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STUDIES ON GROWTH CONDITIONS OF WILD EDIBLE MUSHROOM *ARMILLARIELLA MELLEA* (VAHL EX. FR.) KUMM. FUHR. SELECTED FROM NORTH WEST HIMALAYAN REGION

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Abstract
Armillariella mellea, commonly known as honey fungus, is a basidiomycete fungus. The fruit body commonly known as stump mushroom, stumpie, honey mushroom, pinky, grows typically on hardwoods but may be found around and on other living and dead wood or in open areas. Armillariella mallea 'the Honey Mushroom' is particularly good when saute in butter or mixed with scrambled eggs. Armillaria can be a destructive forest pathogen. It causes "white rot" root disease of forests. Their caps (mushroom tops) are typically yellow-brown, somewhat sticky to touch when moist, and, depending on age, may range in shape from conical to convex to depressed in the center. The stipe may or may not have a ring..Major factors essential for growing mycelium in laboratory are nutrients, temperature, pH and light and dark conditions. The impact of these factors on the growth of Armillariella mellea was investigated under laboratory conditions. The aim of the Armillariella mellea investigation was to determine optimal conditions for the development of the fungus. The results showed Yeastal Potato Dextrose Agar medium as best solid medium, Glucose-Asparagine as best liquid medium, optimal temperature was 25°C, whereas optimal pH was 6.0 under dark conditions.

Key Uses: the Honey Mushroom, basidiomycete, forest pathogen destructive,,Armillariella mellea .

Introduction

Mushrooms have been consumed since ages as a delicacy. Earlier these were collected from natural habitats and cooked either fresh or after drying. With time, cultivation techniques have been worked out for 25-30 edible species out of the 2000 naturally occurring edible mushrooms. Still, there are many more species which are consumed by local inhabitants which are yet to be cultivated.

Before cultivation is taken up of any mushroom it is essential to bring the mushroom into pure culture and to study its nutritional requirements. The present study was undertaken on cultural characteristics of edible mushroom i.e. *Armillariella mellea*

We are still dependent upon forests for the supply of most of the wild edible mushrooms because they have not been artificially and commercially cultivated till date. The reason being little information about their nutritional requirements and entering of some of the mushrooms into mycorrhizal association with forest trees..Hence it was considered worthwhile to investigate the nutritional requirements of these wild edible mushroom..The information is recorded on the following parameters*:* **Growth of mycelium on different solid and liquid media; Recording the effect of temperature; pH and light and darkness**

Materials and Methods

Armillariella mellea was brought into culture. For raising culture, the fruit bodies of mushroom were wiped gently with sterile cotton moistened with 70% ethanol. Bits of tissues were cut aseptically from the region of rapid cell division and planted in the centre of culture tubes containing sterilized potato-dextrose agar medium..After incubating for 10 days at 22^0 C± 2^0 C the actively growing mycelium was transferred to potato-dextrose slants for sub culturing. Throughout the study the culture was maintained on yeastal potato-dextrose agar medium at 5^0C.

i) Composition of media

In order to study the effect of different solid and liquid media on growth, ten solid media of the same composition as given by Tuite (1969) were tried. In case of solid media, inoculations were done in petriplates, whereas inoculations were done in 100 ml conical flasks in case of liquid media, 20 ml of the liquid medium in each flask. Three replicates of each medium were taken for the purpose of study.

ii) Sterilization

All glassware was sterilized in an oven at 180± 5^0C for 90 minutes. The media were autoclaved at 15 lb pressure per sq. inch (1.0545kg /cm^2) for 20 minutes. The inoculation needle and cork borer were initially dipped in ethyl alcohol and then flame sterilized.

iii) Inoculum

Inoculum used during the course of all physiological studies consisted of 5 mm diameter discs cut with the help of pre-sterilized cork borer. Ten days old cultures raised on PDA were used.

iv) Incubation period

Pertiplates containing the basal medium and inoculums were incubated for ten days at 22 ± 2^0 C in order to raise the culture for further studies.

v) Recording of growth

On solid media, the vegetative growth was recorded by taking the average linear growth of mycelia colony in two directions at right angles, till the petriplates were completely colonized. In the liquid media studies, the mycelia mats were filtered through Whatman No. I filter paper discs of 7.5 cm diameter. These filter papers were previously oven dried at 70 ± 5^oC for 3 consecutive days (until constant weight) and weighed, after keeping in moisture free desiccators. After filtration the mycelia mat was again oven dried as above and weighed to record the final dry weight of the same. Throughout the experimentation, three replicates of each treatment were kept and the average was used as a quantitative measure for comparing the growth under different treatments.

vi) Effect of temperature on growth

In this experiment the best solid and liquid medium, out of the 10 tried, were selected for the experiment. The flasks containing basal medium and inoculums were incubated at different temperatures viz. 5, 10, 15, 20, 25, 30, 35 and 40^0C, in separate incubators for studying the optimal temperature requirement.

vii) Effect of Hydrogen ion concentration (pH) on growth

In order to study the effect of pH, inoculation was done in different media with pH adjusted at 3.5, 4.0, 4.5, 5.0, 5.5, 6.0, 6.5, 7.0, 7.5, 8.0, and 8.5, respectively. The pH of basal medium was adjusted with the help of sodium citrate and sodium phosphate buffers.

viii) Effect of light and darkness on growth

The flasks with best basal liquid medium, with optimum temperature and pH, were given the light and dark treatment. For dark conditions flasks were wrapped with black paper so that no light could enter inside.

