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Effect of casing soil thickness on growth and yield of milky mushroom (*Calocybe indica*)

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Abstract

An experiment was carried out during the summer season, 2012 to study the thickness of casing soil layer affected not only initiation of fruiting bodies, number of pinhead and their development but also duration of mushroom, size and weight of sporophore and yield of fruiting body and biological efficiency. Among different thickness of casing soil viz., 1 cm, 1.5 cm, 2.0 cm, 2.5 cm, 3.0 cm, 3.5 cm, 4.0 cm and 4.5 cm, casing soil having 2.0 cm thickness observed maximum number of pinhead, sporophore, size and weight of sporophore, yield and biological efficiency. 2 cm thickness of casing layer was found to be the best as compared to other casing soil thickness. Thickness of casing layer beyond 2.0 cm gradually decreased the mushroom yield and delayed the appearance of sporophores. Biological efficiency in different treatments varied from 51.45% to 100.76%.

Keywords: Casing soil thicken, growth, yield, milky mushroom (*Calocybe indica*)

Introduction

The milky mushroom (*Calocybe indica*) is also called White summer mushroom and Dudh Chhatta. Milky mushroom has become the third commercially grown in India after white button (*Agaricus bisporus*) and oyster (*Pleurotus* spp.) mushrooms. This is a tropical mushroom which is a seven months crop from March – September. This mushroom was first collected in wild form from West Bengal (India) by Purkayastha and Chandra in 1974. Production technology of *Calocybe indica* has been introduced by Purkayastha and Nayak in 1979 which was improved by Purkayastha and Nayak in 1981. This mushroom is gaining popularity due to its attractive robust, white sporocarps long shelf life and taste (Chadha and Sharma, 1995) [1]. Mature sporocarp of *Calocybe indica* contains 4% soluble sugars, 2.9% starch and 7.4% ash (Doshi *et al.*, 1988) [3]. Mushroom culture is currently the only economic way of upgrading lignocellulosic wastes. These organisms can easily use these wastes for their growth and production of protein rich food. It is a labour intensive indoor activity which can help the landless, small and marginal farmers to raise their income, diversify economic activity and can create gainful employment, especially for the unemployed/underemployed youth, weaker section of the society and the women folk. Presently button and oyster mushrooms are commercially cultivated in tropical and subtropical regions of India. The oyster mushrooms can be easily grown under natural condition whereas button mushrooms require controlled conditions. Huge inputs are required to provide ideal condition for button mushrooms. Therefore, button mushroom cultivation is beyond the reach of ordinary farmers. The milky mushroom requires 25-35 °C temperature and can be exploited during lean period. This mushroom has an immense potential in production in the plain region of India due to its better temperature tolerance (30-35° C). Due to variation in temperature, button mushroom cannot be cultivated throughout the year.

Materials and Methods

The experiment was conducted during the summer season, 2012 and the temperature and relative humidity of the cropping room ranged from 30-35 °C and 80-85 percent respectively. 18 to 20 days old spawn raised on wheat grain through standard technique was used during experimentation. There were eight treatments with three replications. The experiment was carried out in completely randomized design (CRD) under room condition. The various casing thickness used in the experiment were as follows:

T₁ - 1 cm, T₂ - 1.5 cm, T₃ - 2.0 cm, T₄ - 2.5 cm, T₅ - 3.0 cm, T₆ - 3.5 cm, T₇ - 4.0 cm, T₈ - 4.5 cm

Preparation of substrate/spawn

The grains were initially pre-weeded by soaking in fresh water for 12 hours and washed 2-3 times under running water.

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The grains were boiled separately for 15 minutes to the extent that no starch was released. Excess water was drained off and the boiled grains were spread over a clean working table to dry slightly. Then calcium carbonate (lime) @ 6 gm/kg and calcium sulphate (gypsum) @ 12 gm/kg of grains were mixed with grains. The gypsum and lime prevents the sticking of grains together and adjusts the pH (6.5 to 6.7). Each of the ready mixture was then filled in 27 × 20 cm size polypropylene bags @ 300 g approximately in each and plugged with non-absorbent cotton. All the filled and plugged bags were sterilized in a autoclave at 20 pound pressure for two hours. Next day in the morning sterilized bags were taken out and allowed to cool down for 20-30 minutes. These cooled bags were transferred in inoculation chamber. Inner surface of laminar air flow, inoculating needle and spirit lamp were sterilized with spirit then bags were kept in laminar air flow and switched on UV light for 30 minutes for sterilization.

