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Peptone Supplementation of Potato Dextrose Agar Medium Proved Better for Mushroom Mycelial Development

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ABSTRACT

An experiment was conducted to assess the suitability of different media on mycelial development of five different mushrooms in College of Agriculture, Vellayani, Kerala. The medium used was potato dextrose agar (PDA), carrot dextrose agar (CDA), yeast malt agar (YMA), malt extract agar (MEA) and potato dextrose peptone agar (PDPA) and study was conducted using *Pleurotus florida, Hypsizygous ulmerius, Calocybe indica, Agaricus bitorquis, Volvariella volvaceae*. Amongst various media used PDPA was found best in enhancing the mycelial coverage of *P. florida* (9 cm on 6th day) followed by *H. ulmerius* (8.96 cm on 6th day) and *V. volvaceae* (8.93 cm on 6th day) in 9 cm petri plates. Peptone supplementation of the PDA media have a pronounced influence on accelerating the mycelia spread of these mushrooms. PDPA was found least effective in mycelial development of *C. indica* which showed its inhibitory effect. At par with PDPA, MEA was also found effective in development of mycelia of *V. volvaceae* (8.76 cm on 6th day). MEA turned out to be the best media for the growth of *A. bitorquis* (8.83 cm on 6th day). PDA media was most effective media for development of *C. indica* (8.63 cm on 6th day).

Key Words: Culture media, Mycelial growth, Mushrooms, Peptone supplemented media, Peptone.

INTRODUCTION

Mushroom bodies are much valued food items from time unmemorable and also gained importance as nutraceutical and pharmaceutical agent due to the ability of producing high protein content with essential amino acids, vitamins, minerals and exopolysaccharides and also numerous useful secondary metabolites, (Adebayo-Tayo *et al*, 2011; Zikriyani *et al*, 2018). Mushrooms are rich sources of nutrients, especially proteins, minerals and also vitamins B, C and D (Panjikkaran and Mathew, 2013; Bellettini *et al,* 2019). Mushrooms naturally grow on any semi-synthetic compost and absorb nutrients for their development and survival. The maintenance and revival of pure culture mycelium with magnificent quality is the first critical stage towards the success of spawn preparation (Sharma *et al*, 2019; Kumar *et al*, 2018). Like all other microbes, mushrooms also require a set of conditions under which they can grow and sporulate best in artificial conditions and culture medium is the crucial factor influencing fungal mycelial growth (Dhingra and Sinclair, 2014). Mycelial growth of mushrooms varies pronouncedly with each culture media. A small variation in the composition of the culture media will positively or negatively influence the mycelial development of mushrooms (Abon *et al*, 2020).

Curvetto *et al* (2002) and Mukhopadhyay *et al* (2002) reported that for the growth of any fungus including mushrooms, both quality and quantity aspects *viz.,* biological productivity and efficiency are much linked to the nutrient type and growth conditions. The mycelium branches

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Table 1. Effect of different media on mycelial development of *P.florida*

Media	Growth on 3rd day	Growth on 5th day	Growth on 6th day	Nature of Mycelia
PDA	4.233	7.900	8.733	++++
CDA	3.666	7.000	7.733	++++
YMA	3.000	6.700	7.033	++++
MEA	2.200	6.000	6.633	+++
PDPA	**4.433**	**8.066**	**9.000**	+++
CD Value	0.252	0.278	0.223	

Table 2. Effect of different media on mycelial development of *H. ulmerius*

Media	Growth on 3rd day	Growth on 5th day	Growth on 6th day	Nature of Mycelia
PDA	4.166	7.900	8.766	++++
CDA	3.933	7.600	8.500	++++
YMA	3.233	7.100	7.900	+++
MEA	2.566	6.666	7.166	++++
PDPA	**4.333**	**8.233**	**8.966**	**++++**
CD Value	0.157	0.196	0.149	

and produces enzymes that digest complex carbohydrates, lipids and protein, which in-turn will be easily absorbed by the developing hyphae (Yadav and Chandra, 2014). Mycelium growth may be considered as the best tool in identification of necessary nutrients for the production of fruiting bodies of many mushrooms as mycelium growth requires much short time in comparison with fruiting bodies development (Kalmis and Kalyoncu, 2006). Many researchers have been driven the use of different agar media as an effective culture platform that supports the mycelial growth of different mushroom species (Cañal *et al*, 2020). Potato Dextrose Agar medium is reported to be effective for the mycelial growth of many species of Oyster mushrooms (*Pleurotus* sp.) , Milky mushroom and Paddy straw mushroom (Dey *et al*, 2007; Neelam *et al*, 2013; Pant *et al*, 2020). Malt extract and peptone agar medium was reported as one of the preferable culture medium for *Agaricus bitorquis* (Ali *et al*, 2015). Peptones and extracts are excellent natural sources of amino acids, peptides and proteins in growth media (Davami *et al*, 2015). Keeping in view the importance of cultural media in the cultivation process of any edible mushroom, the present investigations were conducted with an objective to study the influence and suitability of different media on mycelial development of five different mushrooms.

