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# The Use of Five Manures as An Ingredient In The Making of Straw Mushroom (*Volvariella volvaceae*) spawn

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**ABSTRACT**

The straw mushroom, *Volvariella volvaceae* is delicious and very popular among consumers. It is widely cultivated on the local and industrial scales in tropical and subtropical areas. In Thailand, spawn growers use animal manure, particularly horse manure as a mixture in spawn making. The objective of this research was to determine effects of five animal manures as an ingredient in the making of straw mushroom spawn. The experiment was conducted at Rajamangala University of Technology Suvarnnabhumi, Phra Nakhon Si Ayutthaya, Huntra by means of Complete Randomized Design (CRD) with five replications of five treatments. It was found that the mycelial growth on the horse and the goat manures for thirteen days was the most abundant. In addition, the growth of mycelia in the culture bowl which colonized four days after inoculation on the goat, buffalo and horse manures was rapid. As for the fructification level, six days after inoculation on the horse manure was found to be best with its value of 4.8. The results showed statistically significant differences.

*Key words : Volvariella volvaceae*, straw mushroom spawn, animal manure

## INTRODUCTION

The straw mushroom, *Volvariella volvaceae* is one of the popular mushrooms in Southeast Asia. Cultivation of paddy straw mushroom using paddy straw is the oldest and commonly used technique but it gives very low mushroom yield, 5-15% of dry substrate. Although the yield is not high, paddy straw was practically the only material used to prepare the substrate for cultivation of the Paddy Straw Mushroom under natural conditions, whereas other substrates had been successfully used. Thailand has plenty of straw and can produce numerous straw mushroom. The stages of development of the straw mushroom start with tiny clusters of white hyphal aggregates called primordia known as pinheads and it is followed by several morphological stages in the fruiting body development process [5]. The successive stages are named as "button", "eggs", "elongation", "mature" stages respectively. The nutrient requirement of mushroom are carbon, nitrogen, element, vitamin and growth promoting activity substance. A variety of waste material has been used for straw mushroom cultivation including spawn making [1,9,2,3]. Zakhary *et al* [10] performed seven different media for the cultivation of paddy-straw mushroom, using local materials such as orange juice waste, rice straw, bagasse, pea-canning waste, horse manure and molasses. The good quality *of V. volvaceae* spawn is essential for farmers and the mushroom growers. Spawn producing is a highly specialized part of mushroom cultivation [8]. Little work has been done on *V. volvaceae* spawn making. In Thailand, horse manure was usually used as an ingredient in the straw mushroom spawn making as a traditional source of nitrogen. The spawn in Thailand can be produced simplified. However, Farmers do not want to do mushroom spawn itself . Most of them live in the countryside and some areas had no horse manure. Thus, the purpose of this research was to determine effects of five animal manures as an ingredient in the making of straw mushroom spawn.

### Materials and Methods

The research was carried out at Plant Science Section, Faculty of Agricultural Technology and Agro-industry, Rajamangala University of Technology Suvarnnabhumi, Phra Nakhon Si Ayutthaya, Huntra, Thailand by means of Complete Randomized Design (CRD) with five replications of five treatments. Five animal manures: cow, goat, buffalo, horse and elephant were determined as an

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ingredient in the making of straw mushroom spawn. The growth of mycelia was observed on the spawn bags including the mycelial growth and the fructication level was heeded in the culture bowl.

*Isolation of Volvariella volvaceae:*

Mycelial culture used in this research was obtained by tissue culture of *Volvariella volvaceae* stipes collected from Phra Nakhon Si Ayutthaya, Thailand, and established on potato dextrose agar medium. The medium comprising extract from 200 g. of potato, 20 g, agar and distilled water. 1 litre was sterilized using autoclave at 15 p.s.i. (121 $^{\circ}$C) for 15 min.