Inoculation and incubation

Bags were inoculated with equal amount of inoculums (3 mm diameter bits) of fresh fruiting culture of (*Calocybe indica*) under aseptic condition on the same day considering the date of inoculation as zero day and incubated at room temperature (26 ± 4 °C) for spawn growth. The inoculated bags were kept on B.O.D. incubator for observation and the days required for completion and texture of mycelium in bags were recorded. Further, these fresh spawns bags were used for cultivation on paddy straw using polypropylene bags.

Cultivation of *Calocybe indica*

Preparation of straw substrate

Well dried paddy straw was chopped into 3-4 cm bits and then 10-12 kg straw was immersed in 100 litres water containing 100 ml formaldehyde and 10 gm bavistin to sterilize for 16-18 hours. Sterilization was done in 4 × 3 × 2.5 ft. size hauze. Then the mouth of the hauze is covered with polythene sheet to avoid release of the gases coming out from formaldehyde. Next day in the morning sterilized straw was taken out and drained off excess water by keeping on wire mesh frame and spread over on polythene sheet to dry for 1 to 1.5 hours depending on the prevailing weather condition. The moisture content of the straw is kept at 65-70 percent. It was tested by palm method by squeezing the handful of straw. Then the straw was ready for spawning.

Spawning and Spawn running

Polypropylene bags of 60 cm × 40 cm of 100 gauge were used. The bottom of the bag was tied with a rubber band to make a cylindrical shape to the bed. Then the bag is sterilized with spirit dipped cotton by swapping and then the bag was turned over so that the tied portion comes inside. Bottom of the bag was slightly widen. The bag was filled with alternate layers of straw (1.5 kg sterilized dry straw per bag) and spawn (300 g/kg of dry straw). Press it with palm to let the air go out. The bag was then tied with a rubber band along with a label of the species and date of spawning. About 10-15 holes were made into the polythene bags for the exchange of air and gases. Spawned bag was stacked in racks which were arranged in spawn running room. During spawn running period, temperature of 26 ± 4 °C was maintained. These partially controlled conditions were maintained for 20 to 25 days for complete spawn running period when whitish cottony mycelia growth completely covered the straw in polythene bags. The polythene bags were cut into two halves with a

hacksaw blade. After cutting of bags, casing soil were applied to a height of 2 cm above the newly exposed surface of the bags.

Preparation of casing mixture and casing

Ten days after spawning, casing mixture were prepared in the ratio of 1:1 by using Garden soil (pH 5.3) and two years old farmyard manure (FYM, pH 6.45). Garden soil was taken from the glasshouse field, Department of Plant Pathology and their pH was analysed by using Elico-pH meter in the laboratory of the Department of Soil Science and Agricultural Chemistry. The casing media were prepared by through mixing of the selected substrates in the proper ratio and was chemically sterilized by spraying with 2 percent formalin and then covered with polythene sheet for 3 days. The media were turned on alternate days for 4 days to remove the fumes of formalin from the casing mixture. The selected substrates taken to prepare casing media were mixed in volume-by-volume basis and slightly moistened after casing and then applied to create casing layer thickness of 2.0 cm. After casing beds were kept on racks in cropping room for fruiting. During this period the temperature and humidity were maintained. Observations were recorded.

Cropping room

The temperature and relative humidity for fruiting were kept 30-35 °C and 80-85 percent, respectively. After casing of beds ventilation was reduced. Watering was done two times a day by a hand sprayer and it was withheld a day before harvesting. The yield data were recorded for a period of 35 days.

Harvesting

Pinheads appeared after casing and harvesting was done one week after pinning. Fully matured sporophores of milky white mushroom were harvested from the beds and fresh weight was determined immediately. Likewise, each bed was harvested for 2 cropping in period of 26 days after the first harvesting.

