Effect of Different Composting Periods of Empty Fruit Bunch (EFB) on the Production of *Volvariella volvacea*

Fatin Nabilah Fisol[1], Sumaiyah Abdullah[1*], Mahmud Tengku Muda Mohamed[2], Azizah Misran[2], Azhar Mohamad[3], Mohamad Yuzaidi Azmi[4] and Khalisanni Khalid[5]

[1]Department of Plant Protection, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia
[2]Department of Crop Science, Faculty of Agriculture, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia
[3]Malaysian Nuclear Agency, Bangi, 43000 Kajang, Selangor, Malaysia
[4]Department of Agriculture, Padang Terap, 06300 Kuala Nerang, Kedah, Malaysia
[5]Malaysian Agricultural Research and Development Institute (MARDI), 43400 Serdang, Selangor, Malaysia

*Corresponding Author : sumaiyah@upm.edu.my

Abstract

Volvariella volvacea (*V. volvacea*) or paddy straw mushroom has been cultivated worldwide due to its high nutrient content and pharmaceutical value. One of the factors affecting the cultivation of *V. volvacea* is the substrate materials used as mushroom beds, for example, composted empty fruit bunch (EFB). However, there is still no reported trial on the most suitable composting periods applied on EFB in order to achieve high production of *V. volvacea*. Therefore, the objectives of this study are to investigate the relationship between the composting period of EFB and the production of *V. volvacea*, and to select the best composting period for EFB. In this study, EFB composted for six, nine, and twelve days were selected as substrates for mushroom beds. After harvesting, the weight of *V. volvacea*, which represents their production, was recorded. The results showed that there was no significant difference in the mean weight of *V. volvacea* between all treatments.

Keywords: composting periods, cultivation, spawn, substrate, *Volvariella volvacea*

1. Introduction

The cultivation of edible mushrooms became successful worldwide due to several factors, such as prevailing external climatic conditions, short growing times, low input requirements, and low investment needed due to high availability of agriculture waste. The examples of edible mushroom species that being cultivated in most countries are Oyster mushroom (*Pleurotus* spp.), Ling Zhi (*Ganoderma* spp.), Black Jelly (*Auricularia* spp.), Button mushroom (*Agaricus* spp.), Shiitake (*Lentinus endodes*) and also Paddy Straw mushroom (*Volvariella volvacea*) [14]. According to the Department of Agriculture, Malaysia, the cultivation practise and production of *Volvariella volvacea* is low (0.47%) compared to *Pleurotus* spp., which is 90.8%. Despite the low production rate in Malaysia, the demand and the cultivation practice of this mushroom has increase worldwide due to its pleasant flavor and high protein content [9]. This mushroom belongs to family Pluteacea of Basidiomycetes [15] and is widely grown in tropical and subtropical regions of Southeast Asia such as Thailand, Philippines, China, and Malaysia [2]. It has significant pharmaceutical value, such as immunosuppressive proteins, anti-tumor polysaccharide, and immunomodulactory lectins [15].

Volvariella volvacea (*V. volvacea*) is an edible cultivated mushroom, commonly known as paddy straw mushroom or Chinese mushroom [16]. In cultivation of *V. volvacea*, besides the spawn and the condition (temperature, relative humidity, hydrogen potential) of the surroundings, the media used as the substrate for the mushroom plays a very important role in controlling the production of the mushroom. A proper pre-treatment, which includes composting, addition of supplemental nutrients, and pasteurization of mushroom substrate, is required to improve productivity [19]. According to Bao et al. (2013) [2], *V. volvacea* is a species of mushroom that grow naturally on paddy straw (the substrate). However, this mushroom can also be cultivated on other crop residues such as cotton waste, wheat straw, sugarcane industrial waste, and empty fruit bunch (EFB). Nowadays, Malaysia is becoming one of the biggest producers in terms of palm oil plantations and produces large volumes of palm oil products [4]. Therefore, its residues can be used as a good resource for other agriculture activities.

The continuously increasing production of large quantities of EFB each year is mainly why it can be used as a source of cultivation substrate for mushroom production [8]. This is also because they are available at low cost and the residues contain nutrients that can help in mushroom growth. In previous research, the cultivation of *V. volvacea* on the EFB showed higher production and biological efficiency compared to its production using paddy straw as the substrate [18]. However, in Malaysia, the cultivation of this mushroom on EFB is still at a nascent stage. Therefore, the researchers are still trying to find methods that can improve the quality of the substrate in order to produce high quality of the mushrooms.