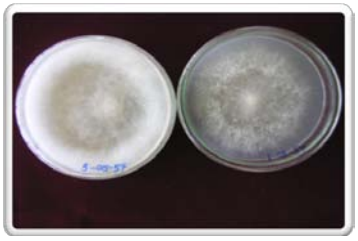

**Fig. 1:** Mycelia of *Volvariella volvaceae* (straw mushroom)

*Preparation of compost:*

*Ingredients:*

4 kg. dried mung bean shell
4 kg. cotton waste
0.5 – 1 kg. rice bran
0.25 kg. calcium carbonate
0.25 kg. molasses
0.5 kg. animal manure

The compost was produced using dried mung bean shell, cotton waste, rice bran, calcium carbonate, molasses, animal manure and water in the ratio of 16:16:4:1:1:2:6 respectively. Dried mung bean shell was soaked and mixed with animal manure and molasses for 10-20 minutes. The five animal manures; cow, goat, buffalo, horse and elephant were seperately mixed with other substrate as five treatments. Those mixture dampen was incorporated with kapok filling, rice bran and calcium carbonate and then stacked. The compost pile was covered with a transparent polyethylene sheet for 5 days to induce the bioactivity of thermophilic decomposers. The compost pile was daily turned over to prevent the development of anaerobic conditions within the stack and to allow the microorganisms exist in the raw materials to begin to degrade the stack.

*Spawn of V. volvacea:*

The composted substrates were put into the autoclavable polypropylene bags which plugged with cotton were took place in 200-litre container for steaming at 100 $^{O}$ C for 2 hour. The *V. volvacea* inoculated spawns were incubated at room temperatures for mycelial growth. The aseptic technique was used in all processes.

*Evaluation of mycelia running on spawn:*

Investigation mycelia running on spawn after inoculation of *V. volvacea* by measuring of mycelia length daily.

*Evaluation of mycelial growth and fructification level in culture bowl:*

The crushed spawn was placed in the bowl. The determination was undertaken by observing the mycelial growth and the occurrence of fruiting body after four days and six days of *V. volvacea* inoculation using following schematics:

1 = no mycelium or no fruitingbody (FB)
2 = possing 1-5 point of mycelia or FB
3 = possing mycelia or FB ¼ of cuture bowl
4 = possing mycelia or FB ½ of culture bowl
5 = possing mycelia or FB > ½ culture bowl

*Data Analysis:*

The result of the experiment was analysed to check the variance and the means of mycelial growth and fructification level were compared to find significant difference at 95 % level by Duncan ′s multiple range Test (DMRT) .

*Results:*

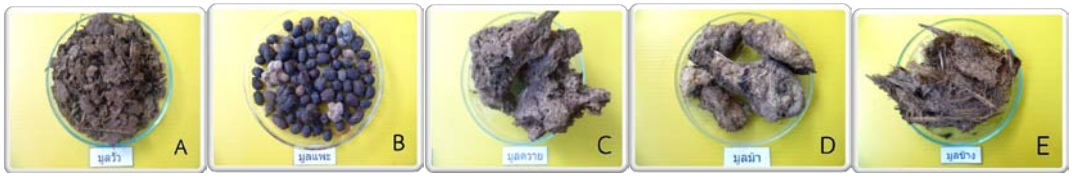


**Fig. 2:** Animal manure A : cow , B : goat , C: baffalo , D : horse and E : elephant

**Table 1** Average Nitrogen (N), Phosphorous ($P_2O_5$) and Potassium ($K_2O$) content of five animal manures in percentage (%).

| dried manure | Average fertilizer quantitive in Percentage (%) | | |
|---|---|---|---|
| | N | $P_2O_5$ | $K_2O$ |
| Cow[1] | 1.10 | 0.4 | 1.60 |
| Goat | 1.03 | 0.66 | 0.64 |
| Buffalo[1] | 0.97 | 0.60 | 1.66 |
| Horse | 2.33 | 0.83 | 1.31 |
| Elephant | 1.18 | 0.53 | 0.89 |

(Source : [1] Buro of rice research and Development, 2013)

According to Table 1, the horse' s maure is the richest sourse of nitrogen, phosphorus and potassium among all animal manures (Table 1).

*Evaluation of mycelia running on spawn:*

**Table 2** Periods (days) for mycelia running reaching to the bottom of the spawn after inoculation

| Treatments | Days post inoculation | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
| cow | - | - | - | - | 0.7 | 2.2 | 3.6 | 5.5 | 7.6 | 9.5 | 11.5 | 13.2 | 14.9 | 16.0 |
| goat | - | - | - | 1.2 | 2.6 | 4.1 | 5.8 | 7.8 | 8.7 | 10.8 | 12.7 | 14.5 | 16.0 | |
| baffalo | - | - | - | 0.9 | 1.8 | 3.5 | 5.3 | 7.4 | 9.0 | 11.0 | 12.8 | 14.3 | 15.7 | 16.0 |
| hourse | - | - | - | 1 | 2.0 | 4.0 | 5.9 | 7.8 | 9.6 | 11.7 | 13.5 | 15.5 | 16.0 | |
| elephant | - | - | - | - | 0.9 | 2.2 | 3.7 | 5.6 | 7.7 | 9.5 | 11.3 | 13.1 | 14.8 | 16.0 |