Observations recorded

Yield attributing characters included were as follows:

Number, size, weight of sporophores length, diameter of stipe and diameter of pileus and yield were recorded after harvest of mushroom.

Biological efficiency

Biological efficiency of mushroom was calculated by using formulae as recommended by Chang and Miles (1989).

$$\text{Percent biological efficiency} = \frac{\text{Fresh weight of mushroom (g)}}{\text{Dry weight of substrate (g)}} \times 100$$

Results and Discussion

The experiment was carried out to know the effect of variation in casing thickness on various parameters on crop duration and yield of *Calocybe indica*

Growth and yield parameters

Growth and yield parameters such as pinhead initiation (days), first picking, number of pinhead per bed, number of sporophore per bed, yield per bed and biological efficiency were studied. The result showed that the earliest pinhead initiation (7 days) after casing was observed from casing layer of 1.0 to 2.0 cm and latest (11.66 days) after casing from 4.5

cm thick casing. There were no significant difference in time taken for pinhead initiation after casing and first picking obtained in response to 1-2 cm casing thickness. The first picking of fruiting bodies was done 14 days after casing from 1-2 cm thick casing which was far earlier than first picking at 18.66 days after casing at 4.5 cm thickness. The maximum number of pinhead (22.66) per bed was observed in casing thickness of 2.0 cm which was found to be significantly superior to other treatments. Minimum number of pinhead (11.33) per bed was observed in 4.5 cm casing thickness. The maximum number of sporophores (14.67) per bed was harvested from 2.0 cm casing thickness which was found to be significantly superior to other treatments. Minimum

number of sporophore (9.33) was harvested in 4.5 cm casing thickness followed by 10 in 4.5 cm casing thickness. Maximum yield (1511.45 g) per bed was recorded in 2.0 cm casing thickness which was found to be significantly superior to other treatments. The minimum yield (771.77 g) per bed was recorded in 4.5 cm casing thickness followed by 850.60 g in 4.0 cm casing thickness. Maximum biological efficiency (100.76%) was recorded in casing thickness of 2.0 cm followed by 89.61 percent in casing thickness of 1.5 cm and 88.61 percent in casing thickness of 1.0 cm. Minimum biological efficiency (51.45%) was recorded in casing thickness of 4.5 cm. (Table 1, Fig 1 and 2)

Table 1: Effect of casing thickness on crop duration and productivity of *Calocybe indica*

Treatments	Period (Days) between casing &		Average No. of pinhead/bed	Average No. of Sporophore/bed	Average yield/bed (gm)	Biological efficiency (%)
	Pinhead initiation	First picking				
T ₁ - 1.0 cm	7.00	14.00	21.00	13.33	1329.26	88.61
T ₂ - 1.5 cm	7.00	14.00	21.66	13.33	1344.19	89.61
T ₃ - 2.0 cm	7.00	14.00	22.66	14.67	1511.45	100.76
T ₄ - 2.5 cm	7.66	14.66	19.68	12.66	1212.82	80.85
T ₅ - 3.0 cm	8.66	15.66	19.33	12.33	1085.40	72.36
T ₆ - 3.5 cm	9.66	16.66	19.00	11.67	1020.65	68.04
T ₇ - 4.0 cm	10.66	17.66	12.66	10.00	850.60	56.70
T ₈ - 4.5 cm	11.66	18.66	11.33	9.33	771.77	51.45
SEm ±	0.26	0.27	0.29	0.33	27.90	
CD at 5%	0.79	0.80	0.87	1.01	84.36	
CV%	5.26	3.04	2.71	4.66	6.44	

