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# Effect of Different Casing Materials on the Production of Button Mushroom (*Agaricus bisporous* L.)

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Article History Received March 18, 2015

**Published Online** May 05, 2016

#### **Keywords:**

Button mushroom, Fruiting bodies, Moisture content, Mycelial growth, Nitrogen content **Abstract:** The experiment was conducted to study the effect of different casing materials on the production of the button mushroom. Results showed that T3 (Lahore compost) and T2 (FYM+sand+lime) were significantly different from the T1 (Clay + Sand + lime) and T0 (peat soil / control) while T1 was also significantly different from T0. The moisture contents in T0, T1, T2 and T3 were 45.95%, 43%, 59.30% and 60.30% respectively. The Nitrogen contents were recorded as 67.18%, 55.60%, and 21.53% higher in T2, T3 and T1 respectively as compared to the control. The time is taken to complete mycelial growth in T3, T2, and T1 were 11.50, 10.75, and 9.50 days respectively. The maximum time taken for completion of mycelial growth was recorded in T3 (19.25days) and the minimum was recorded in control (10days). The time is taken to reach harvesting stage recorded in T3, T2, and T1 were 14.75, 9.50 and 8.75 days respectively, furthermore T1 and T2 were similar to each other but dissimilar to control and T3. T2 produced a maximum number of pinheads (15.50), mature fruiting bodies (14.25) and biomass (369.00g), followed by T1 while control and T3 were at par with regard to the number of mature fruiting bodies.

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Cite this article as: Rehman, M.K., M.A. Ali, A. Hussain, W.A. Khan and A.M. Khan. 2016. Effect of different casing materials on the production of button mushroom (*Agaricus bisporous L.*). *Journal of Environmental & Agricultural Sciences*. 7: 55-61.



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#### 1. Introduction

Mushrooms have been under the human consumption since long and have a good aroma and taste (Kurbanoglu and Algur, 2002). Edible mushrooms are an important constituent of a healthy diet as they have high protein, minerals, vitamins and essential amino acids and used for therapy of many diseases such as cancer and heart disease (Taherzadeh and Jafarpour, 2013). Mushrooms have become more and more important food stuff because of their possible preventive roles against human life style-related diseases such as hyperlipidemia and diabetes (Omowumi and Mukaila, 2013).

Globally button mushroom (*Agaricus bisporus* L.) is widely cultivated and consumed mushroom which contributes 40% of mushroom production contributing to about 40% of the total world mushroom production (Giri and Prasad, 2007). Button mushroom is a basidiomycete fungus, its fruiting bodies are produced in large scale on casing

layers containing specific compost (De Groot et al., 1998).

Wheat straw, chicken manure, straw-bedded horse manure, and gypsum are components of compost used for the production of the white button mushrooms (Straatsma et al., 2000). In commercial mushroom cultivation casing is used to cover the compost, usually after the substrate colonized by mushroom mycelium to encourage fructification. Selection of casing material is vital for the mushroom production as it significantly influences mushroom quality and yield, its availability and price.

In the artificial button mushroom culture, casing layer is an essential part of the total substrate. In addition to the genetic factors mushroom, mycelial growth and development also depend on microbiological, chemical composition and environmental conditions (Pardo et al., 2004). Casing soil protects the compost against desiccation, supports the mushroom against pests and diseases, provides support for developing sporophores, and provides a

gas exchange for development and growth of mushrooms (Colauto et al., 2011). Casing layer provides modified environmental condition which favors the induction of reproductive development of mushroom from vegetative stage mainly attributed to the increased microbial activity in the casing material (Gulser and Peksen, 2003). Bacteria in the casing soil influence productivity, product quality and uniformity (Choudhary, 2011). The required physical and chemical properties of a good casing should possess.