The composting periods of the substrate can be one of the factors that affect its quality; This is because the EFB must be composted before it can be used as the substrate for the mushroom beds. Composting is a process that converts organic residues into stable substances which can be handled, stored, transported, and applied to the field without adversely affecting the environment [13]. The composting period is the period for which the media have been left to be composted. There is still no study that demonstrates the best EFB composting period to optimize the yield production. In this study, the relationship between composting period and production of *V. volvacea* is measured along with the weight of the *V. volvacea* produced. Therefore, the objectives of this study are to investigate the relationship between the composting period of EFB and the production of *V. volvacea* besides determining the optimal composting period of EFB in giving high production of *V. volvacea*. After completion of this study, the use of EFB compost and their best composting period can be applied and practically adopted by the growers in their cultivation practices.

2. Materials and methods

2.1. Preparation of substrate (EFB) and spawn collection

The study was conducted using only *V. volvacea*. The cultivation of the *V. volvacea* was done under Sapucaya nut trees at Vegetable Garden, Taman Pertanian Universiti (TPU), Universiti Putra Malaysia, Serdang Selangor. The EFB was collected from Jugra Oil Palm Sdn. Bhd, Banting, Selangor, Malaysia. The *V. volvacea* mushrooms spawn was obtained from Department of Agriculture at Padang Terap, Kedah. The spawn contains cotton waste as the substrate and the mycelium was fully covered the cotton waste substrate (Fig. 1).

Fig. 1: Spawn of *Volvariella volvacea*.

2.2. The compost preparation

According to the Department of Agriculture, Padang Terap, Kedah, Malaysia, EFB is currently composted for nine days to facilitate cultivation of *V. volvacea*. Therefore, there were three composting periods-six, nine, and twelve days-were used for the experiment in the present study. The EFB was gathered together and covered using canvas. The temperature was maintained in between 30 □C and 35 □C. The wate days interval. The media was prepared starting from twelve days, followed by nine days, and lastly, six days of composting period. This was done to make sure that spawning occurred on the same day.

2.3. Mushroom bed preparation

The beds for each treatment were prepared in triplicates. This means there were nine beds altogether. The beds' size was standardized as 1.0 m width x 1.5 m length. The EFB in each bed of substrate media was arranged in an overlapping manner. After that, the substrate for each bed was watered to reduce the oily surface of the EFB. Then, the substrates were feet-pressed to remove the air gap in the bed to create appropriate compactness for the mycelia growth. The beds were all arranged according to a Randomized Complete Block Design (RCBD).

2.4. Spawning

After the mushroom beds were prepared, the spawning was done by distributing the spawn of *V. volvacea* on the EFB. There was only one bag of spawn (300 g) for each beds of mushroom. After spawning had been done, the beds were covered properly with black polythene sheets and were left for seven days. On the eighth day, the polythene cover was removed for a while, and then, it was lifted into a dome shape to allow the mushrooms to grow (Fig. 2).

Fig. 2: The plastic was lifted into dome shapes on day 8 after spawning.

2.5. Maintenance of the bed

Black polythene sheet was used to cover the bed after spawning until fructification in order to maintain optimum temperature and humidity. The sheet also protects the beds from rain, wind, and external contaminants, and prevents accumulation of moisture in the form of water droplets. Watering at the side of the bed is done depending on the climatic conditions. According to Biswas and Layak, (2014) [3], the optimum temperature for mycelia growth was maintained at 26–30 °C using a Forehead Thermometer (model FT-100D), and at 34–37^{o}C for fructification, and the relative humidity was maintained between 70–90 % using a Hydrometer.

2.6. Harvesting

After 14 days from spawning, the first flush of *V. volvacea* was harvested. Harvesting was done by carefully separating the mushroom from its substrate (EFB). Then, the bed was covered with the sheet again and harvest was continued until there were no more mushrooms growing.

2.7. Parameters measured

In this study, the weight of *V. volvacea* was recorded using a weighing scale. The weight of the mushroom is the most suitable criteria to represent each treatment for comparison purpose.