Observations

Growth of mycelium of *Armillariella mellea* on different solid media

Ten solid media (Tuite, 1969) were tried for the growth (Plate –I) of mycelium of *Armillariella mellea*. The circular growth in petriplates was recorded after ten days of incubation at temperature ($25 \pm .5^oC$).The mean colony diameter of mycelium is numerically and graphically presented in Table 1 and Fig.1, respectively.

One-way ANOVA with Tukey's multiple comparison test was applied to compare the difference of colony diameter means in different solid media. The analysis indicated that the difference of colony diameter means of mycelium between Potato Dextrose Agar and Yeastal Potato Dextrose Agar; Potato Dextrose Agar and Pridham Yeast Malt Dextrose; Yeastal Potato Dextrose Agar and Pridham Yeast Malt Dextrose; Malt Agar and Wheat Grain Extract and Wheat Grain Extract and Maize Grain Extract was non-significant (HSD: 0.000; F-value: 4446.204; $P \leq 0.1$). Whereas, rest of the solid media pairs showed very significant difference between means of mycelial colony diameter (HSD: 0.000; F-value: 4446.204; $P \leq 0.001$) (Table 1.1).

Results of ten solid media tried to grow *Armillariella mellea* revealed that Yeastal Potato Dextrose Agar allowed maximum colony diameter, whereas Czapek's Dox permitted minimum mycelial colony diameter. The mean colony diameter of *Armillariella mellea* on Yeastal Potato Dextrose Agar was significantly more than all other media studied and found to be the best medium .

Growth of mycelium of *Armillariellla mellea* in different liqiud media

To find out the best liquid medium for growing *Armillariella mellea,* five liquid media (Tuite, 1969) were tried. The weight of mycelium was measured after ten days of incubation at temperature ($25 \pm .5^oC$) .The mean mycelial weight (± standard deviation) in different liquid media is numerically and graphically presented in Table 2 and Fig. 2, respectively.

One-way ANOVA analysis with Tukey's multiple comparison test to compare the difference of mycelial weight means revealed that all the liquid media pairs revealed very significant difference between difference of mycelial weight means (HSD:0.000; F-value: 9263.358; $P \leq 0.001$) (Table 2.1).

Results of liquid media tested to grow culture of *Armillariella mellea* proved that Glucose-Asparagine showed maximum growth, whereas, Asthana and Hawker's solution allowed minimum mycelial growth. The mean mycelial growth in Glucose-Asparagine was significantly more than all other media tested and was selected as the best medium.

Effect of temperature

To record the effect of temperature on mycelial growth of *Armillariella mellea*, the mycelium was inoculated in basal liquid medium (Glucose-Asparagine). The flasks were incubated at a temperature range of 5-40^{o}C.The mean mycelial weight (± standard deviation) at different temperatures is numerically and graphically presented in Table 3 and Fig. 3, respectively.
One-way ANOVA with Tukey's multiple comparison test was applied to compare the weight of mycelium and it was observed that difference of mycelial weight means between temperature pairs 5^{o}C and 10^{o}C; 5^{0}C and 40^{0}c; and 10^{o}C and 40^{o}C was non-significant (HSD: 0.000; F-value: 14507.498; P<0.1). The difference in the remaining temperature pairs was found to be very significant (HSD: 0.000; F-value: 14507.498) (Table 3.1).

It was concluded from the results that maximum and minimum mycelial growth of *Armillariella mellea* was recorded at 25^{o}C and 15^{o}C, respectively. The growth ceased completely at 5^{o}C, 10^{o}C and 40^{o}C. The mean mycelial weight at 25^{o}C was significantly more than at all other temperature values studied and was recorded as the optimum temperature.

Effect of Hydrogen ion concentration (pH)

To study the effect of different pH values on the mycelial growth of *Arimillariella mellea*, the pH value of the basal liquid medium in flasks was adjusted in the range of 3.5-8.5 accordingly. The pure culture of *Armillariella mellea* was inoculated and incubated at the optimum temperature(25 ± .2^{o}C).The mean mycelial weight (± standard deviation) (mg) at different pH values is numerically and graphically presented in Table 4 and Fig. 4, respectively.