Adequate water holding capacity (WHC) and porosity are basic requirements of good casing material. Moreover, slightly basic pH (7.2-8.2), low content of soluble inorganic and organic nutrients, pathogen and pest free casing material favor mushroom production. Various materials, either single or in combinations, have been used as casing both commercially and experimentally, however only very few have been shown promising results to be of practical application (Gulser and Peksen, 2003). Among the most suitable casing medium peat is widely used mainly due to its appropriate WHC, organic matter content, nutrient supplying capacity, neutral pH, and structural properties e.g., granular structure. Importantly, peat stays porous and provides appropriate gaseous exchanges and supports microbial population even in moisture saturated condition and its ability to release hormone-like substances, which are very likely involved in the induction of fruiting body formation (Colak, 2004).

problems associated with However, the availability of peat have led to the search for alternative materials (Colauto et al, 2010). Peat is not available in some areas and soil can be used as casing soil. The soil is also accessible throughout the year and its low prices have benefits such as easy management (Zied et al, 2010). Spent mushroom compost as a casing soil material can be used to reduce the cost of production and it can also reduce pollution (Pardo-Giménez and Pardo-González, 2008). Successful cultivation of mushroom often requires good casing material after spawning. The current research was therefore undertaken to study the effect of various casing materials on the productivity of button mushroom, which will be helpful for further improvements in the yield.

# 2. Materials and Methods

The compost used in the study was collected from a private farm i.e., Asjid Mushroom Faisalabad. The compost was prepared by using wheat straw (1000 kg), chicken manure (400 kg), gypsum (28 kg) and urea (7 kg).

# 2.1 Procedure for the preparation of compost

Wheat straw was spread over the ground and sprinkled with water thoroughly for 4 days in order to remove waxy coating film of the straw so that water can penetrate into the substrate. All ingredients, except gypsum, were mixed in the wet straw, which was stacked into a pile about one-meter width and one-meter length. The heap was compressed by applying light pressure, in order to open the pile and for the uniform composting process. Gypsum was added on 3rd and 4<sup>th</sup> turning in two split doses.

# 2.2 Pasteurization technique

Compost was pasteurized in the room, where bed temperature of compost was gradually increased to 58-60°C, through steam generator placed outside the room. The temperature was kept maintained for six hours.

### 2.3 Spawning

Spawn was applied to the trays @ 10gm having 750gm substrate per tray. The size of the tray was (3x7) inches and depth was 3 inches. Mycelial growth was observed after spawning.

### 2.4 Casing treatments

Three type of casing materials were applied @ 45 mm thickness. The temperature of compost was maintained at 22-25°C which is optimum for mycelial colonization of casing layer. Appropriate relative humidity is important for the ideal growth of fungus, which was maintained at 80-90 %. Treatments included, T0: Normal casing treatment (peat soil) (control); T1: Clay + Sand + lime (2:1+4%); T2: FYM + Sand + lime (2:1+4%); T3: Lahore compost (contained 2.5 %N, 1%P, 1%K, and other micro nutrients)

### 2.5 Moisture contents of the casing

The moisture content of casing materials before spawning was recorded on both fresh and dry weight basis. To measure the moisture content of the casing, 5grams of the sample were kept in the oven for 48 hours at 50°C up to a constant weight. Moisture percentage was calculated by the difference in fresh and dry weight as described in the following formula

Moisture %age of the casing material = (fresh weight – dry weight)/ fresh weight  $\times$  100

# 2.6 Determination of nitrogen percentage of casing materials

Total nitrogen (N) was determined according to A.O.A.C (1990) method which involved digestion of casing material with a concentrated H<sub>2</sub>SO<sub>4</sub> and digestion mixture, containing 10:0.5:1 ratio of K<sub>2</sub>SO<sub>4</sub>, CuSO<sub>4</sub>, and FeSO<sub>4</sub>. Digestion was performed by adding one gram of oven dried casing powder in

Kjeldahl digestion flask along with 20ml concentrated  $H_2SO_4$  and digestion mixture (5 g). The mixture was kept for 30 minutes, subsequently, it was gradually heated to obtain green transparent liquid material. After cooling, contents were shifted in a volumetric flask (100 ml) and volume was made up to the mark.