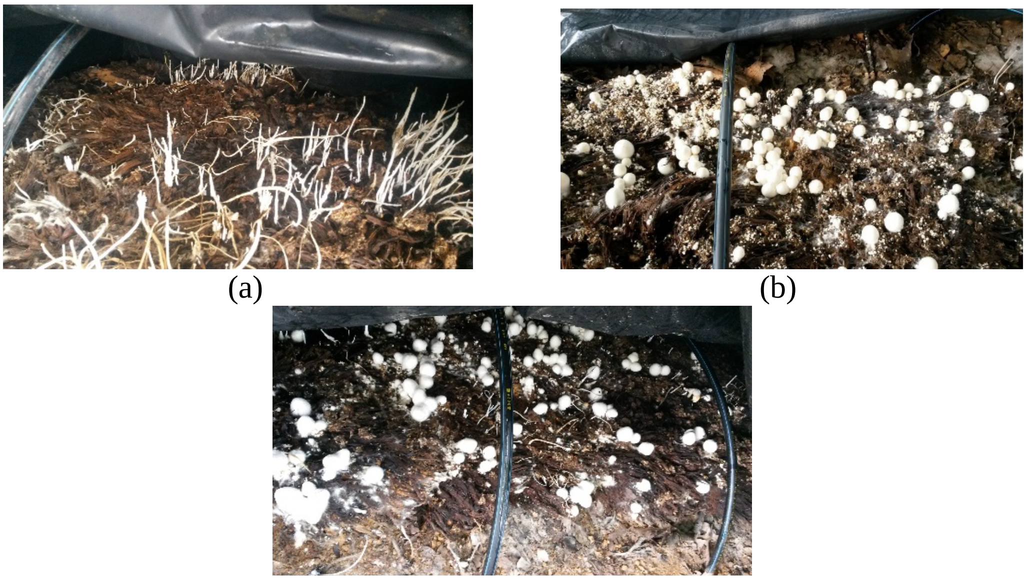
2.8. Statistical analysis

The raw data of each treatment that were recorded was processed using Microsoft Excel 2010 and the test for the data was performed using Statistical Analysis System (SAS) Version 9.4 software. Results of weight between treatments were analyzed using Analysis of Variance (ANOVA) and Duncan's Multiple Range Test was used for multiple mean comparison.

3. Results and discussion

Theoretically, composting is a biological process that depends on the microorganism population that converts the organic substance waste into stabilized and less complex compounds by breaking down the organic residue [20]. Since composting process is time-consuming, the grower will need to achieve the minimum period for composting in order to obtain high production of *V. volvacea* in a shorter time [17]. The composting period is very important as it is the period that determines the level of readiness of EFB to be used as the substrate for *V. volvacea* cultivation.

The treatments of the composting period for EFB were decided at six, nine, and twelve days. The weight of the mushroom is the most suitable parameter that represents the production of the *V. volvacea* for each treatment. Figure 3 showed the production of *V. volvacea* from each treated bed before harvest. On the first day of harvesting, a large number of *V. volvacea* at the button stage of growth appeared on the bed of nine days (Fig. 3 (b)) and twelve days of composting (Fig. 3 (c)). Conversely, the bed of six days composting did not produce any fruiting body of *V. volvacea* (Fig. 3 (a)). Data collected for each treatment were analyzed and summarized in Table 1.



(a) (b) (c)

Fig. 3: The condition of EFB's growing bed of *V. volvacaea* cultivation for (a) six days composting period, (b) nine days composting period and (c) twelve days composting period.

Table 1: The summary of data on number and weight of *V. volvacea* for each treatment after fourteen days of harvesting.

Composting Period (days)	Number of mushrooms	Weight of mushrooms (g)	Mean weight of mushrooms (g) ±S.E.
6	84	2630.00	876.67±412.53[a]
9	100	3450.00	1150.00±341.22[a]
12	132	4890.00	1630.00±151.33[a]

Note. S.E. is short form for standard error. It should be noted that, Mean ± S.E. followed by same letters are not significantly different to each other according to Duncan's ($p > 0.05$).

Based on the results from the analysis (Table 1), there was no significant difference between the mean weight of *V. volvacea* for each treatment (CP6=CP9=CP12). A study by Triyono et al. (2019) [19] on the composting period of EFB (two days, five days, and eight days) for production of the *V. volvacea* fruiting body also showed no significant difference between all treatments. However, from the study, the size reduction of EFB significantly decreases the yield of *V. volvacea* fruit body. Conversely, a study by Arifestiananda et al., (2015) [1] on the effect of composting period of paddy straw on production of *V. volvacea* found that the rice straw composted for five days produced significantly higher yield in term of total weight and number of fruits body, compared to composting periods of ten days and fifteen days. Besides that, Hernandez et al. (2003) [7] observed that the biological efficiency of *Pleurotus ostreatus* for mushroom bed with composted substrate (30% coffee pulp and 70% dry pangola grass) was significantly higher compared to non-composted substrate of the same mixture.

