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Comparative Study on Growth Parameters and Yield Potential of Five Strains of Milky Mushroom (*Calocybe indica*)

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Milky mushroom is the third most largely growing mushrooms and has the good demand in the market and world trade too. An experimental was carried out with five strains of *Calocybe indica* viz. CI-4, CI-13, CI-14, CI-15 and CI-18 for growth behaviour and yield potential. Minimum spawn run period was observed from CI-14 and CI-4 where completed in 15.66 days while CI-15 took maximum time to complete spawn run (18 days). Pinhead was appeared early (28 days) in CI-14 while strain CI-15 took maximum time to initiate pin head (34 days). The harvesting of 1st, 2nd and 3rd flushes were completed early (35, 47 and 58.33 days respectively) from strain CI-14 followed CI-4, CI-18, CI-13 and CI-15. Mushroom strain CI-14 was better in performance for total number of fruit bodies (25), total length of mushroom (10.41 cm) and cap diameter (8.50 cm) but in strain CI-13 length of stalk was better (7.81 cm). The strain CI-18 showed better performance for stalk diameter (3.17 cm) followed by CI-13 (3.03cm), CI-14 (2.97 cm), CI-4 (2.50 cm) and CI-15 (4.71 cm). Finally strain CI-14 was in better performance while CI-13 showed lowest yield. Highest average yield of 1st 2nd and 3rd flush was obtained from CI-14 (441.67g, 285g and 85g). The total yield was also better for CI-14 (811.67g). This experiment will help to mushroom growers for selection of mushroom strains for obtaining better growth behaviour and yield potential of milky mushroom (*Calocybe indica*).

Keywords: *Calocybe indica*; strains, spawn, growth behaviour and yield potential.

Mushroom cultivation is the most suitable technology for creating wealth and health out of wastes from plants, animals and industries which are abundantly available on earth. In Eastern U.P., three types of mushrooms *viz*. white button mushroom (*Agaricus bisporus*), oyster mushroom (*Pleurotus* spp.) and milky mushroom (*Calocybe indica*) are commonly grown for commercial purpose.The milky mushroom (*Calocybe indica*) is relatively new to the world of mushroom industry and third most important in production and that can be cultivated on lignin rich agricultural wastes. This mushroom was first reported from West Bengal, India (Purakayasha and Chandra 1974). Even though attempts were made to grow *C. indica* (Purkayastha and Nayak, 1981); Chakravarty *et al.*, 1981, Doshi *et al*., 1989. Pandey and Tewari, 1993) only limited success was achieved in increasing the bio-efficiency and productivity of this mushroom. The advantages of this mushroom over other mushrooms are easy method of cultivation, less investment, very attractive fruiting body, pleasing milk white color, long shelf life, more nutritious and less time to grow.

The choice of the farmers for growing of any crops variety depends upon its yielding ability. It means the cost-benefit (C: B) ratio should always be in favour of the farmer. Scientists are suggesting to farmers for growing the high yielding mushroom strains. Therefore present investigation was based on the comparison of five strain of milky mushroom *(Calocybe indica)* for growth behaviour and yield potential.

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MATERIALS AND METHODS

Collection of Mushroom Culture

Pure culture of four strains CI-4, CI-13, CI-15 and CI-18 of *Calocybe indica* were introduced from D.M.R. Solan (Himachal Pradesh) and one strain CI-14 of Milky mushroom, *Calocybe indica* was already maintained in "Mushroom Spawn Laboratory" Department of Mycology and Plant Pathology, IAS, BHU Varanasi. These cultures were sub-cultured and maintained on PDA medium in a BOD incubator at 30 ±2 º C temperatures for further investigation.

Mushroom Spawn

Wheat grains were used as medium for preparation of mushroom spawn. Well cleaned and healthy wheat grains were washed and soaked for 6 to 8 hours in water and then boiled for 40 minutes or until they become soft. Excess water was drained off after boiling and the grains were cooled or air dried in wooden/ plastic tray up to 60% moisture. These cooled grains were supplemented with 2% calcium carbonate and 2% calcium sulphate on dry weight basis of grains to avoid clumping of grains. These supplemented boiled wheat grains were filled (300g/ bottle) in clean 500ml saline bottle and plugged these bottles with non-absorbent cotton plugs. These wheat grain filled bottles were sterilized in autoclave at 15 lb pressure (121ÚC) for one hour and then allowed to cool at room temperature for a day.

After sterilization and cooling these grain filled bottles were aseptically inoculated separately with 7-10 days old mushroom culture of five strains of *Calocybe indica.* These inoculated bottles were incubated at 28 ± 2 ÚC in B.O.D. incubator for mycelial growth. These inoculated bottles were shaken at 4 days interval to allow proper spread of the mycelium between the grains.

Preparation of substrate

Wheat straw was used as substrate for production of milky mushroom and this was soaked in water for 15 hours in large plastic buckets. After soaking, excess water drained out and substrate was sterilized in autoclave (at 121°C, 15 psi for 1 hour). For sterilizing this moist substrate filled in gunny bags and put in autoclave. After autoclaving sterilized substrate cooled and spread on cleaned floor. This ready substrate was used for spawning.

