# Effect of Substrate Weight Variation and Spawn Method in Production of White Oyster Mushroom (Pleurotus Ostreatus)

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# Abstract

The aim of this research was to obtain a bag log size of white mushroom (Pleurotus ostreatus) capable of producing optimal fruit body with a relatively short time and high Biological Efficiency (BE) value. The study was conducted from February to October 2017. The design of the study used a Completely Randomized Design with two factors. The first factor is the mushroom spawn method (M) consisting of 2 (two) levels which include:  $m_1 = Direct$  Eksplan Planting (DEP); and  $m_2 = Pure$  Mycelium Culture (PMC). The second factor is the weight of the substrate (B) consisting of 4 (four) levels which include:  $b_1 = 1.0$  Kg;  $b_2 = 1.5$  Kg;  $b_3 = 2.0$  Kg;  $b_4 = 2.5$  Kg. The conclusion (1). The pattern of growth of white oyster mushroom mycelium is best to use the seeds of DEP method with weight of 1 Kg Substrate in 5 weeks long mycelium able to cover bag-log. (2). Production of white oyster mushroom is best using a combination of spawning method of DEP or PMC with a weight of 1.0 kg substrate indicated by Biological Efficiency value of 63.1%.

Keyword: Pleurotus ostreatus, spawn, substrate weight

## 1. Introduction

Asian countries produce more than 74.64% of world mushroom markets followed by Europe (19.63%) respectively in 2014 (FAO, 2015). In recent years, about 40% of total world mushroom are exported from China as the world's biggest producer of mushroom. However, 95% of the total China production is for domestic consumption (Zhang, et al., 2014).

One of the benefits of mushroom cultivation is their potential contribution to a more sustainable and environmentally friendly way of farming. Mushroom cultivation using an agricultural waste as a growing medium and the subsequent use of a spent substrate has high value for horticultural activity; organic fertilizer and potential utilize for animal feeding. The value of mushroom to diets, coupled with reported medicinal properties, can also provide valuable additional new small and medium-scale business options (Stanley and Odu. 2012; Rosmiza et al. 2016). Roughly about 300 mushrooms are edible, but only 30 have been domesticated and 10 are grown commercially globally. The best-known specialty mushroom and easiest to market is Oyster mushrooms and Shiitake (Barney, n.d). Different mushrooms lend themselves to different growing systems. The most important condition for mushrooms fruiting is for the growing environment to be carefully controlled with temperature, humidity, light and sometimes atmospheric gases (Tisdale et.al. 2006). Since mushrooms containabout 90% of water, it is also desirable to grow them under a relative humidity above 85-90% (Sugianto, at al., 2017).

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An existing shade house was modified for mushroom production and proved to be an adequate fruiting site. Nitrogen-fixing trees (C. equisetifolia, T. orientalis, and F. moluccana) supported greater yields (275.5, 272.4 and 268.8 g/bag, respectively), biological efficiency (70.1, 78.5, and 74.0%, respectively) (Tracy et al., 2006). Pleurotus genus is one of most extensively studied white-rot fungi due to its exceptional ligninolytic properties. It is an edible mushroom, and it also has several biological effects, as it contains important bioactive molecules. In basidiomycete fungi, lignocellulolytic enzymes are affected by many typical fermentation factors, such as medium composition, ratio of carbon to nitrogen, pH, temperature, air composition, etc. (Sugianto, et al. 2017).

The cultivation of the white oyster mushroom depends on factors such as: substrate planting composition, environmental condition, spawn quality, substrate weight, and Biological Efficiency value. Fungal cultivation substrate should be able to be transformed into a fruit body by inoculated fungi. High biological efficiency (BE) values indicate that from planting substrate was successfully converted by mushrooms into high fruit bodies. Spawn quality is determined by the method of preparation, because the resulting mycelium is related to the ability to degrade fibers in the substrate (Sunawan and Sugianto 2005). Hafifi's research (2013), indicating there is difference between the methods used, DEP method gives effect faster than PMC method. DEP method has better adaptability with the difference reaching the range of 1 to 2 days. Different if the calculation between generations of seeds, then for DEP method tends to be stable compared to PMC method.