Aliquot from this prepared material was taken and distillation was performed in the micro-Kjeldahl apparatus by adding 40% NaOH. Ammonia vapors were obtained in 4% boric acid and mixed indicators of bromocresol green and methyl red. This distillate was titrated against H<sub>2</sub>SO<sub>4</sub> (0.1 N) till the original color of methyl red was recorded. N percentage was calculated by using the following formula:

 $A\text{-}B\times100\times100\times0014/Volume of digested sample used$ 

A= volume of N/10  $H_2SO_4$  used, B= blank reading, 100=volume made after digestion, 100= for percentage, 0.0014=factor (which is equal to gram of N present in 1 mol. of 0.1 N  $H_2SO_4$ )

### 2.7 Mycelial growth studies

Linear mycelial growth characters were also observed on the compost kept in trays and plastic bags after casing. The average of mycelial growth was calculated.

Data was recorded to check the different parameters to compare the mycelial growth in different casing materials used. The following parameters were:

- time required for the completion of mycelial growth, a period of time taken for the appearance of fruiting bodies after casing
- the time period to reach fruiting bodies at a harvesting stage after casing the number of fruiting bodies as pinhead after casing
- total weight of fruiting bodies (g) after casing the number of mature fruiting bodies after casing

### 2.8 Statistical analysis

Data was subjected to statistical analysis by constructing an analysis of variance (ANOVA) tables. Least Significant Difference test (LSD) at 5 % probability level was used to compare mean values for various treatments with a significant difference (Steel and Torrie, 1997).

### 3. Results and Discussion

# **3.1.** Influence of casing material pH on mushroom growth

Like for other fungus mushroom growth is significantly influenced by pH of growth medium. ANOVA (Table 1) revealed that different casing materials were significantly different in their pH ranges. As shown in Fig.1, maximum pH was recorded in T3 (8.44), followed by T2 (8.41) and T1 (8.05). Minimum pH was observed in control (6.65). T3 and T2 were at par, while significantly different from T1 and T0. T1 was also significantly different from T0. The pH has been reported to be correlated negatively with the mushroom yield (Jarial and Shandilya, 2004).

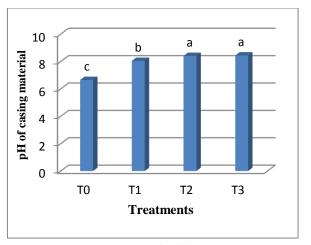


Fig. 1. Variations in pH of different casing materials (means sharing different letters differ significantly).

**Table 1: ANOVA for Statistical Analysis** 

	Mean Squares of Parameters									
SOV	d.f.	pН	MC	N	DMG	DFB	DH	PinN	FBW (g)	MFB
				Conc.	(days)	(day)	(days)			
Treatments	3	2.85**	315.53**	2.56**	10.75**	60.06**	58.57**	47.73**	53850**	46.20**
Error	12	0.016	0.378	0.015	1.125	4.021	1.125	3.313	1832.292	3.458

DFB: Days for appearance of fruiting bodies; DH: Days to reach harvesting stage after casing; DMG: Days for completion of mycellial growth in days; FBW: Total weight of fruiting bodies; MC: moisture contents; MFB: number of mature fruiting bodies after casing; N Conc.: Nitrogen concentration; PinN: Number of pinheads after casing.

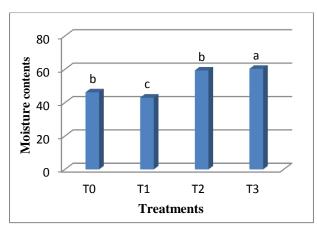


Fig. 2. Moisture contents of different treatments (means sharing different letters differ significantly).