MATERIALS AND METHODS

Mushroom cultures used

The pure culture of 5 edible mushrooms *viz., Pleurotus florida, Hypsizygous ulmerius, Calocybe indica, Agaricus bitorquis* and *Volvariella volvaceae* from the Department of Plant Pathology, College of Agriculture, Vellayani, Kerala was utilized for this study.

Media preparation

Potato Dextrose Agar (PDA) media: The medium was prepared by using 200 g peeled potato, 20g dextrose and 20g agar in a liter of water, *Carrot Dextrose Agar (CDA) media*: The medium was prepared by using 200g peeled and sliced carrot, 20g dextrose and 20g agar in a

Table 3. Effect of different media on mycelial development of *C. indica*

Media	Growth on 3rd day	Growth on 5th day	Growth on 6th day	Nature of Mycelia
PDA	**3.833**	**7.233**	**8.633**	+++
CDA	2.933	6.800	7.900	+++
YMA	2.800	6.533	7.466	+++
MEA	2.900	6.466	7.733	++++
PDPA	1.033	1.200	1.333	+
CD Value	0.149	0.192	0.238	

Table 4. Effect of different media on mycelial development of *A. bitorquis*

Media	Growth on 3rd day	Growth on 5th day	Growth on 6th day	Nature of Mycelia
PDA	0.533	0.533	0.533	+
CDA	0.633	0.633	0.633	+
YMA	1.166	2.833	3.000	++
MEA	**3.866**	**7.100**	**8.833**	++++
PDPA	1.933	3.666	4.100	++
CD Value	0.177	0.207	0.192	

liter of water, *Malt Extract Agar (MEA) media*: The medium was prepared by dissolving 20g malt extract and 20g agar in a litre of water,

Yeast Malt Agar (YMA) media: The medium was prepared by dissolving 10g dextrose, 5g peptone, 3g malt extract, 3g yeast extract and 20g agar in a liter of water, *Potato Dextrose Peptone Agar (PDPA) media*: The normal PDA medium was modified by adding peptone. *i.e.,* the medium was prepared by using 200g peeled potato, 20g dextrose, 5g peptone and 20g agar in a liter of water.

Sterilization of medium

The above five media prepared were sterilized in an autoclave at 1.05 kg/cm^2 for 20 min and then poured into 90 mm petri dishes inside the laminar airflow chamber. Media were cooled and solidified to room temperature at 30$\pm$2^o C.

The petri dishes were inoculated with 5 mm mycelial bit of actively growing pure cultures (10d old) of *P. florida, H. ulmerius, C. indica, A. bitorquis* and *V. volvaceae* taken using sterile cork-borer. The plates were incubated at 30$\pm$2^o

Table 5. Effect of different media on mycelial development of *V. volvaceae*

Media	Growth on 3rd day	Growth on 5th day	Growth on 6th day	Nature of Mycelia
PDA	3.800	7.500	8.066	+++
CDA	2.166	6.466	7.033	+
YMA	3.800	7.800	8.166	+++
MEA	**3.966**	**7.866**	**8.766**	+++
PDPA	**3.966**	**8.033**	**8.933**	++++
CD Value	0.212	0.192	0.259	

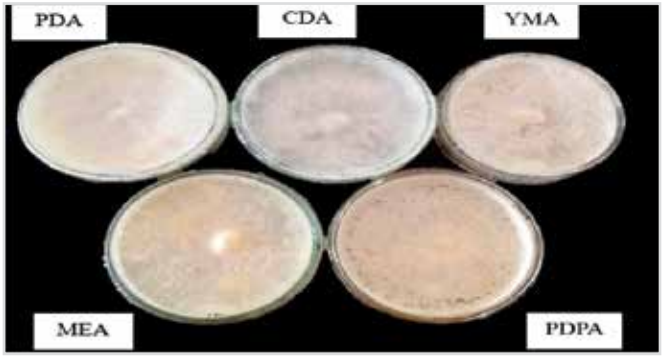
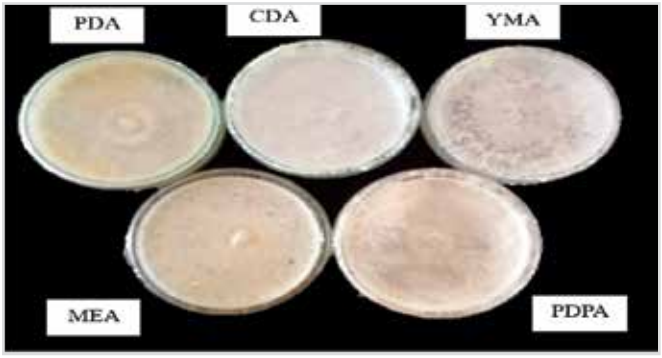


