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Comparative Effect of Different Culture Media on the Submerged Mycelial Biomass Production of *Hericium erinaceus*

SAKSHI KHURANA, ARVIND, VIKASH KUMAR[1], JAGDEEP SINGH[2] AND ANIL SINDHU*

Department of Biotechnology, Deenbandhu Chhotu Ram University of Science and Technology, Murthal, Sonepat-131 039 (Haryana), India
**(e-mail : Sindhu.biotech@gmail.com; Mobile : 94683 49982)*

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ABSTRACT

Medicinal edible mushroom, *Hericium erinaceus* (Lion's mane) includes several key bioactive metabolites that have been linked to a variety of health benefits and have been employed in nutraceuticals and pro-health products. For maximum biomass production of mushroom, commonly used different culture media such as : Sabouraud's Dextrose Broth (SBD), Potato Dextrose Broth (PDB), modified Potato Dextrose Broth (mPDB) and Basic Medium Broth (BMB) were studied with Malt Extract Broth medium as control. Among the most commonly used mushroom growing media, mPDB was most favourable media with the maximum mycelium fresh weight biomass of 373 g/l. The findings are encouraging as increased production of mycelium biomass has significant medicinal and pharmacological value.

Key words : *Hericium erinaceus,* culture medium, modified potato dextrose broth

INTRODUCTION

Mushrooms are popular all around the world because of their delicate flavour and health-giving characteristics. They are high in unsaturated protein, fatty acids, minerals and vitamins. These are high in dietary fibre and low in fat, carbs and salt. Furthermore, because mushrooms have a low nucleic acid content, they are regarded an ideal diet for people suffering from obesity, hypertension and diabetes (Chaturvedi *et al.*, 2018; Berovic *et al.*, 2022). *Hercium erinaceus*, a mushroom that is both edible and medicinal, has been used in traditional Chinese medicine and culinary traditions since the beginning of time (Liu *et al.*, 2021). Terpenoids, phenolics, steroids, pyranones, fatty acids and alkaloids are among the chemical constituents of *H. erinaceus.* According to research, about 80 small molecular compounds have been isolated and identified from *H. erinaceus* (Atila *et al.*, 2021). However, a growing body of evidence suggests that the active compounds in *H. erinaceus* have a variety of pharmacological effects, including cardiovascular, antitumor, neuroprotective, immunomodulatory and hepatoprotective properties (Chong *et al.*, 2020; Lew *et al.*, 2020; Liu *et al.*, 2021). Coronary heart disease, arteriosclerosis, alzheimer's disease, arthritis, parkinson's disease, hepatitis, nerve pain, hypertension, cancer, gastric ulcer and obesity are among the modern uses of *H. erinaceus* (Wong *et al.*, 2015; Chen *et al.*, 2016; Bailly and Gao, 2020; Lee *et al.*, 2020; Turk *et al.*, 2021). Its potential in the treatment of viral infection has been highlighted in new research. It's known for its balancing abilities (Lu *et al.*, 2016; Li *et al.*, 2018; Lee *et al.*, 2020). According to the most recent estimates, the global value of *H. erinaceus* products exceeds $ 1 billion per year. Because of the increased demand, artificial cultivation methods like liquid spawn have improved. Spawning can also be accomplished by cultivating mycelium on liquid media and then homogenizing it and it is referred to as liquid spawn. It may be used to automate the spawn multiplication inoculation process or to inoculate substrates. The use of liquid culture technologies to the generation of mycelia of higher fungi opens the door to industrial-scale applications for this

[1]Regional Mushroom Research Center (Maharana Pratap Horticultural University), Murthal, Sonepat-131 039 (Haryana), India.
[2]Department of Plant Pathology, College of Agriculture, Chaudhary Charan Singh Haryana Agricultural University, Hisar-125 004 (Haryana), India.
Presently at Department of MBBB, Chaudhary Charan Singh Haryana Agricultural University, Hisar-125 004 (Haryana), India.

group of organisms. The following are some of the benefits of applying liquid culture technology to the spawn production process : increased process control (growth rates and nutritional content), increased automation in the spawn plant, shorter production cycle times, more uniform inoculums distribution in the substrate, and inoculation of the substrate under more stringent aseptic conditions. Therefore, the aim of the present study was to maximize the mycelium biomass production by evaluating the culture media which were most commonly utilizing in the mushroom cultivation.

MATERIALS AND METHODS

The isolate He 204 of *H. erinaceus* used in present study was collected from the Regional Research Center, Murthal (Sonipat) of Maharana Pratap Horticultural University (MHU), Karnal (Haryana).The strain was transferred to potato dextrose agar (PDA) plates and stored at 4°C for future use.