Sumiati research results (2005), substrate weight of 1 to 2 kg per bag log are best used in Indonesian plantations because if there is contamination it wills easier handling. This is because the climates in the tropic with high moisture levels are more susceptible to pest and disease attacks. In subtropical countries where moisture is low pests and diseases are difficult to develop. The combination of spawns of DEP and PMC methods with different substrate weights is expected to increase the value of harvest production shown through Biological Efficiency values. The aim of this research is to get bag-log size from white oyster mushroom (Pleurotus ostreatus) which able to produce optimal fruit body with a relatively short time and height Biological Efficiency value.

## 2. Materials and Methods

The research was conducted at Faculty of Agriculture Integrated Laboratory and Central Laboratory of Islamic University of Malang, from February to October 2017. The research site is located at an altitude of 500 meters above sea level, with an average daily temperature of 27°C. Spawns used in this study using two methods namely DEP and PMC. The production of white oyster mushroom spawns by DEP method was made by explants taken from the stem and put into a bottle filled with sterilized corn kernels using autoclave at 121°C, pressure 15 Lbs for 15 minutes. Substrate that has been inoculated then incubated in dark space with room temperature 27-28°C until the mycelium meet the media and ready to be lowered in the next generation.

Seeds made by the method of PMC are done by taking part of the body of mushroom fruit and inserted into petridish that has been filled with media potatoes dextrose agar (PDA) then incubated at a temperature of 26-28°C, until the media is full with white mushroom mycelium. Derivative seed production was done by using a mixture of substrate which consisted of the sawmill 40%, bran 15%, 2% gypsum, CaCO<sub>3</sub> 1%, SP-36 0,5% and water 41,5%. The substrate mixture is inserted in a saucer bottle and sterilized in a 15 lb pressure autoclave, with a temperature of 121°C for 20 minutes. The bottles are cooled and inoculated with pre-made culture results. For 20 days after inoculation, the surface of the spawning bottle will be covered with white mycelium. Stages of making of planting substrate (bag-log) with mixture of sawdust of Albasia wood 47,0%; bran 10%; lime 0.5%; gypsum 1.5%; 0.5% corn flour; SP-36 0,5%; and 40% water. The mixing results are put into a 0.03 mm PP plastic bag. Bag-log weight is made with the size of 1 kg, 1.5 kg, 2 kg and 2.5 kg according to the treatment settings. Bag-log sterilized with temperature stemer 121°C, pressure 15 pound, for 3 hours.

The design used was a Factorial Complete Random Design with two factors. The first factor is Spawn Mushroom (B) consisting of 2 (two) levels which include:  $b_1 = DEP$ ; and  $b_2 = PMC$ . The second factor is the size (weight) of the substrate (U) consisting of 4 (four) levels which include:  $u_1 = 1.0$  kg;  $u_2 = 1.5$  kg;  $u_3 = 2.0$  kg;  $u_4 = 2.5$  kg. From these two factors, 8 (eight) treatment combinations were obtained and each treatment unit was repeated 6 (six) times. Observed variables included: growth pattern of mycelium, average number of hoods, total fruit body fresh weight (TFBFW) and Biological Efficiency (BE) were calculated using formula BE = TFBFW/Weight of substrate x 100% (Sugianto, 2007).

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#### 3. Results and Discussion

## 3.1 Mycelium Growth Pattern

The growth pattern of mycelium has the real interaction between the spawnings method and the substrate weight from the first week to the fifth week presented in Figure 1.

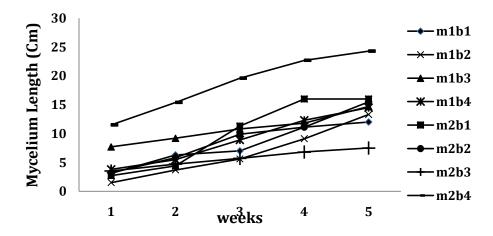


Figure 1. Mycelium Growth Pattern Average Fulfill The Media On Spawning Method Treatment And Different Substrate Weight in Five Weeks

Figure 1, showed the best pattern of weekly mycelium growth trends in combination of  $m_1b_1$  (PMC spawning method + substrate weight of 1.0 kg) resulted in the fastest mycelium growth with an average trend of 3.2 cm per week. The ability to prolong mycelium depends on the available nutrients. Nutrition is a stimulus for the formation of mycelium and fruit body. The process of fruiting body formation is indirectly influenced by the growth of mycelium, because the initial stage of the formation of the fruit body requires nitrogen-containing material (Sugianto 2005).