Application of one-way ANOVA with Tukey's multiple comparison test to compare the difference of mycelial weight means revealed that the difference between means of mycelial weight (mg) between pH values 3.5 and 8.0; 3.5 and 8.5; 4.0 and 7.0 and 8.0 and 8.5 was found non-significant (HSD: 0.000; F-value: 1203.820; $P \leq 0.1$). The pH pair 4.5 and 7.5 showed least significant difference between means of mycelial weight (HSD: 0.000; F-value: 1203.820; $P \leq 0.05$). Whereas, the remaining pH pairs showed very significant difference (HSD: 0.00; F-value: 1203.820; $P \leq 0.001$) (Table 4.1).

Results concluded that maximum growth *Armillariella mellea* was observed at pH 6.0 and minimum at pH 3.5. The mean mycelium weight (mg) at pH 6.0 was significant more than at all other pH value studied.

Hence, pH 6.0 was considered as optimum pH value for culturing *Armillariella mellea* in subsequent studies.

Effect of light and darkness

To record the effect of light and dark conditions on the mycelial growth of *Armillariella mellea*, the basal medium (Glucose-Asparagine) adjusted at pH 6.0 was inoculated and incubated at optimum temperature (25 ± .2^{o}C) in light and darkness (flasks wrapped in black paper). The mean mycelial weight (± standard deviation) (mg) in light and dark conditions is numerically and graphically presented in Table 5 and Fig. 5, respectively.
On application of Student's t-test, it was found that weight of mycelium in dark was significantly more than in light conditions (t-value: -82.025; $P \leq 0.001$) (Table 5.1).
Hence, results proved that maximum growth of *Armillariella mellea* occurred in dark conditions.

Table 1: Colony diameter of *Armillariella mellea* on different solid media

Sr. No.	Name of the medium	Colony diameter (cm) (mean ± SD)
1.	Potato Dextrose Agar (PDA)	8.60 ± .100
2.	Yeastal Potato Dextrose Agar (YPDA)	8.87 ± .058
3.	Pridham Yeast Malt Dextrose (PYMD)	8.40 ± .100
4.	Glucose Yeast Agar (GYA)	7.68 ± .275
5.	Malt Agar (MA)	6.82 ± .029
6.	Wheat Grain Extract (WGE)	6.53 ± .464

7.	Maize Grain Extract (MGE)	6.22 ± .029
8.	Horse Gram Extract (HGE)	6.07 ± .076
9.	Pea Extract (PE)	5.42 ± .029
10.	Czapek's Dox (CD)	2.83 ± .058

* Incubation period of 10 days

Table 2: Weight of mycelium (mg) of *Armillariella mellea* in different liquid media

Sr. No.	Name of the medium	Weight of mycelium (mg) (mean ± SD)
1.	Glucose-Asparagine	113.02 ± .875
2.	Czapek's solution	89.32 ± .553
3.	Dimmick's solution	80.51 ± .559
4.	Richard's solution	35.63 ± .550
5.	Asthana and Hawker's solution	28.81 ± .648

* Incubation period of 10 days

Table 1.1: Differences between the means and One-Way ANOVA with Tukey's Multiple Comparison Test for colony diameter (cm) of mycelium of *Armillariella mellea* on different solid growth media

Sr. No.	Growth Media		1 PDA	2 YPDA	3 PYMD	4 GYA	5 MA	6 WGE	7 MGE	8 HGE	9 PE	10 CD	HSD	F-value
		Mean (cm)	8.60	8.867	8.400	7.683	6.817	6.533	6.217	6.067	5.417	2.833	0.00	4446.204
1.	**PDA**	8.600	0.00	.267 NS	-.200 NS	.917***	1.783***	2.067***	2.383***	2.533***	3.183***	5.767***		
2.	**YPDA**	8.867		0.00	.467 NS	1.183***	2.050***	2.333***	2.650***	2.800***	3.450***	6.033***		
3.	**PYMD**	8.400			0.00	.717***	1.583***	1.867***	2.183***	2.333***	2.983***	5.567***		
4.	**GYA**	7.683				0.00	.867***	1.150***	1.467***	1.617***	2.267***	4.850***		
5.	**MA**	6.817					0.00	-.283 NS	.600***	.750***	1.400***	3.983***		
6.	**WGE**	6.533						0.00	.317 NS	.467 NS	1.117***	3.700***		
7.	**MGE**	6.217							0.00	.150 NS	.800***	3.383***		
8.	**HE**	6.067								0.00	.650***	3.233***		
9.	**PE**	5.417									0.00	2.583***		
10.	**CD**	2.833										0.00		

Table 2.1: Differences between the means and One-Way ANOVA with Tukey's Multiple Comparison Test for weight (mg) of mycelium of *Armillariella mellea* in different liquid media