Spawning

It is the process of uniformly mixing of spawn in the well prepared substrates. Wheat straw was selected as substrate for milky mushroom cultivation. Spawn of all five strains of *Calocybe indica* were mixed separately @ 50g/kg dry substrate and filled in separate polythene bags. Each bag was contained 2.5 kg moist substrate. The upper surface of these bags were covered with cleaned news paper and put on wooden ranks in mushroom growing room where temperature ranged between 28 to 34 ºC.

Casing

The process of applying casing layer on the surface of the mushroom bed is called casing. Casing also provides moisture and strength for growth and development of fruit bodies and maintains humidity and temperature in the growing chamber. After completion of spawn run, the news paper sheet was removed and surface of mushroom bed was covered (3-4 cm thick layer) by casing soil. The casing soil was prepared from 2 years old farm yard manure and loam soil (garden soil) (1:1 ratio). The casing materials were wetted with water and sterilized by autoclaving at 121°C and 15 psi for 1 hr. before casing. Mushroom beds were sprayed regularly with water to keep the casing layer adequately moist**.**

Harvesting

Pinheads were generally appears within 28 days after casing and they become ready for harvesting within another one week. The mature unopened mushroom fruiting body was harvested when they achieved proper size and shape at most once a day. They were picked by gently twisting of the base of stalk without disturbing casing. Small pits were formed after harvesting and these pits were immediately re-cased with casing soil for development of next fruit bodies. The moisture in the casing soil was maintained by regular spraying of water. Three to four successive harvesting were done from each bag.

Observation and Measurement

The following growth parameter were observed during this investigation-

Growth behaviour (recorded in days)

1. Spawn run period
2. Initiation of pinhead
3. First harvesting

4. Second harvesting
5. Third harvesting

Yield potential

1. Yield of first flush (g)
2. Yield of second flush (g)
3. Yield of third flush (g)
4. Total yield (g)
5. Biological efficiency (%)

Biological efficiency (B.E.) was determined by following given formula-

$$B.E. = \frac{\text{Fresh weight of mushroom}}{\text{Dry weight of substrate}} \times 100$$

Morphological parameters of fruit bodies

1. Total no. of fruiting bodies
2. Maximum weight of fruit body (g)
3. Minimum weight of fruit body (g)
4. Average length of stalk (cm)
5. Average width of stalk (cm)
6. Average diameter of mushroom cap (cm)
7. Total length of mushroom (cm)

RESULTS

Comparative study on growth behaviour of five strains of milky mushroom (*Calocybe indica*)

Spawn run period

The spawn run period of five strains viz. CI-4, CI-13, CI-14, CI-15 and CI-18 of milky mushroom (*Calocybe indica*) were presented in Table 1. Results showed that two strains CI- 4 and CI- 14 of milky mushroom were found very fast, where it took in 15.66 days. The mushrooms bags of other two strains CI-18 and CI- 15 were completely colonized in 17 days and 18 day respectively.

Initiation of pin head

The mushroom pin head was first initiated in 28 days from strain CI-14 followed by strain CI-4 (28.67 days). The pin head of CI-18 strain was initiated in 30.33 day while CI-13 strain was initiated pin head in 31.33 days. Strain CI-15 was taken maximum time (34 days) for pin head initiation.

Harvesting of flushes and total crop period

Milky mushroom were harvested in three flushes and total crop period. There was found variation in period of harvest 1st, 2nd and 3rd flush in five strains presented in Table. Strain CI-14 was showed better performance and taken minimum time for harvest 1st, 2nd and 3rd flush (35, 47 and 58.33 respectively), followed by CI-4 where harvested in 35 day, 47.67 day and 59.67 day for 1st, 2nd and 3rd flush respectively. Maximum cropping period was recorded from strain CI-15 where it took in 70.67 days.

Comparative study on growth parameters and yield potential of five strains of milky mushroom (*C. indica*)

Total number of fruit bodies

The growth parameters of fruiting bodies like average number of fruit bodies, length of stalk, diameter of stalk, length of fruit bodies' minimum & maximum weight of fruit bodies and diameter of cap were measured and results showed in Table 2. The maximum numbers of fruit bodies were obtained from the strain CI-14 (25) followed by CI-18 (22.67), CI-4 (21.33), CI-15(19.33) and CI-13(17.66) respectively.

Table 1. Growth Period of five strains of Milky mushroom (*Calocybe indica*)-(in days)

Strains	Growth period (in days)					Total
	Spawn run period	Pin head initiation	First Flush	Second Flush	Third Flush	Crop Period (in days)
CI-4	15.66	28.67	35	47.67	59.67	59.67
CI-13	16.33	31.33	39.67	52.67	66	66
CI-14	15.66	28	35	47	58.33	58.33
CI-15	18	34	42.33	55.33	70.67	70.67
CI-18	17	30.33	38	49.67	60.67	60.67
SEM	2	2.94	4.88	6.34	7.40	7.40
CD (0.05)	4.46	5.40	6.96	7.94	8.58	8.58
CV	6.63	5.29	7.02	6.88	6.43	6.43

Maximum and minimum weight of fruit bodies

Strain CI-4 was produced biggest fruit bodies where it weighted as 112.67g followed by strain CI-14 where it weighted 99.67g. Smallest fruit body was produced by strain CI-13 and it weighted as 82.67g. Another observation like highest minimum fruit body was measured from CI-15 strain (26.33g) followed by CI-18 strain (24.33g).