It was found that the mycelial growth on the horse and the goat manures for thirteen days was the most abundant (Table 2 and 3). One of the major problems of spawn making is the contamination during the incubation period of inoculated spawn. In general, The faster mushroom mycelia run, the lesser contamination is.

**Table 3:** Average days for mycelia running reaching to the middle and to the bottom of l spawn after inoculation

| Length | Average days for mycelia running (days) | | | | |
|---|---|---|---|---|---|
| | cow | goat | buffalo | horse | elephant |
| In the middle of the spawn | 9 - 10 | 8 - 9 | 9 - 10 | 8 - 9 | 9 - 10 |
| In the bottom of the spawn | 14 | 13 | 13-14 | 13 | 14 |

*Evaluation of mycelial growth and fructification level in culture bowl:*

The growth of mycelia in the culture bowl which colonized four days after inoculation on the goat, buffalo and horse manures was rapid (Table. 4). As for the fructification level, six days after inoculation on the horse manure was found to be the best with its value of 4.8 (Table 4). The results showed statistically significant differences. Although the horse manure gave the best as an ingredient in the making of straw mushroom spawn, the other animal manure such as the buffalo and cow manure were also used.

**Fig. 3:** Fruiting bodies of *Volvariella volvaceae* (straw mushroom) in culture bowl

**Table 4:** Average level of mycelia growth and fructification in the culture bowl four days and six days post inoculation (dpi)

| Treatments | mycelia growth level<br>4 days post inoculation | fructification level<br>6 days post inoculation |
|---|---|---|
| cow | 4.60 ab | 4.00 abc |
| goat | 5.00 a | 3.40 bc |
| buffalo | 5.00 a | 4.40 ab |
| horse | 5.00 a | 4.80 a |
| elephant | 4.40 b | 3.00 ab |
| C.V. (%) | 7.22 | 19.09 |

Different letters represent statistically significant difference at P < 0.05 by DMRT

Mycelia growth and fructification level :1= no mycelium or no fruitingbody (FB), 2 = possing 1-5 point of mycelia or FB, 3 = possing mycelia or FB ¼ of cuture bowl, 4 = possing mycelia or FB ½ of culture bowl and 5= possing mycelia or FB > ½ culture bowl.

*Discussion:*

Mushroom are heterotrophic organisms which require an external source of organic carbon. They need the essential elements: carbon, nitrogen, sulfur, phosphorus, potassium, magnesium and some elements which are required in lesser quantities and that are known as trace elements [6]. The data (Table 1) suggested that the hourse manure provide high nitrogen content than the other animal manures, including the amount of Phosphorus. Phosphorus is very beneficial to the fungal decomposer.

A major problem for mushroom growers is lack of good quality spawn. Some spawns were infected by other microorganisms due to non aseptic condition. In Thailand, there are the following process. :

Subculture ⟶ mother spawn (compost) ⟶ planting spawn (compost) ⟶ bed ⟶ fruits

*V. volvaceae* mother spawn is generally made up of cotton waste, animal manure, rice bran and lime. It is inoculated with an mycelial agar plug after sterilization. The planting spawn were usually prepared by using the composts which were steamed at 100 $^{O}$c for two hrs. Thus, if the mycelia on spawn run fast, it is a good chance to decrease the risk of loss. The local farmers often use this method because the pressure cooker is expensive for them. The substrates for preparing *V. volvaceae* planting spawn adapted according to the availability of area. Some places where has no such material, the small spawn growers try to find other substrates, or give up to make the spawn. Thailand has plenty of agricultural waste materials such as rice straw, palm waste, water hyacinth etc. There are many kinds of livestock vary to the regions in Thailand, hence It will be an alternative applying of baffalo and cow manure as an ingredient in the spawn making.

*Conclusion:*

It can be concluded that the horse manure achieved the best as an ingredient in the making of straw mushroom spawn. However, the buffalo and cow manure can be used as a substitute for the spawn making.

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