Fig. 1. Growth on 6th day of P. florida on different media



Graph 1. Effect of different media on mycelial development of P. florida

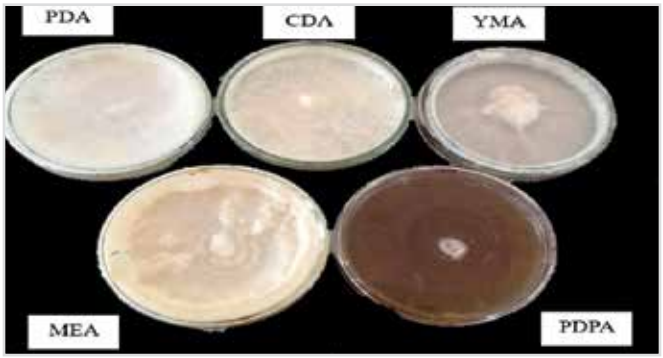
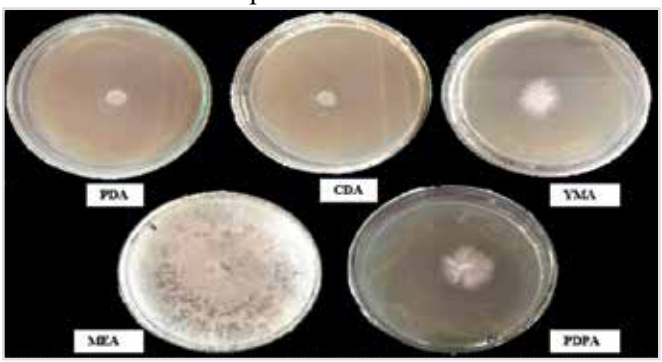
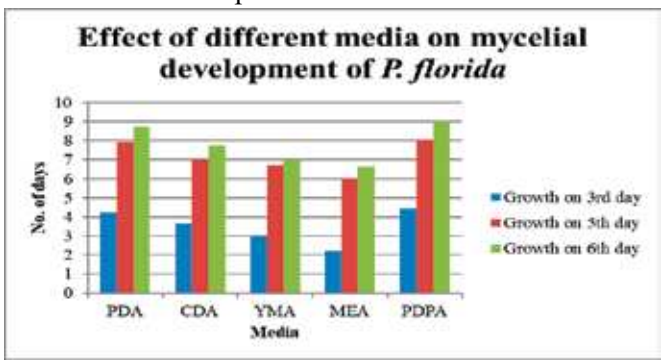


Fig. 2. Growth on 6th day of H. ulmerius on different media



Graph 2. Effect of different media on mycelial development of H. ulmerius

Fig. 3. Growth on 6th day of C. indica on different media



Graph 3. Effect of different media on mycelial development of C. indica

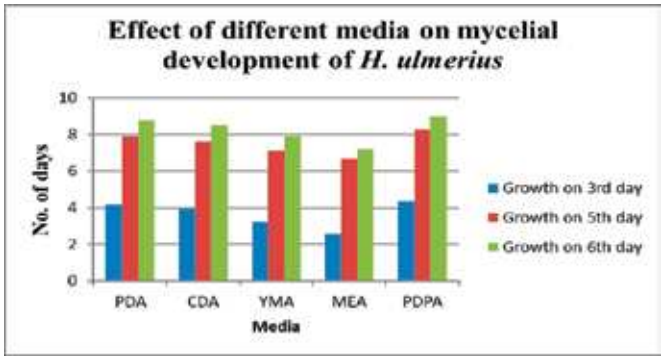
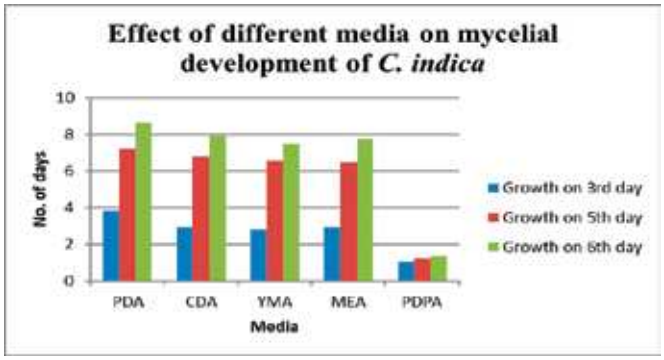


