	177.50	Dense
T ₆	Sucrose	90.00	322.50	Dense
T ₇	Mannitol	84.68	261.25	Dense
T ₈	Control	25.13	105.00	Sparse
SE(m)±		1.24	5.52	
CD (0.05)		3.02	13.44	

*Mean of 4 replications.

sharma *et al.*, 2019 that on an average malt extract agar medium was observed to support the maximum average diametric growth of the fungus significantly followed by sweet potato dextrose agar and oat meal agar while, the minimum average diametric growth was recorded in Czapek's dox agar followed by potato dextrose agar.

Effect of different pH on mycelial growth of *Volvariella volvacea*

Maximum radial growth of the fungus was achieved at pH 7 (90 mm), followed by pH 8, 6 and 5 with radial diameter growth of 85.13 mm, 85.03 mm and 84.53 mm respectively. Maximum dry weight of mycelium was obtained at pH 7 with a dry weight of 284.75 mg followed by pH 7 (284.75 mg), 8 (236 mg), 6 (235 mg), 5 (227.25 mg) and 9 (212.50 mg) (Table 2).

In support of our observations, Shivani sharma *et al.* (2019) studied the effect of different pH regimes. It is clear from the observation that maximum average diametric growth was recorded at pH 7.0 significantly followed by pH level 8.0 which was statistically at par with the growth at pH 9.0 and pH 6.0 while, significantly minimum average diametric growth was recorded at pH 5.0, irrespective of different strains and hours of incubation.

Akinyele and Adetuyi (2005) and Kumar *et al.* (2016) also revealed that, pH 7.0 was found to be the best for the mycelial growth of *V. volvacea* as it supported the maximum growth of the test fungus followed by pH 8.0.

Effect of different temperatures on mycelial growth of *Volvariella volvacea*

Out of the total six temperatures evaluated to know the effectiveness of each, highest radial growth (90 mm) was found to be achieved at temperature 35°C followed by 30°C and 40°C with radial diameter growth of 71 mm and 55 mm respectively. Maximum dry weight of mycelium (555.75mg) was obtained from the growth under same temp (35°C) as in case of solid media followed by 30°C (266.5 mg), 25°C (178 mg) and 40°C (143.75 mg) (Table 3).

Similar observations has been reported by Rukhsana *et al.* (2013) who studied the influence of temperature on *Volvariella volvacea* at 25°C, 30°C and 35°C by keeping

pH constant (7.5). They resulted that the fungus can grow on all the temperature tested but the maximum radial growth 82.3 mm was observed at 35°C. It is followed by 30°C and 25°C at which 71 and 60 mm growth was observed, respectively. In partial agreement with our observations Sivani Sharma *et al.* (2019) while studying cultural characteristic of *Volvariella* spp reported that average diametric growth was recorded at 30°C followed by growth at 35°C, 40°C and 25°C irrespective of the strains and time of incubation under study.

Effect of different carbon sources on mycelial growth of *Volvariella volvacea*

Among the seven different carbon sources evaluated against our test fungus, it has been revealed that maltose, starch and sucrose recorded maximum radial diameter growth on solid base medium with an average diameter growth of 90mm each. Sucrose medium produce maximum dry weight of the test fungus after 10 days of inoculation (322.5 mg) (Table 4).

In partial agreement with our observations Rukhsana *et al.* (2013) evaluated five carbon compounds viz. glucose 13.5 g, sucrose 12.5 g, starch 12.5 g, maltose 13.5 g and mannitol 13.5 g amended in Sabouroud' s agar medium for the growth of *Volvariella volvacea* and reported that among the carbon sources, glucose considerably supported the growth of fungus with radial growth of 82.7 mm and regarded as the best carbon source. Starch and maltose also showed momentous effect on fungus with radial growth 72.3 and 74.3 mm, respectively. Sucrose has a least effect among all carbon sources having radial growth 21 mm.

Effect of different nitrogen sources on mycelial growth of *Volvariella volvacea*

Among the seven different nitrogen sources evaluated against the test fungus *Volvariella volvacea*, peptone gives highest radial growth with an average diameter of 90 mm. Same peptone recorded maximum dry weight of the mycelial mat of the test fungus (400 mg) (Table 5).

Similar observations has been reported by Rukhsana *et al.* (2013) who evaluated three nitrogen sources for the

Table 5: Growth of *Volvariella volvacea* using different nitrogen sources.

Treatments	Nitrogen sources	Radial growth of mycelium 7 DAI (mm)*	Dry weight of mycelium 10 DAI (mg)*
T ₁	Sodium nitrate	0.00	62.65
T ₂	Potassium nitrate	57.50 (sparse growth)	38.00
T ₃	Ammonium phosphate	0.00	117.50
T ₄	Ammonium ferrous sulphate	0.00	37.50
T ₅	Ammonium chloride	0.00	40.00
T ₆	Glycine	48.25 (sparse growth)	32.50
T ₇	Peptone	90.00	400.00
T ₈	Control	81.75 (sparse growth)	73.25
	SE(m)±	1.54	2.14
	CD (0.05)	3.75	5.22

*Mean of 4 replications.

growth of *Volvariella volvacea* viz. Potassium nitrate 10 g, sodium nitrate 8.5 g, urea 8.5 g and peptone 2.5 g individually in Sabouroud's agar medium. The results of the experiment determined that the peptone supports the best growth of the fungus among all the tested nitrogen sources. KNO_3 and NaNO_3 , are the inorganic nitrogen sources, represent the moderate radial growth 56.8 and 48.2 respectively, Whereas urea act as a least effective one having radial growth 27.7 mm.

CONCLUSION

From the above experiment it can be concluded that Potato dextrose agar (90 mm), Yeast extract agar (90 mm) and oat meal agar (90 mm) were found suitable as they produce dense mycelium and cover the whole petriplate on 7th day of inoculation. From the various liquid media studied it has been found that dense mycelium was obtained on Malt extract agar medium (449.25 mg). Maximum radial growth of the fungus was achieved at pH 7 and at 35°C temp (90 mm). Maltose, starch and sucrose recorded as best carbon sources for *V. volvacea* growth on solid base medium with an average diameter growth of 90mm and sucrose proved to produce dense mycelium on liquid media. Peptone was found to be best nitrogen source as it gives highest radial growth of the test fungus with an average diameter of 90 mm and also produced dense mycelial mat of the test fungus (400 mg). The findings will be helpful for commercial or large-scale production without loss due to environmental conditions as in Eastern region the climate is very much variable throughout the year.

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Conflict of interest

All authors declared that there is no conflict of interest.

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