In this study, although there is no significant difference in terms of average weight of the fruiting body between all composting periods, EFB with six days composting showed a slower production of fruiting body compared to other treatments. Besides that, the six-days mushroom beds showed zero production on the first day of harvesting which might be because the EFB condition is not yet at the level of compost suitable for *V. volvacea* to grow due to the shorter period. However, at day six of harvesting, the production of the mushroom from the six-day beds shows similar results as the other two beds during first day of harvesting. Farid's (2011) [5] study on effect of composting of paddy straw on growth of *V. volvacea* showed that the composted paddy straw ensures a significantly faster harvesting stage compared to non-composted paddy straw.

In addition, shorter time of composting process does not ensure the organic substance complex has been completely broken into nutrients, such as Carbon and Nitrogen [11], which can be readily absorbed by the mycelium and other stages of the mushroom to grow. This is very important as the mycelium of the mushroom obtain nutrients supplied from the EFB to grow and mature into fruiting body of *V. volvacea.* According to Arifestiananda et al. (2015) [1], the composting periods of paddy straw affect the protein content in the fruit body of *V. volvacea*. The research produces the paddy straw composted with ten days produced fruit body with highest protein content, which is 41.94 mg/g, compared to five days composted paddy straw. This supports that the hypothesis that composting period could affect the amount of nutrient absorbed by the mushroom.

According to Meng et al. (2019) [12], temperature is one of the primary parameters used to monitor the composting process, because it changes in a manner related to the decomposition of organic matter and the growth of microorganisms. During the composting process, the microorganisms release heat and energy which can increase the temperature to achieve optimum temperature for *V. volvacea* to grow [10]. However, a shorter period causes the temperature to be quite lower for *V. volvacea* mycelium to grow (Table 2).

Table 2: The data of temperature during composting period at three days interval.

Day	Temp (□C) of E		
	6	9	12
0	-	-	26.8
3	-	27.7	29.8
6	27.3	29.7	31.6
9	30	31.6	33.1
12	31.1	33.0	33.7

Note. '-' means there is no data recorded as the composting have not been started yet.

Based on data as recorded in Table 2, the temperature of the EFB was different during the composting process. The temperature of EFB for all treatments increased with time and showed the highest temperature at day 12. According to Thiribhuvanamala et al. (2012) [18], the temperature should be maintained between 30 –35 □C. Therefore were at the suitable range of temperature to be used as the substrate for *V. volvacea*. However, the temperature of EFB that has been composted for six days is lower compared to EFB that have been composted for nine and twelve days, respectively. Therefore, this might be the reasons for slower production of *V. volvacea* compared to other treatments.

Moreover, the lower temperature triggered the other fungi species to grow (Fig. 3 (a)). According to Goyal et al. (2005) [6], generally during composting, fungi are involved actively in the decomposition of the cellulose, hemicelluloses, and lignin that are present in the organic matter, which is the EFB in this. Fungi possess enzymes that can break down the materials. The examples of the cellulolytic fungi that inoculate the organic matter to accelerate the composting process include *Aspergillus* and *Trichoderma* [17]. These fungi grow at low temperatures and high moisture conditions. Therefore, compared to the *V. volvacea*, the condition is more favorable for other fungi to compete with *V. volvacea*, thus lowering the production of *V. volvacea* in the six-day beds.

4. Conclusion

In conclusion, the cultivation of *V. volvacea* using agricultural residues such as EFB is a value-added process to viably convert this waste into useful materials. Based on the results obtained, the effects of EFB composting periods on the yield of *V. volvacea* statistically were not significantly different at $p < 0.05$ among the three treatments (six days, nine days and twelve days). This means that the composting period of EFB in the range of six days to twelve days are suitable for the production of *V. volvacea*. However, the weight of the mushroom noticeably increases with increasing composting periods. This study may become the benchmark for the mushroom growers in choosing their preferred composting periods for the cultivation substrate used.

Acknowledgements

The authors would like to thank Malaysian Higher Education (MOHE) for the FRGS grants (FRGS/1/2016/WAB01/UPM/02/15).

References

[1] S. Arifestiananda, and S. R. Setiyono, “The effect of medium composting time and dosage of chicken manure on the yields and protein content of paddy straw mushroom”, Agric. Science Bull, 10, 10, 2015, pp. 1-6.

[2] D. Bao, M. Gong, H. Zheng, M. Chen, L. Zhang, H. Wang, and Y. Zhou, “Sequencing and comparative analysis of the straw mushroom (*Volvariella volvacea*) genome”, PLoS One, 8, 3, 2013, pp. e58294.