Oyster mushroom as a saprofit, using carbon source which is from organic materials to elaborated to be simple carbon compound then absorbed into mushroom mycelium. Water needs are using for chemical particle flow between the cell which helps the growth and development of mycelium to form fruit body and spores at once (Chang and Miles, 2004). The ability to produce oyster mushroom organic compound can growth at various materials which contain carbohydrates or other organic carbon compounds. The absorbable carbon source is soluble such as monosaccharides or compounds like sugar, organic acids, amino acids, and other organic compounds (Stanley and Odu, 2012). In addition to the carbon element as a metabolic process, nitrogen elements are needed as a composer of organic amino in proteins and enzymes.

# 3.2 Average Number of Hoods

The average number of fruit body hoods is presented in Figure 2. Treatment interaction of spawning method DEP + weight of substrate 2.5 kg  $(m_1b_4)$  has the highest average number of hoods 14.33 fruits which are not significantly different with method of spawning of PMC + weight of substrate 1 kg  $(m_2b_1)$  of 13.67 pieces. Method of spawning PMC + substrate weight of 1 kg  $(m_2b_2)$  is 13.00. Spawning method DEP + weight of 1 kg substrate  $(m_1b_1)$  of 13.00 pieces.

Spawning method DEP + weight substrate 2 kg  $(m_1b_3)$  of 13.00 fruit and method of spawning PMC + weight of substrate 2 kg  $(m_2b_3)$  equal to 12,67 pieces, significantly different with treatment combination of spawning method DEP + weight substrate 1.5 kg  $(m_1b_2)$  of 11.33 fruit and spawning method DEP + weight substrate 1 kg  $(m_2b_4)$  of 8.00 pieces.

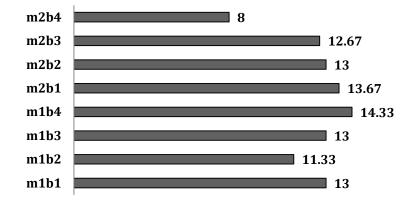
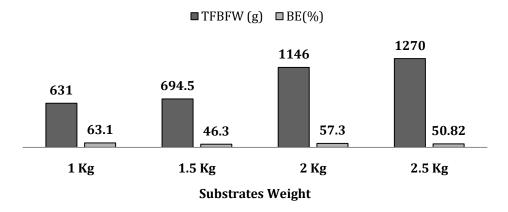
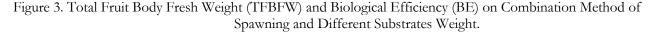


Figure 2. Average Number of Hoods Each Harvest (Pieces) Combination Treatment Spawning Methods and Different Substrates Weight.

# 3. Total Fruit Body Fresh Weight (TFBFW) and Biological Efficiency (BE)

Separate TFBFW and average BE were only affected by the weight of the substrate (Figure 3). The weight of 2.5 kg substrates  $(b_4)$  was not significantly different from the weight of 2 kg substrate  $(b_3)$  and was significantly different from the other treatments.





The highest value in treatment  $b_1$  (1 kg substrate weight) was 63.10% and the lowest value in treatment  $b_2$  (substrate weight 1.5 kg) was 46.30%. This phenomenon shows that the heavy substrate is not proportional to the average value. The larger the substrate size the larger the resulting hood diameter and the fresh weight measurement of the fruit body. The substrate is the main nutrient for the fungus. The nutrients can be used after the fungus excretes Extra cellular enzymes that can elaborated a simpler concoction (Suharnowo, 2012). Nutrition for white oyster mushrooms (Plerotus ostreatus) is needed to support growth and development. Nutrition is taken from the substrate either directly in the form of substances, ions and molecules such as hexose sugar (6-C). Indirectly, the fungus degrades automatically, or the polymer is decomposed into simple molecules or monomers. Elements that do not require white oyster mushrooms (Plerotus ostreatus) are absorbed in the form of compounds such as carbon, nitrogen, phosphorus, potassium, magnesium, calcium, sulfur, boron, molybdumine, cobalt and vitamin B complex (Sugianto, 2017).