# **3.2.** Influence of casing material moisture contents on mushroom growth

Water is a basic need for the mycelial growth and for pinhead formation. Analysis of variance showed that different casing materials were significantly different from each other for moisture content (Table 1). Fig.2 shows that moisture content decreased from T3 to T1, (60.3%, 59.3%, and 43.0% respectively), where as in control treatment, moisture content (46.2%) were higher than T1. After applying LSD test, results showed that treatments were significantly different from each other. Optimum moisture contents for the mushroom growth were 60-65%. Moisture contents were in range in the T3 and T2 while in T1 and T0 moisture contents were slightly lower than the recommended value as reported by Nobel et al., (1999) and Jarial and Shandilya, (2005).

### 3.3 Determination of nitrogen contents

Different casing materials significantly varied in their N contents. High N contents significantly affect the mushroom growth. ANOVA presented in Table 1 revealed that different casing material was significantly different from each other. As shown in Fig-3, maximum N content was recorded in T2 (4.33%) followed by T3 (4.03%) and T1 (3.15) while minimum N contents were observed in control (2.59%). After applying LSD test, results showed that all treatments were significantly different from each other. Our results are in agreement with the findings of Demirer et al., (2005), who found that maximum mushroom production w obtained by high application dose of calcium ammonium nitrate followed by ammonium nitrate.

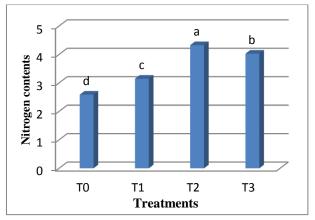


Fig. 3. Nitrogen contents of different treatments (means sharing different letters differ significantly).

# 3.4 Days to completion of mycelial growth

Data showed that all treatments were significantly different from each other on the basis of time taken to complete the mycelial growth. Time taken to complete mycelial growth in T3, T2 and T1 was 11.50, 10.75 and 9.50 days respectively (Fig. 4). Results showed that T2 and T3 were statistically similar to each other while T1 and T2 remained at par. T0 was statistically different from all other treatments and took the lowest time for mycelial growth. The late growth in other treatments was due to low moisture contents and higher N level. Our results support the conclusion of Ram and Kumar (2010) who reported that within 20 days mushroom bags were completely colonized by mushroom mycelium.

### 3.5 Duration of appearance of fruiting bodies

The casing treatments imparted differential impact on the time of appearance of fruiting bodies (Fig. 5).

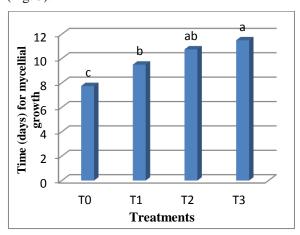


Fig. 4. Mycelial growth under different casing material (means sharing different letters differ significantly).

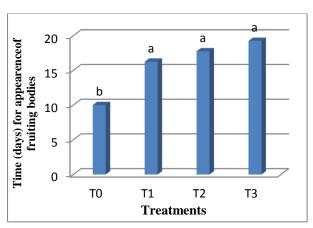


Fig. 5. Time (days) for the appearance of fruiting bodies of different treatments (means sharing different letters differ significantly).

The maximum time was taken (days) for completion of fruiting bodies was recorded in T3 (19.25days), followed by T2 (17.75 days) and T1 (16.25 day) with no significant difference. Minimum time (10days) was recorded in control (T0), which was significantly different from all other treatments. Observed late mushroom growth can be attributed lower moisture content supplemented with high N contents. These results are in close agreement with the findings of Khan et al (1991). The micro flora presents in the casing soils play a vital role in the mushroom fruiting body initiation and development (Vijay and Gupta, 1992; Fermor et al., 2000).

### 3.6 Days to reach harvesting stage after casing

Significant differences were observed among the time requirement for treatments to reach the harvesting stage. The maximum time was recorded in T3 (14.75days), followed by T2 (9.50days) to complete mycelial growth (Fig.6).

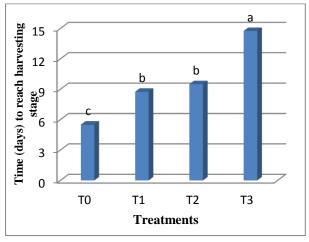


Fig. 6. Times (days) to reach the harvesting stage of different treatments (means sharing different letters differ significantly).