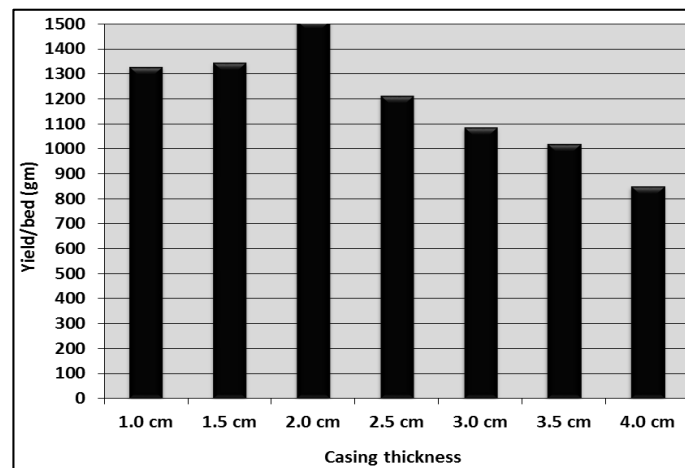


Fig 1: Effect of casing thickness on yield of *Calocybe indica*

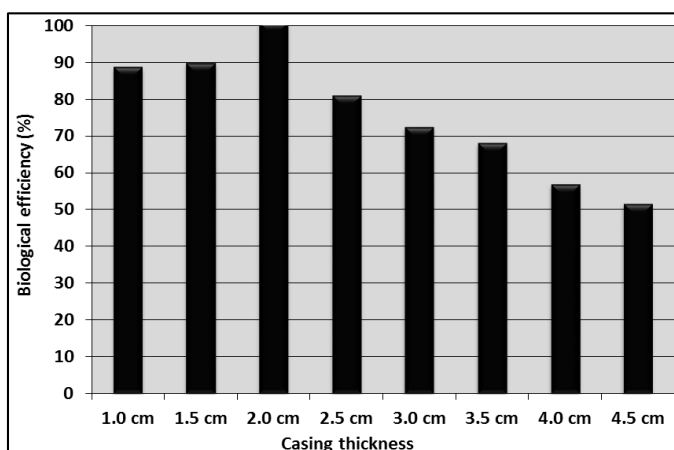


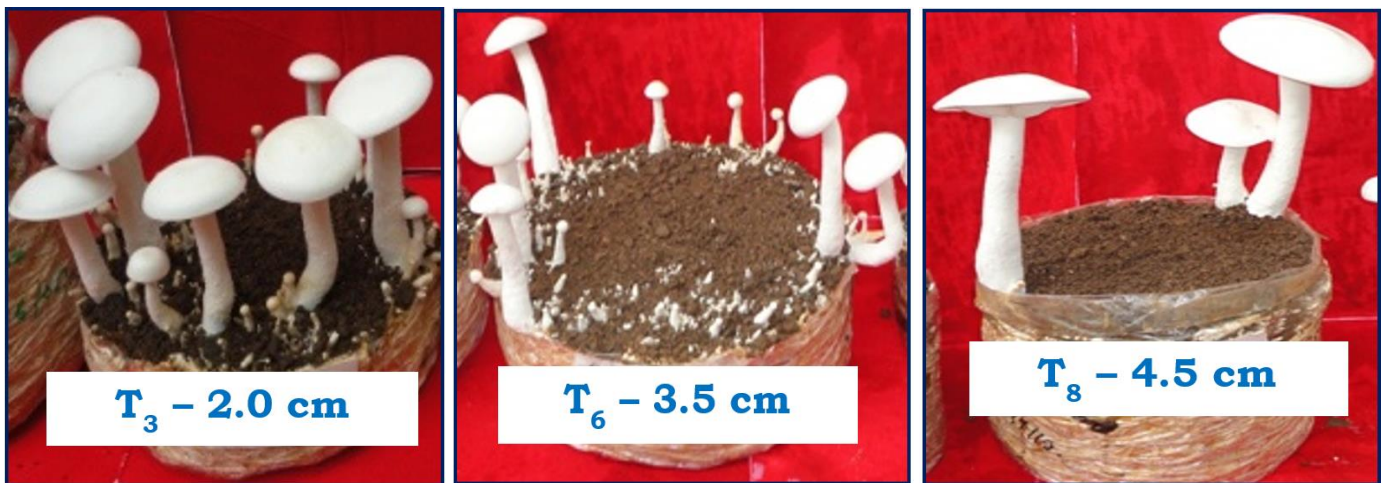
Fig 2: Effect of casing thickness on biological efficiency of *Calocybe indica*