[3] M. K. Biswas, and M. Layak, “Techniques for increasing the biological efficiency of paddy straw mushroom (*Volvariella volvacea*) in Eastern India”, J. Food Sci. Technol, 2, 4, 2014, pp. 52-57.

[4] S. H. Chang, “An overview of empty fruit bunch from oil palm as feedstock for bio-oil production”, J. Biomass Bioenergy, 62, 2014, pp. 174-181.

[5] A. Farid, " Effect of composting and kinds of carbohydrate sources on growth and results of straw mushroom", M.S. thesis, Faculty of Agriculture, University of Jember, Indonesia, 2011.

[6] S. Goyal, S. K. Dhull, and K. K. Kapoor, "Chemical and biological changes during composting of different organic wastes and assessment of compost maturity", J. Bioresour. Technol. 96, 14, 2005, pp. 1584-1591.

[7] D. Hernandez, J. E. Sanchez, and K. Yamasaki, " A simple procedure for preparing substrate for *Pleurotus ostreatus* cultivation", Bioresour. Technol, 90, 2, 2000, pp. 145-150.

[8] T. K. Hoe, M.R. Sarmidi, S. S. R. S. Alwee, and Z. A. Zakaria, "Recycling of oil palm empty fruit bunch as potential carrier for biofertilizer formulation", J. Teknol, 78, 2016, pp. 165-170.

[9] K. Kumud, S. Lily, B. Behera, and K. B. Mohapatra, "Improvement of biological efficiency of paddy straw on mushroom through substrate management", J. Environ. Eco. 32, 1, 2014, pp. 33-39.

[10] R. Kulcu, and O. Yaldiz, "The composting of agricultural wastes and the new parameter for the assessment of the process", J. Ecol. Eng, 69, 2014, pp. 220-225.

[11] S. Malayil, H. N. Chanakya, and R. Ashwath, R. "Biogas digester liquid—a nutrient supplement for mushroom cultivation", J. Environ. Nanotech. Monitor. Management, 6, 2016, pp. 24-31.

[12] X. Meng, B. Liu, H. Zhang, J. Wu, X. Yuan, and Z. Cui, "Co-composting of the biogas residues and spent mushroom substrate: Physicochemical properties and maturity assessment", Bioresour. Technol, 276, 2019, pp. 281-287.

[13] N. Mohammad, M. Z. Alam, and N. A. Kabashi, "Optimization of effective composting process of oil palm industrial waste by lignocellulolytic fungi", J. Mater. Cycles Waste Manage. 17, 1, 2015, pp. 91-98.

[14] M. Z. Rosmiza, W. P. Davies, R. A. CR, M. J. Jabil, and M. Mazdi, "Prospects for increasing commercial mushroom production in Malaysia: challenges and opportunities", Mediterranean J. Social Sci, 7(1S1), 2016, pp. 406.

[15] A. Roy, P. Prasad, and N. Gupta, N. "*Volvariella volvacea*: A macrofungus having nutritional and health potential", Asian J. Pharm. Technol, 4, 2, 2014, pp. 110-113.

[16] A. Shi, H. Hu, F. Zheng, L. Long, and S. Ding, "Biochemical characteristics of an alkaline pectate lyase PelA from *Volvariella volvacea*: roles of the highly conserved N-glycosylation site in its secretion and activity", J. Appl Microbiol. Biotechnol, 99, 8, 2015, pp. 3447-3458.

[17] S. Siddiquee, S. N. Shafawati, and L. Naher, "Effective composting of empty fruit bunches using potential *Trichoderma* strains", Biotechnol. Reports, 13, 2017, pp. 1-7.

[18] G. Thiribhuvanamala, S. Krishnamoorthy, K. Manoranjitham, V. Praksasm, and S. Krishnan, " Improved techniques to enhance the yield of paddy straw mushroom (*Volvariella volvacea*) for commercial cultivation", African J. Biotechnol, 11, 64, 2012, pp. 12740-12748.

[19] S. Triyono, A. Haryanto, M. Telaumbanua, J. Lumbanraja, and F. To, "Cultivation of straw mushroom (*Volvariella volvacea*) on oil palm empty fruit bunch growth medium", Int. J. Recycling Org. Waste in Agric, 8, 4, 2019, pp. 381-392.

[20] J. Zhang, G. Zeng, Y. Chen, M. Yu, Z. Yu, H. Li, ..., and H. Huang, H. "Effects of physico-chemical parameters on the bacterial and fungal communities during agricultural waste composting", J. Bioresour. Technol, 102, 3, 2011, pp. 2950-2956.