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EFFECT OF FIVE DIFFERENT CULTURE MEDIA ON MYCELIAL GROWTH OF *AGROCYBE AEGERITA*

Nagalakshmi Muthu and Krishnakumari Shanmugasundaram*

Department of Biochemistry, Kongunadu arts and science college (Autonomous), Coimbatore – 641 029, Tamil Nadu, India.

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Correspondence to Author:

Dr. S. Krishnakumari

Associate Professor,
Department of Biochemistry,
Kongunadu Arts and Science College,
Coimbatore – 641029, Tamil Nadu, India

E-mail: krishnashanmugambc@gmail.com


ABSTRACT: A preliminary experiment was carried out to analyse the growth performance of *Agrocyba aegerita* mushroom cultures using chemosynthetic media. This study is mainly aimed to different media such as Yeast Malt Agar Medium (YMA), Saboraud's Dextrose Agar Medium (SDA), Glucose Peptone Agar Medium (GPA), Malt Extract Agar Medium (MEA) and Potato Dextrose Agar Medium (PDA) on the growth of mycelium of the mushroom. Studies revealed that the joint portion of cap and stripe produced vigorous mycelium growth in minimum time; the average maximum growth was obtained on Malt Extract Agar (MEA) than Potato Dextrose agar (PDA) medium.

INTRODUCTION: Mushrooms have been recorded as a source of vegetable and medicines for human beings throughout the world. They are generally divided into edible and non-edible. Mushroom fruit bodies are well known food items since ancient times and became important as nutraceutical and pharmaceutical agent due to the capability of producing many useful secondary metabolites, high protein content with essential amino acids, vitamins, minerals and exopolysaccharides¹. *Agrocyba aegerita* mushroom with high economic, nutritional, and pharmaceutical value. Mushrooms have become attractive as a functional food and as source for the development of drugs and nutraceuticals^{2, 3} namely for antioxidant⁴⁻¹⁰ and antimicrobial compounds¹¹.

In nature mushrooms grow wild in every country from snowy mountains to sandy deserts on all types of soils, pastures, forests, cultivated fields or water lands. They appear in all seasons, chiefly during the rainy weather, wherever organic matter or its decomposition products are available¹². Many fungi and their mycelium biomass are reported as good source of food, protein supplement, lipid source and many more metabolites^{13, 14, 15}.

The media listed above, with the exception of compost agars, are all devoid of insoluble material and are rich in simple sugars, few of which are encountered by the mushroom on its 'natural' substrate. The insoluble fraction, which includes lignin's, cellulose, hemicelluloses, protein and microbial biomass, is preferentially used by the mushroom mycelium for growth¹⁶.

Mushrooms are amongst the most popular food items accepted the world over. The increased consumer demand over the years has resulted in production of mushrooms in large proportions¹⁷ through cultivation which is a highly efficient

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method for recycling the agricultural residues so as to produce nutritious food¹⁸.

Today China is the major producer of mushroom in the world¹⁹. In the present study, stock culture of vigorous growing mycelia of *Agrocyba aegerita*, was cultivated on different media (i.e.) YMA, PDA, SDA, GPA and MEA and their mycelial growth rate was determined. The purpose of the present study was to determine the most suitable media, for the growth of the mushroom.

MATERIAL AND METHODS:

Collection and Maintenance of *A. Aegerita* Culture:

The pure culture of *A. aegerita* was obtained from the Directorate of Mushroom Research (DMR), Chambaghat, Solan, Himachal Pradesh and were used for mass culture production.

Media preparation:

Following media were used for the purpose:

PDA- Potato Dextrose Agar Medium:

Potato dextrose agar - 39g
Water - 1000ml

MEA - Malt Extract Agar Medium:

Malt extract - 30 g
Agar-Agar - 15 g
Water - 1000 ml

GPA - Glucose Peptone Agar Medium:

Peptone - 20 g
Dextrose - 10 g
Nacl - 5 g
Agar-Agar - 15 g
Water - 1000 ml

YMA - Yeast Malt Agar Medium:

Malt - 20 g
Yeast - 2 g
Agar-Agar - 15 g
Water - 1000 ml

SDA - Saboraud's Dextrose Agar Medium:

Dextrose - 40 g

Agar-Agar - 15 g
Peptone - 10 g
Water - 1000 ml

All five media were prepared according to above mentioned composition.