Sr. No.	Liquid Growth Media		1 Glucose Asparagine	2 Czapek's Solution	3 Dimmick's Solution	4 Richard's Solution	5 Asthana and Hawker's Solution	HSD	F-value
		Mean (mg)	113.02	89.32	80.51	35.63	28.81	0.00	92.63.358
1	**Glucose-Asparagine**	113.02	0.00	23.69***	32.51***	77.38***	84.21***		
2	**Czapek's Solution**	89.32		0.00	8.81***	53.69***	60.51***		
3	**Dimmick's Solution**	80.51			0.00	44.88***	57.70***		
4	**Richard's Solution**	35.63				0.00	6.82***		
5	**Asthana and Hawker's Solution**	28.81					0.00		

***P ≤ 0.001;** P ≤ 0.01; * P ≤ 0.05; NS: Non-Significant Difference at P ≤ 0.10;
HSD: Honestly Significant Difference as revealed through Tukey's multiple comparison test.

Table 3: Weight of mycelium (mg) of *Armillariella mellea* at different temperatures

Sr. No.	Temperature(in ^{0}C)	Weight of mycelium (mg) (mean ± SD)
1.	5	0.00 ± .000
2.	10	0.00 ± .000
3.	15	24.26 ± .554
4.	20	54.06 ± .997

5.	25	112.40 ± .470
6.	30	70.85 ± 1.031
7.	35	26.30 ± .360
8.	40	0.00 ± .000

* Incubation period of 10 days

Table 4: Weight of mycelium (mg) of *Armillariella mellea* at different pH values

Sr. No.	pH	Weight of mycelium (mg) (mean ± SD)
1.	3.5	21.32 ± .291
2.	4.0	34.43 ± .55
3.	4.5	39.45 ± .440
4.	5.0	51.03 ± .152
5.	5.5	83.89 ± 4.557
6.	6.0	107.04 ± .206
7.	6.5	75.08 ± .200
8.	7.0	43.33 ± .416
9.	7.5	35.77 ± .724
10.	8.0	22.25 ± .136
11.	8.5	22.01 ± .160

* Incubation period of 10 days

Table 3.1 :Differences between the means and One-Way ANOVA with Tukey's Multiple Comparison Test for weight (mg) of mycelium of *Armillariella mellea* at different temperatures

Sr. No.	Temperature		1 5	2 10	3 15	4 20	5 25	6 30	7 35	8 40	HSD	F-value
		Mean (mg)	0.00	0.00	24.26	54.06	112.40	70.85	26.30	0.00	.000	14507.498
1	**5**	.00	0.00	0.00 ^{NS}	-24.26***	-54.06***	-112.40***	-70.85***	-26.30***	0.00 ^{NS}		
2	**10**	0.00		0.00	-24.26***	-54.06***	-112.40***	-70.85***	-26.30***	0.00 ^{NS}		
3	**15**	24.26			0.00	-29.79***	-88.14***	-46.59***	-2.04***	24.26***		
4	**20**	54.06				0.00	-58.34***	-16.79***	27.76***	54.06***		
5	**25**	112.40					0.00	41.55***	86.10***	112.40***		
6	**30**	70.85						0.00	44.55***	70.85***		
7	**35**	26.30							0.00	26.30***		
8	**40**	0.00								0.00		

Table 4.1: Differences between the means and One-Way ANOVA with Tukey's Multiple Comparison Test for the weight of mycelium of *Armillariella mellea* at different pH values

Sr. No.	pH		1 3.5	2 4.0	3 4.5	4 5.0	5 5.5	6 6.0	7 6.5	8 7.0	9 7.5	10 8.0	11 8.5	HSD	F-value
		Mean (mg)	21.32	34.43	39.45	51.03	83.89	107.04	75.08	43.33	35.77	22.25	22.01	.000	1203.820
1.	**3.5**	21.32	0.00	-13.11***	-18.13***	-29.71***	-62.57***	-85.72***	-53.76***	-22.01***	-14.45***	.93 ^{NS}	1.30 ^{NS}		
2.	**4.0**	34.43		0.00	-5.02***	-16.60***	-49.46***	-72.61***	-40.65***	8.90***	-1.34^{NS}	12.18***	14.42***		
3.	**4.5**	39.45			0.00	-11.59***	-44.44***	-67.59***	-35.64***	-3.89*	3..68^{NS}	17.19***	19.43***		
4.	**5.0**	51.03				0.00	-32.85***	-56.01***	-24.05***	7.70***	15.27***	28.78***	31.02***		
5.	**5.5**	83.89					0.00	-23.15***	8.80***	40.55***	48.12***	61.63***	63.87***		
6.	**6.0**	107.04						0.00	31.95***	63.71***	71.27***	84.79***	87.03***		
7.	**6.5**	75.08							0.00	31.95***	39.32***	52.83***	55.07***		
8.	**7.0**	43.33								0.00	7.57***	21.08***	23.32***		
9.	**7.5**	35.77									0.00	13.51***	15.75***		

10.	**8.0**	22.25										0.00	2.24^{NS}	
11.	**8.5**	22.01											0.00	

***P ≤ 0.001; ** P ≤ 0.05; * P ≤ 0.10; NS: Non- Significant Difference at P ≤ 0.10;
HSD: Honestly Significant Difference as revealed through Tukey's multiple comparison test.