# 4. Conclusions

Based on the results of data analysis from the research that has been done can be summarized as follows: Production of white oyster mushroom (Pleurotus ostreatus) best use combination of spawning method DEP or PMC with weight of 1.0 kg substrate shown by biological efficiency value 63, 1%.

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#### References

- Chang S.T. and Miles, P.G. (2004). Mushrooms: Cultivation, Nutritional Value, Medicinal Effect and Environmental Impact (Second Edition), 451 Pp. Boca Raton, Fl: CRC Press.
- Hafifi, S. (2013). Test the Effectiveness and Efficiency of Direct Eksplan Planting (DEP) Method as an innovation for White Oyster Mushroom (Pleurotus ostreatus) Spawning. J. Agronisma. 1(1): 5-10.
- Rosmiza MZ, Davies, WP, Rosniza Aznie CR, Jabil MJ, and Mazdi M. (2016). Prospects for Increasing Commercial Mushroom Production in Malaysia: Challenges and Opportunities. Mediterranean Journal of Social Sciences.MCSER Publishing, Rome-Italy. 7(1): 406-415
- Sugianto, A. 2005. Response of White Oyster Mushroom (Pleurotus ostreatus) to Variable Rice Straw Substrate C / N Ratio and Addition of AGS + Varied Nutrient Dosages. Science Research Media. 1(1): 34-41.
- Sugianto, A. (2007). White Oysther Mushroom Seed Testing Made With Cold Shock Methods The Eksplant And Mycellium Pure Culture . J. Agritek 15(6): 1512-1517
- Sugianto, A. 2017. Development of Wood Mushroom Technology as Alternative Food. Intimedia Publishing. Malang. Indonesia. ISBN. 978-602-1507-37-7.
- Sugianto, A., A.1 Solihah, and P. Hartono. (2017). Acceleration of Five Types Edibel Wood Mushroom Production through Varied Harvest Synchronization Temperature Settings. Journal of Agriculture and Environmental Sciences. 6(2):128-133.
- Suharnowo.( 2012). Growth of Mycelium and Fruit Body Production. Jurnal Lantera Bio.1(3): 12-17
- Sunawan dan A. Sugianto. (2005). Spawning Study of Two Types of Wood Mushrooms Lentinus edodes and Pleurotus ostreatus and their Tests Against Various Kinds of Nutrition. J. Agritek. 13(4): 715-725
- Sumiati.(2005). Repair of White Oyster Mushroom Pleurotus ostreatus Florida Strain with Modification of Main Raw Materials Substrate. J. Hort 16(2).
- Stanley HO, Odu NN. (2012). Cultivation Of Oyster Mushroom (Pleurotus Tuber-Regium) On Selected Organic Waste.International Journal of Advance Biological Research 2: 446-448
- Tisdale, T. E. Susan, C. And Miyasaka-Hemmes, D.E. (2006). Cultivation Of The Oyster Mushroom (Pleurotus ostreatus) On Wood Substrates In Hawai. World. J. Microbiol.Biotechnol. (22) 201-20
- Tracy E. TisdaleSusan C. MiyasakaEmail author. Don E. Hemmes. (2006). Cultivation of the oyster mushroom (Pleurotus ostreatus) on wood substrates in Hawaii.World Journal of Microbiology and Biotechnology. 22(3):201
- Zhang, Y., Geng, W., Shen, Y., Wang, Y., & Dai, Y. (2014). Edible mushroom cultivation for food security and rural development in China: bio-innovation, technological dissemination and marketing. Sustainability. 2(6): 2961-2973.