

GROWTH PERFORMANCE AND MORPHOLOGY OF FUNGAL FOAM DERIVED FROM EDIBLE MUSHROOM

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Abstract. Production of mushrooms has grown steadily in global market. This have gain interest from researchers to utilize mushroom for better use. *Pleurotus florida* is one of the well edible mushrooms cultivated in Malaysia. The abundance of mushroom production leads to a new alternative of fungal foam production which is derived from the fungal species and natural resources obtained from agricultural wastes. Since agricultural wastes contribute to environmental problems, hence utilizing them into beneficial products is a great idea. Sawdust was chosen as substrate to grow the mushroom for forming fungal foam. The purpose of this study is to evaluate the growth performance of *Pleurotus florida* with different spawn loading on sawdust. The produced fungal foam was analyzed on the morphological structure and chemical structure. Sawdust was mixed with 10 % w/w and 20 % w/w of spawn loading for forming fungal foam. Growth performance was observed where it takes 24 days for 20 % w/w of spawn loading while 10 % w/w spawn loading fully grown into foam after 32 days under room temperature and 20 % w/w of spawn shows higher specific growth rate at 0.2415 cm/day. Morphological testing on the fungal foam was determined using scanning electron microscope (SEM) and 20 % w/w of spawn loading was highly compact compared to 10 % w/w of spawn loading. Result proved that the high growth rate in 20 % w/w of spawn loading can be promoted for future use since it also has a better morphological structure that will give good mechanical properties.

Keywords: Fungal foam, growth, morphology, mycelium, sawdust

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Introduction

The need for more environmentally friendly materials has increased in recent years due to the severe environmental problems caused by plastic waste pollution. Alternatively, biodegradable polymers derived from natural resources such as lignocellulosic materials are an excellent solution to this problem. They can be easily renewed, are relatively cheap, and can be found in abundant supply. The lignocellulosic substance usually consists of 35-50 % cellulose, 20-35 % hemicellulose, and 10-25 % lignin [1] that are needed for growing fungal.

Fungal is a eukaryotic type of species kingdom that makes up multiple cells. Mycelium is one of the parts in fungi or mushroom is known as the roots of species. It is made up of a fibrous filament known as hyphae [2]. Mycelium is the primary part of fungi material that are used to produce the fungal foam. The mycelia and the hyphae are vital fungus elements, more often known as the mushroom. The two phrases relate to the building components of fungal. Few types of mushrooms have been widely cultivated in Malaysia such as *Agaricus*, *Auricularia*, and *Pleurotus* [3] and this study used *Pleurotus florida* to grow in sawdust.

Meanwhile, the high demand for fungi in the industry cause induces the usage of agar as the medium for the fungi to grow. However, the cost production of media preparation is expensive [4] since the substrate used for cultivating spawn which is minimal agar is high cost. Therefore, the previous research shows that a variety of substrates are used in the cultivation of fungal foam [5]. In tropical climates, sawdust is utilized as a growth medium, depending on the areas and accessible trees. The most widely used sawdust is made from rubber trees, followed by *Acacia auriculiformis*, *Mangifera indica*, and *Tamarindus indica* [6]. A combination of sawdust with fungi will produce similar to common commercial foam in the industry.

The production of fungal foam consists of few processes. Primarily choosing the types of substrates was very important to assist fungal growth within a period of cultivation. Fungal foam was created by combining fungal mycelium with a lignocellulosic substrate. However, the characteristics of the produced material are dependent on the fungus and substrate used, the growth circumstances, and the procedure of cultivation and growth environment [5]. Besides that, the fungal foam may be created in two ways which are by utilizing mycelium's ability to interconnect other substances inside its network to form a bulk material. On the other hand, imprecise formula during the mixing of lignocellulose substrates creates a poor type of growth profile for the fungal foam. This is important to maintain the final product of fungal foam for future applications such as packaging materials or absorbent since the properties are comparable to the current polymeric foams in the industry.

Hence, the main focus of this research is to investigate the uses of different inoculum loading on sawdust in fungal foam production of *Pleurotus Florida*. The study will cover from growth profile and morphology of fungal foam. The Fourier transform infrared spectrophotometer (FTIR) was used to identify the various functional groups present in the sample.

Materials and Methods

Samples Preparation

The *P. florida* was obtained from mushroom grower near Padang Besar, Perlis. Rice bran is also provided by the same merchant. Next, the substrate which is sawdust was earned by a local supplier near Kangar, Perlis. Medium for cultivating spawn was prepared with ratio 100:10:1 of substrate, rice bran and calcium carbonate respectively. Sawdust used was 1000 g with 10 g of rice bran and 1 g of calcium carbonate and all the medium was mixed [7].

The distilled water (1200 ml -1300 ml) was added into the medium until it becomes clump. The sample then placed into 3/4 of cup with 6 cm height. The upper part of the cups was exposed with small holes filled by cotton ball to allow the oxygen to enter the cup for the help in cultivation process. Then, samples were placed into an autoclave with 121 °C for 30 minutes to ensure the mixing medium to be free from any contamination.

Fungal Foam Production

Sawdust with 6 cm height of the cups was mixed with 10 % w/w and 20 % w/w of the spawn loading. The medium inoculated with the *P. florida* was incubated in the dark cabinet at 25 °C for the spawn running process. The cabinet's relative humidity was controlled in between 75 - 85 % with no presence of light [8]. The fungal foam was cultivated in a dark cabinet and the growth has been observed in every 4 days. This was to determine which sample infuse to become the foam first. Fungal started to grow on the top of the substrate until it reached the bottom of the cup and fully covered the substrate. Till this point, it is considered that fungal foam was formed. After fungal foams were fully formed, the samples were dried in an oven at 70 °C for 24 hours. The primary function of this method was to stop the production of fungi and remove any impurities on fungal foam. The samples were collected after have been fully dried and left at room temperature for 20 minutes before keeping in the storage seal with plastic to make sure there was no water vapor left on the sample which lead to contamination.