Fig. 4. Growth on 6th day of A. bitorquis on different media



Graph 4. Effect of different media on mycelial development of A. bitorquis

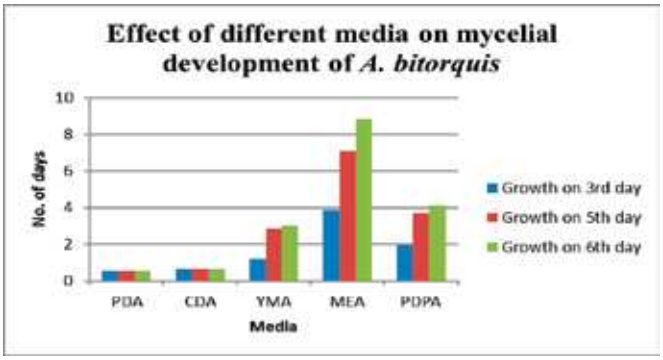
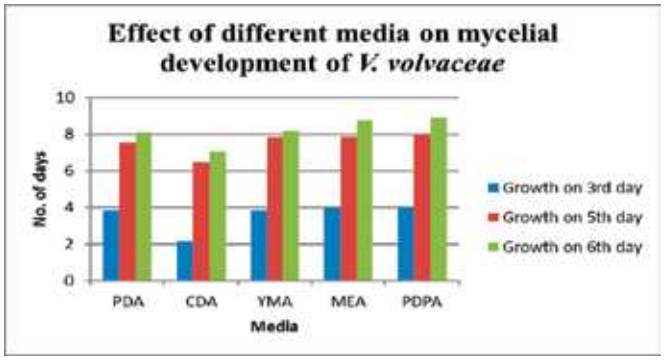


Fig. 5. Growth on 6th day of V. volvaceae on different media



Graph 5. Effect of different media on mycelial development of V. volvaceae

C. The radial mycelial growth and mycelial density were observed and recorded continuously.

Daily mycelial growth was measured using a ruler placed across the petri dish horizontally. The mycelial density was rated as follows (Kadiri, 1998):

+ = Very Scanty mycelial density

++ = Scanty mycelial density

+++ = Moderate mycelial density

++++ = Abundant mycelial density

+++++ = Very abundant mycelial density.

The experiment was conducted in a completely randomized design (CRD) with three replications for each treatment under in vitro conditions and the data were analyzed by using statistical package of program WASP2.0. Critical difference was calculated at 5 per cent probability.

RESULTS AND DISCUSSION

It was found that PDPA media was best in fastening the mycelial coverage of *P. florida* (9 cm on 6th day) followed by *H. ulmerius* (8.96 cm on 6th day) and *V. volvaceae* (8.93 cm on 6th day) in the petri plates. But the mycelial density of *P. florida* was moderate in PDPA medium while it was abundant in PDA, CDA and YMA media. *H. ulmerius* showed abundant mycelial density in PDA, CDA, MEA and PDPA while it showed only moderate mycelial density in YMA. Thus, animal nitrogen source peptone have a profound influence in the mycelial development of these mushrooms which is evident from the vigorous mycelial growth as well as from the abundantly developed mycelial density.

Among the five solid media used, PDPA was found as least effective in the mycelial development of *C. indica.* The mycelial density of *C. indica* on PDPA media was also scanty which showed its inhibitory effect. PDA media was the most effective media for development of *C. indica* (8.63 cm on 6th day) with moderate mycelial density. *C. indica* showed abundant mycelial density in MEA the mycelial development was not appreciable in MEA. On par with PDPA, MEA also founded effective in the development of mycelia of *V. volvaceae* (8.76 cm on 6th day). However, the mycelial density of *V. volvaceae* was found to be abundant in PDPA medium and only moderate in MEA medium. *V. volvaceae* also showed moderate mycelial density in PDA and YMA but showed very scanty mycelial density in CDA.

MEA turned out to be the best medium for the growth of *A. bitorquis* (8.83 cm on 6th day) with abundant mycelial density followed by PDPA. The mycological peptone present in the malt agar could have rapid influence on the development of luxuriant growth of the mycelia of *A. bitorquis.* The mycelial density of *A. bitorquis* was scanty in PDA and CDA and was scanty in YMA and PDPA.

CONCLUSION

The peptone supplemented potato dextrose agar (PDPA) medium was found more effective in mushroom mycelial development in the present study. Peptone supplementation of the PDA media had shown a pronounced influence on accelerating the mycelial spread as well as the mycelial density of *P. florida*, *H. ulmerius* and *V. volvaceae.* In the case of *C. indica,* PDPA was not preferable as it has an inhibitory effect. MEA turned out to be the best media for the mycelial growth of *A. bitorquis* and *V. volvaceae.* Since mushrooms are good source of various bioactive compounds of industrial and therapeutic importance also, the in-vitro developed mycelia may be used for the large scale production of the compounds as most mushrooms are seasonal. The present study may be useful in order to make the bioactive production technology from the mycelia and to obtain high cost effectivity.

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