Table 5: Weight of mycelium (mg) of *Armillariella mellea* in best liquid medium (Glucose- asparagine) in light and darkness

Sr. No.	Treatments	Weight of mycelium (mg) (mean ± S.D)
1.	Light	78.107 ± .093
2.	Dark	88.050 ± .095

*** Incubation period of 10 days**

Table 5.1: The significance of differences between the Means as determined by Student's t-test for mycelium weight of for mycelium weight of *Armillariella mellea* in light and dark conditions

Sr. No.	Treatments	Weight of mycelium (mg) (mean ± SD)	t-value
1.	Light	78.107 ± .093	-129.329
2.	Dark	88.050 ± .095	

*** Incubation period of 10 days**
***** P ≤ 0.001;** P ≤ 0.01; * P ≤ 0.05; NS: Non-Significant Difference at P ≤ 0.10**

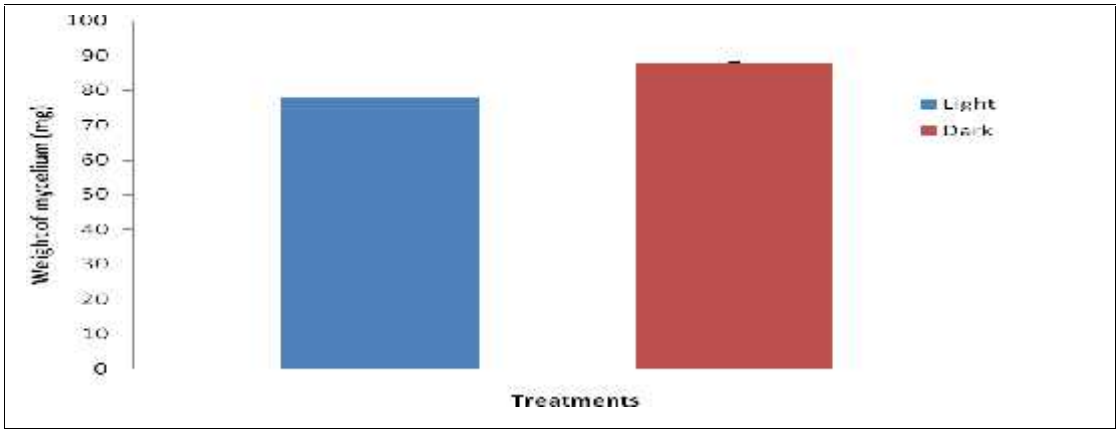

Fig. 5: Weight of mycelium (mg) of *Armillariella mellea* under light and darkness