Sterilization of medium:

The flasks having media were sterilized in the autoclave at 15lb/sq. inch pressure for one hour and then poured in 90 mm Petri dishes under the laminar flow hood to avoid contamination. Media were cooled to 37°C. The joint, stalk and veil of the fresh mushrooms were inoculated on culture media.. Radial growth of mycelium of different portions was observed until the Petri dishes were filled with it. The experiment was repeated for 5 times. The plates was incubated at 37°C and observed for 15 days during which the mycelial vegetative growth and mycelial density of *Agrocyba aegerita* were recorded.

The mycelial density was rated as described²⁰ as follows:

+ = Very Scanty mycelial density
2+ = Scanty mycelial density
3+ = Moderate mycelial density
4+ = Abundant mycelial density
5+ = Very abundant mycelial density

The growth rate is given by the formula below:

Growth rate = Colony diameter on the last day (cm) / Number of day's measurement was taken after inoculation

Daily mycelial growth was determined using a ruler across the Petri-dish horizontally.

RESULTS AND DISCUSSION:

The result in (**Fig; Table 1** and **2**) on the 15th day showed that *Agrocyba aegerita* had the highest mycelial colony diameter 8.82 ± 0.27 cm, density (5+) and growth rate (1.5cm/day) on MEA media followed by PDA media with 8.00 ± 0.86 cm, 5+ and 1.3 cm/day being colony diameter, mycelial density and growth rate respectively.

On YMA media colony diameter was 7.76 ± 1.46 cm, mycelial density 5+ and growth rate was

1.3 cm/day. Furthermore, the growth on GPA was poor with 6.20 ± 0.38 cm as colony diameter, 3+ as mycelial density and 1.0 cm/day as growth rate. The least growth was recorded on SDA media

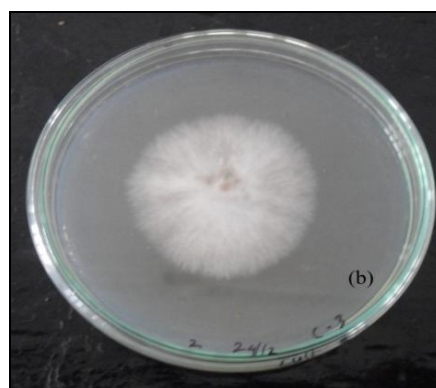
TABLE 1: MYCELIAL DENSITY AND GROWTH RATE OF AGROCYBA AEGERITA IN DIFFERENT MEDIUM

| S. No | Medium | Average Mycelial Density On 15 th Day Of Incubation | Growth Rate cm/day |
|-------|--------------------------|--|--------------------|
| 1. | Saboraud's dextrose agar | 3 + | 1.0 |
| 2. | Potato dextrose agar | 5 + | 1.3 |
| 3. | Yeast malt agar | 5 + | 1.3 |
| 4. | Glucose peptone agar | 4 + | 1.1 |
| 5. | Malt extract agar | 5 + | 1.5 |

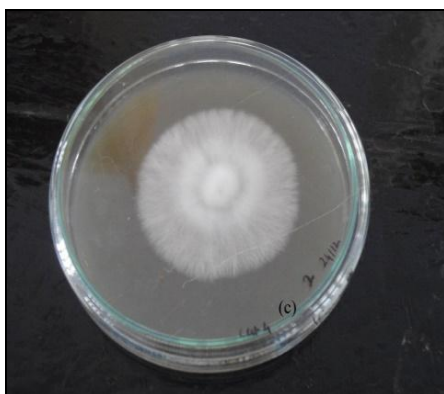
3+ = Moderate, 4+ = Abundant, 5+ = Very abundant