Stalk length

The average stalk length was observed from five strains of milky mushroom (*Calocybe indica*).Maximum length of stalk was observed in strain CI-13 (7.81 cm) followed by CI-14 (7.43 cm), CI-18 (6.13 cm), CI-4 (5.22 cm) and CI-15 (4.74 cm).

Diameter of stalk

This result showed that the strain CI-18 was reported as maximum average diameter of stalk (3.17 cm) followed by other four strains viz. CI-13 (3.03 cm), CI-14 (2.97 cm), CI-4 (2.50 cm) and CI-15 (2.39 cm).

Total length of fruit bodies

It was showed the result in Table 2 that maximum length of fruit bodies was observed in strain CI-14 (10.41cm) followed by four other strains as CI-13 (10.32 cm), CI-18 (8.69 cm), CI-4 (7.36 cm) and CI-15 (6.90 cm).

Cap diameter

Maximum cap diameter was recorded from strain CI-14 (8.50 cm) followed by CI-4 (8.28 cm), CI-13 (7.78 cm), CI-18 (6.73 cm) and CI-15 (6.72 cm).

Total yield

Three flushes were harvested during experiment, and observed yield of each flush. A continuous reduction in yield was recorded during harvest of next flush. Data related to harvest of 1st, 2nd and 3rd flushes and comparative total yield of five strains of milky mushroom are presented in Table 3. The results from Table 3 shown that strain CI-14 was better in performance, yielded 811.3g/kg of dry wheat straw substrate. Strain CI-18 was found second in given production with 760.66g followed by CI-4 (716.67g) and CI-15 (681.33g). Minimum yield (608 g) was obtained from strain CI-13. Finally highest Biological efficiency (81.1%) was obtained from strain CI-14 followed by CI-18 (B.E.-76%).

Table 2. Growth parameters and fruit bodies of five strains of Milky mushroom (*Calocybe indica*)

Strains	No. of Fruit Bodies	Max. Wt. of Fruit Body (gm)	Min. Wt. of Fruit Body (gm)	Av. Length of Stalk (cm)	Av. Width. of Stalk (cm)	Av. Dia. of Mushroom Cap (cm)	Total Length of Fruit Body (cm)
CI-4	21.33	112.67	24	5.22	2.50	8.28	7.36
CI-13	17.66	82.67	20	7.81	3.03	7.78	10.32
CI-14	25	99.67	22.67	7.43	2.97	8.50	10.41
CI-15	19.33	85	26.33	4.74	2.39	6.72	6.90
CI-18	22.67	90	24.33	6.13	3.17	6.73	8.69
SEM	3.86	14.80	5.23	0.84	0.62	0.75	0.82
CD (0.05)	6.19	12.13	7.20	2.88	2.48	2.72	2.86
CV	9.97	8.63	12.20	7.30	12.03	5.37	5.16

Table 3. Yield potential of five strains of milky mushroom (*Calocybe indica)*

Strains	Yield of each flush and total yield				Biological efficiency (%)
	First flush (gm)	Second flush (gm)	Third flush (gm)	Total yield (gm)	
CI-4	380	240	63.33	716.67	71.67
CI-13	323.33	206.67	75.33	608	60.80
CI-14	441.67	285	85	811.67	81.16
CI-15	379.33	231.67	70.33	681.33	68.13
CI-18	438.33	256	66.33	760.66	76.06
SEM	39.43	40.57	28.91	28.62	2.86
CD (@ 5%)	19.79	20.07	16.94	16.86	5.33
CV	5.50	9.11	24.10	2.19	2.19

This result is confirmative with the findings of Sharma and Kumar (2008) who evaluated yield potential on different strains of *Calocybe indica* viz. APK-2, CI-1, CI-3, CI-6 and CI-7. The strain CI-6 resulted in maximum biological efficiency which was significantly different from other strains. The highest average weight of fruit bodies was in APK-2, which was significantly higher than CI-6, CI-7 and CI-1.

DISCUSSION

Milky mushroom is the third most largely growing mushrooms and has the good demand in the market and world trade too. There are various strains are available for production of milky mushroom. In the present investigation, five strains of *C. indica* were evaluated for proper selection of strains. Finally strain CI-14 of milky mushroom has been performed better for yield potential while CI-13 showed lowest yield and it also shows that growth parameter for strain CI-14 were better than other strains of milky mushroom. This investigation will help to mushroom growers for selection of mushroom strains for obtaining better growth behaviour and yield potential of milky mushroom (*Calocybe indica*).

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