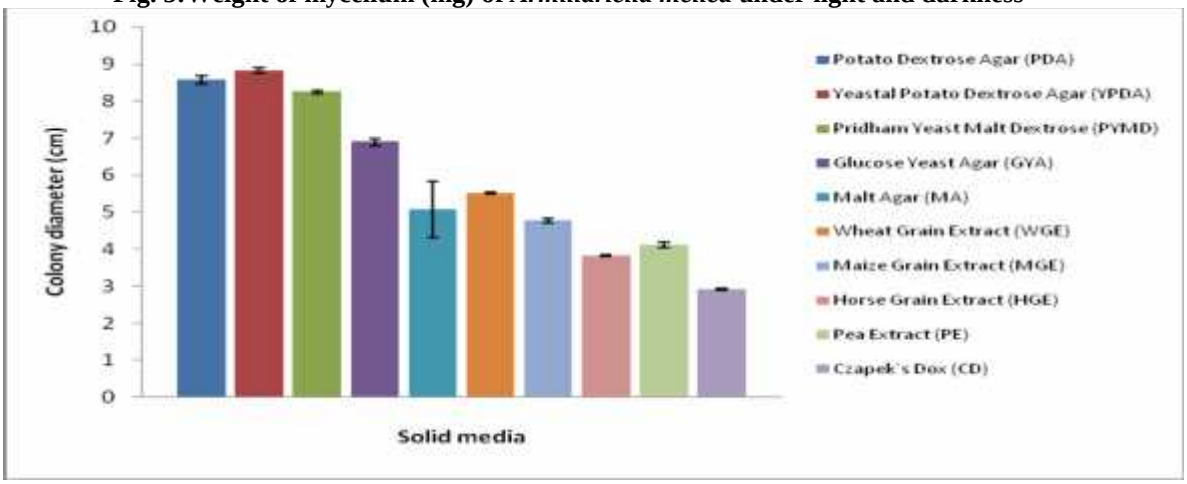

Fig. 1: Colony diameter (cm) of *Armillariella mellea on* different solid media

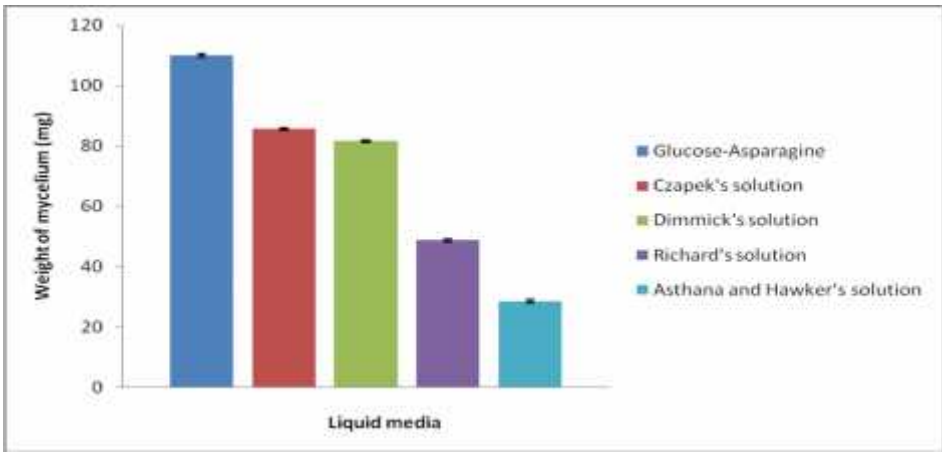

Fig. 2:Weight of mycelium (mg) of *Armillariella mellea* in different liquid media

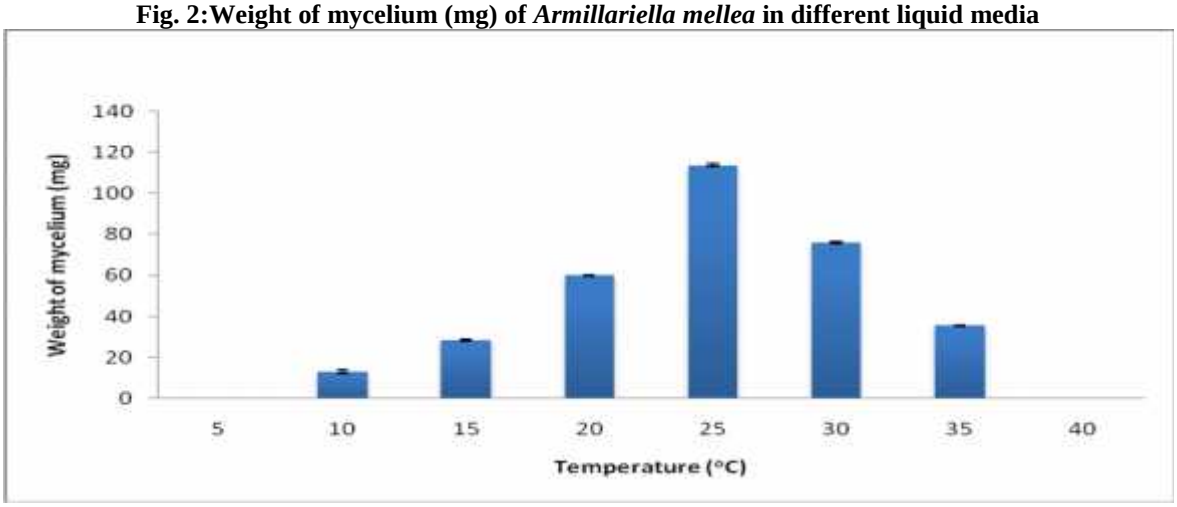

Fig. 3:Weight of mycelium (mg) of *Armillariella mellea* at different temperatures