Size and weight of sporophore

The result showed that the length of stipe, diameter of stipe and diameter of pileus in different treatments ranged from 9.20 cm to 14.40 cm, 5.40 cm to 7.75 cm and 8.35 cm to 11.40 cm respectively. Maximum length of stipe (14.40 cm), diameter of stipe (7.75 cm) and diameter of pileus (11.40 cm) were recorded in casing thickness of 2.0 cm which was found statistically at par with casing thickness of 1.5 cm and 1.0 cm. Maximum weight of sporophore (103.03 g) was recorded in casing thickness of 2.0 cm which was found statistically at par with casing thickness of 1.5 cm and 1.0 cm. The minimum weight of sporophore (82.72 g) was recorded in casing thickness of 4.5 cm. (Table 2, Plate 1)

Table 2: Effect of casing thickness on size and weight of sporophore of *Calocybe indica*

Treatments	Average length of stipe (cm)	Average diameter of stipe (cm)	Average diameter of pileus (cm)	Average weight of sporophore (gm)
T ₁ - 1.0 cm	13.44	6.55	10.36	99.72
T ₂ - 1.5 cm	13.47	6.63	10.45	100.84
T ₃ - 2.0 cm	14.40	7.75	11.40	103.03
T ₄ - 2.5 cm	12.22	6.43	9.84	95.80
T ₅ - 3.0 cm	10.92	6.39	9.68	88.03
T ₆ - 3.5 cm	10.58	6.15	9.34	87.46
T ₇ - 4.0 cm	9.82	5.66	8.79	85.06
T ₈ - 4.5 cm	9.20	5.40	8.35	82.72
SEm ±	0.32	0.44	0.35	1.52
CD at 5%	0.98	1.31	1.04	4.68
CV%	4.59	7.19	5.66	3.46

**Plate 1:** Fruiting bodies of *Calocybe indica* grown on different casing thickness

The thickness of casing layer not only affects initiation of fruiting bodies and their development but also the duration of crop and fruiting body yield. In the present studies, observations indicated that the earliest pinhead initiation (7 days) after casing was observed from casing layer of 1.0 cm to 2.0 cm and latest from 4.5 cm thick casing. In general it was observed directly proportional to the casing thickness. Pinhead initiation was observed only from the margins of casing layer touching polythene at the thickness of 4.0 cm and 4.5 cm. It can alter the crop duration by 6 to 7 days. There was no significant difference in time taken for pinhead initiation after casing and first picking obtained in response to 1.0-2.0 cm casing thickness. Casing thickness of 2.0 cm was ideal for higher sporophore number and maximum yield and biological efficiency. Thickness of casing layer beyond 2.0 cm gradually decreased the mushroom yield and delayed the appearance of sporophores. Least weight and size of sporophores was recorded in the substrate with maximum casing depth (4.5 cm). Similar result were reported by Pani (2012) [4] and Shukla (2007) [10]. Deb (2002) [2] also reported that 1.0 cm thick casing gave maximum yield, whereas, Sharma *et al.* (1997) [9] suggested that the number of fruiting bodies harvested per bed were less than 8.0 and 11.0 respectively at and above the casing thickness of 2.5 cm. Sassine *et al.* (2005) [8] suggested that the casing should be very loose, otherwise the primordia cannot penetrate from the bottom to the top of the casing layer.

The present study concluded that the earliest pinhead initiation (7.00 days) was observed from casing layer of 1.0 to 2.0 cm and latest (11.66 days) from 4.5 cm thick casing.

Pinhead initiation was observed only from the margins of casing layer touching polythene at the thickness of 4.0 cm and 4.5 cm. However, there was no significant difference on days for first picking from the day of pinhead formation obtained in response to 1-2 cm casing depth. Casing thickness alone can alter the crop duration by 6 to 7 days. The first picking of fruiting bodies was done 14 days after casing from 1.0-2.0 cm thick casing which was far earlier than the casing thickness of 4.5 cm in which first picking was done 18.66 days. Casing thickness of 2.0 cm was ideal for higher sporophore, maximum yield and biological efficiency. Thickness of casing layer beyond 2.0 cm gradually decreased the mushroom yield and delayed the appearance of sporophores.

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