To investigate vegetative development potato dextrose broth, modified potato dextrose broth, sabouraud dextrose broth, malt extract broth and basic medium broth were used. The media compositions used in this study are presented in Table 1. The media were autoclaved at 121°C with 15-pound square inch (psi) pressure for one hour after being poured 150 ml of media into a 300 ml flask.

The pH of each medium was set at pH 6 for that 250 ml stock solution of 1N HCl acid and 1N NaOH base was prepared. Autoclaving raised the pH of the media due to the loss of CO_2. Use of a pH meter with an unsterilized electrode to adjust the pH after autoclaving was a risk of contamination in the culture. In order to solve these issues, the media for the pH 6 treatment were made in a 300 ml flask with acid and base stock solutions, autoclaved, and the pH was measured; the pH was increased to 6.6. Now 150 ml of fresh media with pH treatments of 5.4 was prepared so that the pH rose to the desired pH of 6 after autoclaving. After autoclaving, the treatment media were poured into a separate conical flask, and the final pH was recorded at 6.1, which was almost the desired pH with a minor increase of 0.1 from the desired pH. The remaining media were used in the experiment. Five replications of a 5 mm mycelial disc of strain He 204 were inoculated in the medium flask and cultured at 25°C in the BOD incubator with 150 rpm. Pure culture discs were extracted from a 25-day old prior culture. Different media were compared to malt extract broth as a percentage increase or decrease in vegetative growth relative to control.

Five replications were used to test the studies. The experiments were subjected to analysis of variance (ANOVA) for statistical evaluation, and the averages were compared using the Tukey HSD (honestly significant difference) test at a significance level of 5%.

RESULTS AND DISCUSSION

Five different media were tested to examine how they affected *H. erinaceus* growth and biomass (strains He 204). The outcomes obtained are depicted in Fig. 1. Different media exhibited substantial differences in their growth and fresh mycelia weight of He 204. On modified potato dextrose broth medium, growth was significantly higher, followed by potato dextrose broth and basic medium broth, which differed significantly from one another. After 14 days of inoculation, modified potato dextrose broth had the highest fungal growth (59.612 g/150 ml) of all the media tested. Sabouraud dextrose broth had the lowest

Table 1. Composition of media used for the growth of *H. erinaceus* He 204

S. No.	Contents of media	PDB	mPDB	BMB	SDB	MEB
1.	Potato dextrose broth	24 g	24 g	-	-	-
2.	Malt extract	-	-	-	-	30 g
3.	Peptone	-	5 g	3 g	-	-
4.	Glucose	-	-	15 g	-	-
5.	Yeast extract	-	-	5 g	-	-
6.	Dipotassium phosphate	-	0.5 g	1 g	-	-
7.	Magnesium sulphate	-	0.5 g	0.5 g	-	-
8.	Monopotassium phosphate	-	0.5 g	-	-	-
9.	Sabouraud dextrose [special peptone + dextrose (1 : 2)]	-	-	-	30 g	-
10.	Distilled water	1 L	1 L	1 L	1 L	1 L

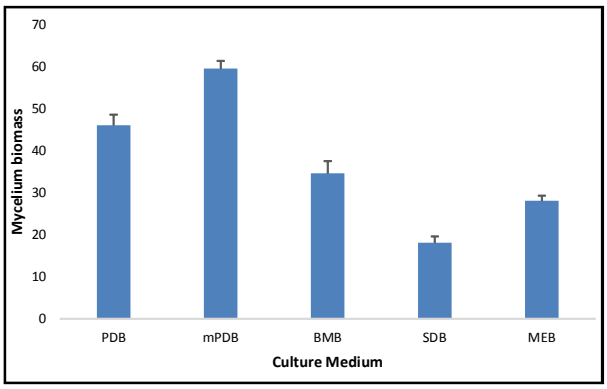

Fig. 1. Effect of culture medium on the mycelium biomass production of *H. erinaceus.*

concentration after 14 days (18.108 g/150 ml). On the other hand, potato dextrose broth showed better growth medium (46.128 g/150 ml) for *H. erinaceus* as compared to basic medium broth (34.652 g/150 ml) and malt extract medium (28.136 g/150 ml). In the study, mPDA and PDA were determined to be the best for *H. erinaceus* mycelial development, which was consistent with the findings of Julian *et al.* (2018) who demonstrated the superiority of PDA for *H. erinaceus* growth and biomass. Cohen *et al.* (2014) found that *H. erinaceus* grew more quickly on PDA. Gang *et al.* (2016) reported that 3% glucose, 3% peptone, 0.1% $MgSO_4$, and 0.2% KH_2PO_4 greatly improved mycelium development.

CONCLUSION

The findings of this study provided valuable insight and knowledge into the growth rate and productivity of *H. erinaceus* He 204. The present study concluded that use of modified potato dextrose broth as a medium appeared to be the most suitable for large-scale culture establishment of *H. erinaceus* mushroom due to increased mycelial growth.

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