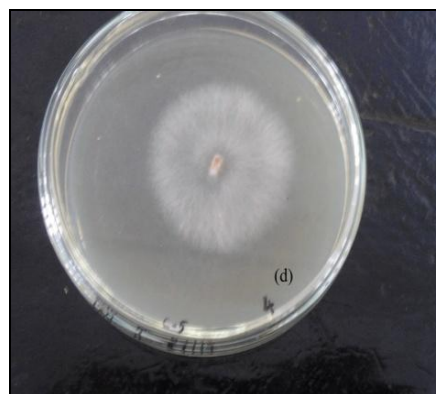
(a) Saboraud's dextrose agar



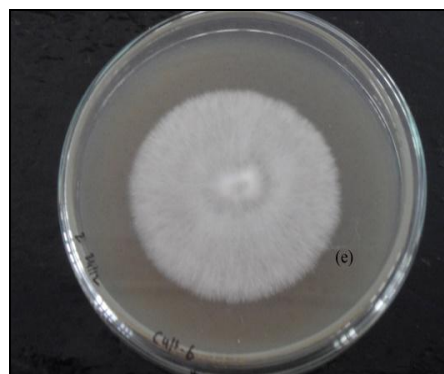
(b) Potato dextrose agar



(c) Yeast malt agar



(d) Glucose peptone agar



(e) Malt extract agar

FIG.1: MYCELIAL GROWTH IN DIFFERENT MEDIA

TABLE 2: MYCELIAL GROWTH PATTERN OF AGROCYBA AEGERITA IN DIFFERENT MEDIUM

| S. No | Medium | Colony diameter in different days (in cm) | | | | | | | | | |
|-------|--------------------------|---|-----------------|-----------------|-----------------|------------------|------------------|------------------|------------------|------------------|------------------|
| | | 6 th | 7 th | 8 th | 9 th | 10 th | 11 th | 12 th | 13 th | 14 th | 15 th |
| 1. | Saboraud's dextrose agar | 0.58 ±0.19 | 1.22 ±0.40 | 2.06 ± 0.39 | 3.30 ± 0.27 | 3.60 ± 0.29 | 3.90 ± 0.31 | 4.40 ± 0.30 | 4.90 ± 0.31 | 5.50 ± 0.24 | 6.20 ± 0.38 |
| 2. | Potato dextrose agar | 0.92 ± 0.19 | 1.72 ± 0.36 | 2.32 ± 0.31 | 2.89 ± 0.27 | 3.80 ± 0.25 | 4.70 ± 0.34 | 5.46 ± 0.38 | 6.24 ± 0.24 | 6.94 ± 0.09 | 8.00 ± 0.86 |
| 3. | Yeast malt agar | 0.74 ± 0.36 | 1.30 ± 0.26 | 1.78 ± 0.31 | 2.66 ± 0.60 | 3.56 ± 0.95 | 4.54 ± 1.44 | 5.44 ± 1.66 | 6.44 ± 1.67 | 7.14 ± 1.56 | 7.76 ± 1.46 |
| 4. | Glucose peptone agar | 0.80 ± 0.23 | 1.84 ± 0.21 | 2.36 ± 0.21 | 3.00 ± 0.15 | 3.54 ± 0.21 | 4.04 ± 0.21 | 4.66 ± 0.32 | 5.40 ± 0.37 | 6.16 ± 0.29 | 6.78 ± 0.11 |
| 5. | Malt extract agar | 0.90 ± 0.16 | 1.78 ± 0.19 | 2.98 ± 0.46 | 4.14 ± 0.46 | 4.84 ± 0.27 | 5.64 ± 0.34 | 6.50 ± 0.34 | 7.36 ± 0.31 | 8.34 ± 0.38 | 8.82 ± 0.27 |

Among the five solid growth media, malt extract agar media found to support fastidious growth of mushroom cultures with the presence of higher amount of carbohydrates particularly maltose, protein and other medium. This is followed by potato dextrose agar medium, yeast malt agar medium, saboraud's dextrose agar medium and glucose peptone agar medium. Though the mycelial diameter of the mushroom cultures in saboraud's dextrose agar medium, glucose peptone agar medium were in par with rest of the growth media, the density of the mycelia were low in these two growth media.

Hence, malt extract agar medium was found to be an ideal media for growth of these mushroom cultures. Mycological peptone present in the malt agar rapidly gives a luxuriant growth with typical morphology and pigmentation.

Mycelium is an important part for mushroom production as well as for production of several secondary metabolites used for therapeutic purpose. The mycelium growth depends on several factors such as growth media, pH, temperature, nutrient elements and some environmental factors ²¹. Growth medium is the most important factor because it supplies necessary nutrient for the growth of mushroom mycelium.

Different media such as potato dextrose agar, yeast extract agar, malt extract agar, lamberts agar and compost extract agar are mostly used for the growth of mycelium ²². Mycelium growth is the best tool to identify necessary nutrients for the production of fruiting bodies as mycelium growth requires short time in comparison with fruiting bodies development ²³.

CONCLUSION: Since mushrooms are good source of bioactive compounds of anticancer, antifungal and anti-diabetic in nature, the mycelia may be used for the large scale production of the compounds as mushrooms are seasonal. To make the bioactive production technology cost effective, present study may be useful in order to obtain more biomass ultimately to have bioactive compounds in hand. Further standardization regarding quantification of substrate as nutritional source for biomass production and its cost economics is required to reach more constructive conclusion

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