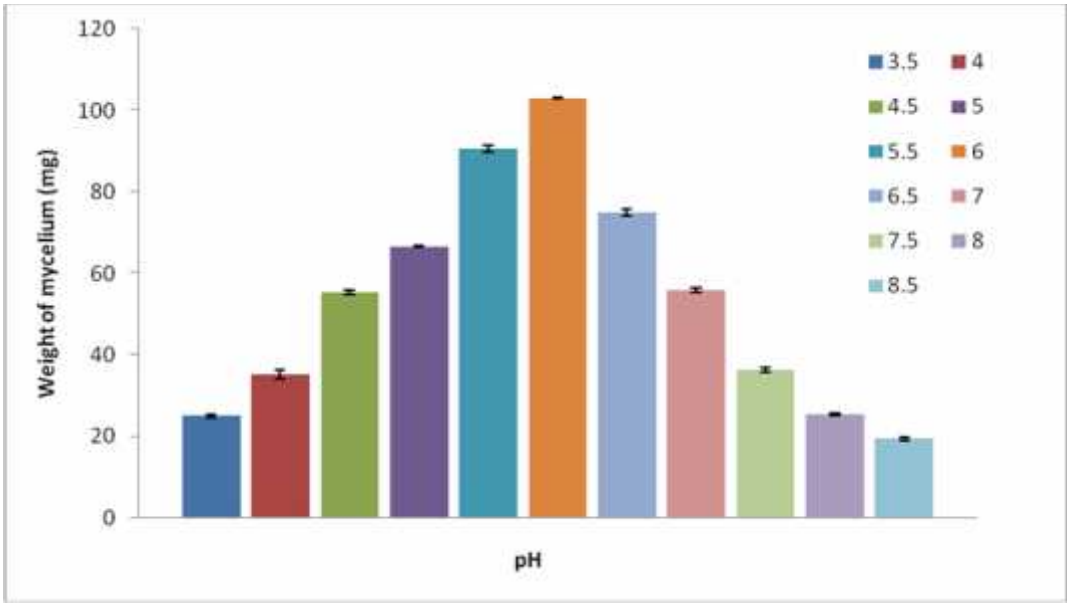

Fig. 4: Weight of mycelium (mg) of *Armillariella mellea* at different pH values

Results

Results derived from the ten solid media tried for the growth of *Armillariella mellea*, clearly indicated that Yeastal Potato Dextrose Agar medium supported maximum growth of mycelium while, Czapek's Dox permitted minimum colony diameter(Table 1). The mean colony diameter of Yeastal Potato Dextrose Agar was significantly more than all other tested solid media (Table 1.1).

Results of five liquid media tried for the growth of *Armillariella mellea* proved that Glucose-Asparagine showed maximum mycelial weight whereas minimum growth was recorded in Asthana and Hawker's solution (Table 2). Whereas, the comparison of mycelial weight means observed in all the five liquid media pairs was very significant (HSD: 0.00; F-value: 9283.584; $P \leq 0.001$) (Table 2.1).

Maximum and minimum growth of *Armillariella mellea* occurred at 25°C and 10°C, respectively (Table 3). The growth ceased completely at 5°C and 40°C. The mean mycelial growth was significantly more than at all other temperature values studied (Table 3.1).

Maximum growth **of** *Armillariella mellea* was recorded at pH 6.0 and (Table 4). The mean mycelial weight (mg) at pH 6.0 was significantly more than all other pH values studied (Table 4.1).

Regarding growth of mycelium of *Armillariella mellea* was better in dark than under light conditions (Table 5). Student's t-test, revealed that weight of mycelium in dark was statistically very significant under dark conditions than under light conditions (Table 5.1).

Discussion

A study of detailed growth conditions of an organism is as important as the study of any of its other aspects. In the present study growth conditions regarding (media, temperature, hydrogen ion concentration light and darkness) of *Armillariella mellea* were investigated with the cultures raised from their basidiocarps.

The literature has references showing evidence of best growth of *Armillariella mellea* mycelium on Yeastal Potato Dextrose Agar (YPDA) and Potato Dextrose Agar (PDA). Good mycelial growth on YPDA has been recorded by Jandaik and Kapoor (1975a) in case of *Pleurotus sajor- caju, Podaxis pistillaris and Phellorina inquinans.* Rangad and Jandiak (1977) also reported YPDA as best medium for growth of different species of *Pleurotus, Agrocybe aegerita Flammulina valutipes and Stropharia rugoso- annulata.* Thianga and Jandaik (1979) also recorded best growth of *M. procera* on YPDA. Chaturvedi (1987) recorded YPDA as best medium for the growth of *P. ostreatus.* Shad (1989) recorded best growth of *M. esculenta, M. conica and M. deliciosa* on PDA. Nair and Devi *et ai.,* (1987) also recorded the YPDA as the best medium for culturing *Coprinus lagopus.*