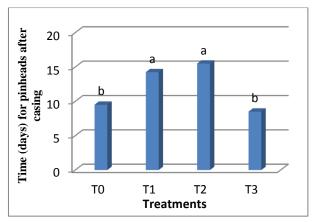


Fig. 7. Time (days) for pinheads after casing of different treatments (means sharing different letters differ significantly).

After reaching a maximum number, fruiting bodies declined slowly over harvest of successive flushes of mature fruiting bodies. In our experiment, the late growth of mushroom was recorded due to the high N level and moisture deficient conditions which delay growth and development. Similar findings were presented by (Kalberer, 1995).

### 3.7 Number of pinheads after casing

The treatment had a significant impact on the number of the pinheads. A maximum number of pinheads were observed in T2. This may be due to the reason that its casing material had farmyard manure (FYM), which supply adequate nutrients especially N (Fig. 7). Our results are accordance with Nobel et al., (2003) depicted that peat casing showed good results of primordia initiation than in other casing materials.

### 3.8 Total weight of fruiting bodies after casing

Biomass depends on media composition and growth conditions. ANOVA Table 1 revealed that casing treatments used in this study had a significant impact on the total biomass production. T1 and T2 were similar to each other but differed with control and T3. Significant differences were also noted between T3 and control. The amount of biomass produced in T2, T1, control and T3 was 369.00g, 317.75g, 288.00g, and 104.25g respectively (Fig. 8).

T2 produced maximum biomass due to the presence of FYM in its casing material which provides adequate nutrients to the button mushroom. Similar results were observed by Colak (2004), who obtained a high yield of button mushroom by using wheat straw casing material. Our results are also supported by Shandilya (1989) who reported that maximum yield of mushroom was achieved with casing material having farmyard manure.

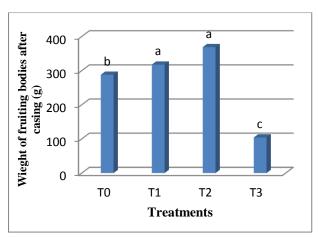


Fig. 8. Weight of fruiting bodies after casing (g) of different treatments (means sharing different letters differ significantly)

# 3.9 Number of mature fruiting bodies after casing

The casing treatments imparted differential impact on the time to complete mycelial growth. Results presented in Fig. 9 showed that the maximum number of fruiting bodies was observed in T2 (14.25), followed by T1 (12.50), while T3 and control produced 7.75 and 7.5 respectively. Results revealed that casing material containing farmyard manure produced a maximum number of fruiting bodies. Results indicated that T1 and T2 were remained at par to one another, but significantly different from other treatments i.e., control and T3. Our results are well supported by the findings of Pardo et al., (2004). Taherzadeh and Jafarpour (2013) reported that treatments having different casing materials were significantly different from each other for the production of the fruiting bodies.

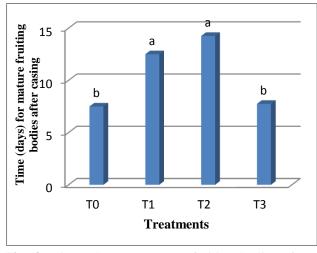


Fig. 9: Time (days) to mature fruiting bodies after casing of different treatments (means sharing different letters differ significantly).

#### 4. Conclusion

Mushrooms had long been used for medicinal and dietary purposes since decades. Our study was aimed to compare the effect of different casing materials on the productivity of button mushroom. Our results were consistent with the previous findings but also can serve as a valuable guideline for future studies on casing materials for mushroom production.

**Acknowledgements:** Authors acknowledge Mr.Mueen Alam Khan, Assitant Professor, Islamia University Bahawalpur (IUB) for constructive comments and suggestions on the manuscript.

**Competing Interests:** The authors declare that there is no potential conflict of interest.

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