For determining the comparative suitability for vegetative growth of *Armillariella mellea,* five liquid media were tested. Glucose-Asparagine supported maximum average mycelial growth. Rangad and Jandaik(1982) also recorded maximum growth of *F.velutipes,Agrocybe aegerita* and *Stropharia-rugoso-annulata* in Glucose- Asparagine, Mehta (1985) and Chaturvedi (1987) observed Glucose- Asparagine medium to favour maximum vegetative growth of *Pleurotus sapidius and Pleurotus ostreatus.* Singh and Lakhanpal (1988) also recorded maximum growth of *T. himalayansis* in Glucose Asparagine solution. Shad (1989) also found glucose asparagine to support maximum growth of *M. esculanta, M. deliciosa, M. Conica, M. crassipes and M. semilibra.*With regard to the effect of temperatures, it was recorded that all the sixteen mushrooms studied could grow in a wide temperature range of 10- 35°C but failed to grow below 10°C and above 35°C. Rangad and Jandaik (1977) have reported maximum growth of *Agrocybe aegerita and Stropharia rugoso- annulata* at 25°C. Mehta and Bhandal (1988) also recorded growth of *P. ostreatus, P. florida, P. saroj- caju, P. flabellatus, P. sapidus and P. cystidiosus at* 25^{O}C. While, Gupta (1990) recorded 25°C to be the optimum temperature for vegetative growth of *M. esculenta, M. conica, M. crassipes, and M. angusticeps*. The highest radial diameter, mycelia density and dry mycelia weight were recorded at temperature 25°C for *Pleurotus ostreatus (Ali et al., 2004).*Effects of temperature (5-34^{o}C)were investigated on hyphal growth of *Pleurotus flabellatus* . The temperature for hyphal growth of *Pleurotus flabellatus* varied from 20^{o}C to 31^{o}C with optimum temperature at 25°C (Li Rong et al., 2004). Song *et. al.,* (2004) conducted studies on growth conditions of liquid culture for *Morchella conica.* The optimum temperature for *Pleurotus nebrodensis* was 25^{o}C. The studies indicated that the suitable temperature for mycelial growth was 22-28°C although 25°C was optimum (HongTao et. al., 2005). Similarly, Yadav and Yadav (2012) observed 25°C to be the optimum temperature for the growth of *Cantharellus cibarius and Scleroderma bovista.*

It is evident from the results that showed maximum growth at 25°C. The growth of mycelium starts decreasing with increase or decrease in optimum temperature. The results are in agreement with the references quoted in the literature.

For recording Optimum pH level for their growth of *Armillariella mellea* the mycelium was grown in the best suited liquid medium at different levels of pH. It was recorded that maximum growth occurred at slightly acidic pH i.e. 6.0. This was closely followed by 5.5 and 6.5 in acidic pH range. This finding is in agreement with the optimum pH for *Podaxis pistillaris* which had been recorded to be 6.0 by Jandaik and Kapoor (1975b). Thind and Jandaik (1979) also recorded pH 6.0 as best pH for growth of *Macrolepiota procera.* Rangad and Jandaik (1982) also recorded maximum growth of mycelium at pH 6.0 in *Stropharia- rugoso- annulata.* Nair and Devi (1986-87) also recorded pH 6.0, as optimum pH for the growth of *Calocybe lagopus.* Further, Ali *et al.,* (2004) reported pH 6.0 for the maximum mycelium growth of *pleurotus ostreatus.* During the screening of culture conditions for *P. pulmonarius and P. columbinus,* the best pH was reported 6.0 (QinnGhe *et al.,* 2004). Studies of Song *et al.* (2004) on growth conditions of *Ramaria botrytis* revealed pH 6.0 as suitable for mycelial growth.

It is evident from the results that there is decrease in mycelial growth of *Armillariella mellea*, on either side of optimum pH..In other words, the growth of mycelium increased with decrease in acidity and decreased with increase in basicity upto optimum pH.

Mycelium of *Armillariella mellea* was found to grow better under dark conditions in comparison to light conditions. Better growth of *S. crispa* and *T. himalayansis* was also recorded in dark conditions by Sharma (1987) and Lakhanpal *et al.,* (1988).

Conclusion

All the solid media tested, supported good to moderate growth of *Armillariella mellea.* However the highest growth rate of these fungi was recorded in Yeastal Potato Dextrose Agar medium; Czapek's Dox medium supported least growth of *Armillariella mellea* . Good growth on YPDA may be ascribed to yeast extract which is known to contain growth enhancing substances like riboflavin. Least growth of this mushroom in remaining extracts may be attributed to the lack of nutrient content required for the growth of fungus used in present investigations.(Table 1 and Fig.1). Out of five liquid media tried Glucose-Asparagine showed maximum mycelial weight. (Table 2 and Fig.2). The better growth of fungi in Glucose asparagines may be ascribed to free amino acid asparagine present in the solution.

Maximum and minimum growth of *Armillariella mellea* occurred at 25°C and 10°C, respectively. The growth ceased completely at 5°C and 40°C.(Table 3 and Fig.3). The mean mycelial growth was significantly more than at all other temperature values studied. Maximum growth of *Armillariella mellea* was recorded at pH 6.0.(Table 4 and Fig.4). Growth of mycelium of *Armillariella mellea* was better in dark than under light conditions. (Table 5 and Fig.5).

The study on *Armillariella mellea* concludes that it also behaves in the same manner in culture as the other commercially cultivated mushrooms like *Agaricus bisporus, Pleurotus* and *Volvariella* spp.etc. There is need to develop and standardize the cultivation technology of these wild edible mushrooms for making them commercially cultivable and popular among the common people like other cultivated mushrooms.

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