Fungal Foam Analysis

Fungal foams produced were analyzed in terms of growth performance, morphological structure and chemical structure. The growth of fungal foams was monitored every four days by ticking the level of fungal foam growth height in cm on the cup and the result obtained were recorded to track the progress. The growth rate of fungal was determined by linear regression model by referring to Mitchell et al. [9]. The slope of the graph from the linear line is the growth rate of fungal. As for the morphology structure of fungal foam, it was determined by the scanning electronic microscope (SEM). To prevent electrostatic charge during the inspection with an accelerating 1015 kV voltage, the fungal foams were sputter-coated with a thin coating of platinum [10]. Moreover, FTIR was used to analyze the chemical structure contained in the fungal foam. Both fungal foam samples; 10 % w/w and 20 % w/w spawn loading were tested and evaluated to compare the characteristic and structure.

Results and Discussion

Growth Performance of Fungal Foam

Figure 1 and Table 1 showed that fungal foam that used 20 % w/w of spawn loading exhibited a higher growth rate of at 0.2415 cm/day compared to spawn loading of 10 % w/w at 0.2373 cm/day. At 20 % w/w spawn loading, fungal foam took a shorter time to fully grow which was only 24 days, and 10 % w/w loading took 32 days. This was due to the low activity of enzyme that is secreted by *P. florida* to absorb nutrients to stimulate mycelium growth [5]. The result revealed that spawn loading impacted the production of fungal foam, where a higher spawn loading increased the growth rates of fungal foam. This result in an agreement with Nashiruddin et al. [5], that grew of *Pleurotus ostreatus* (white oyster mushroom) on rice husk with different loading and found that higher spawn loading resulted in faster mycelium growth. It was believed that high mycelium growth was due to high laccase activity to extract nutrients to initiate mycelium growth. This was also agreed by Idowu et al. [11] that using 3 % of spawn loading and had taken a longer time to grow mycelium as compared to 13 % of spawn loading. On the other hand, Sabu et al. [12] also reported similar findings as well. However, there was a limit on the increasing of the spawn loading. When the limit was exceeded by more than 60 %, the enzyme production may be reduced due to the rapid depletion of nutrients, leading to a decrease in metabolic activity [5]. Besides, it is important for the substrate to have enough moisture content to allow the hyphae cells to grow longer and the mycelium to spread throughout the substrate. The optimum moisture content for the *P. florida* growth is ranged between 55 % -70 % [5]. Moreover, increases in period of cultivation were cause decrease in compressive strength. This was due to the large amount of organic substrate breaking down [13].

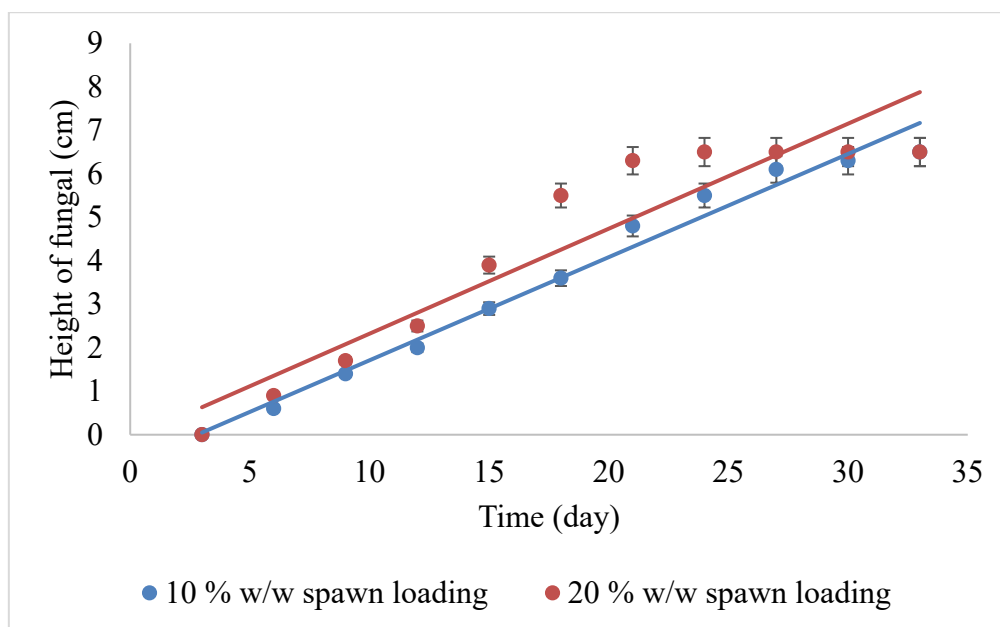


Figure 1: Growth of fungal on sawdust at different spawn loading of *P. florida*

Table 1: Growth rate of fungal in different spawn loading

Spawn loading (% w/w)	Growth rate (cm/day)
10	0.2373
20	0.2415

Morphological Properties

Using a scanning electron microscope (SEM), the morphology of fungal foam was analyzed under 1000 $\times$ magnification power as shown in Figure 2. Surface features of the fungal foam were characterized by the colonization of mycelium branches in the varied spawn loading of 10 % w/w and 20 % w/w. The entangled tube - like structure were indicated the appearance of hyphae. The combination of hyphae produces branches representing the mycelium. The more branches formed in the fungal foam, the more formation of hyphae in the sawdust which shows that the 20 % of spawn loading grows effectively in sawdust. Figure 2(b) shows fungal foam of 20 % w/w spawn loading that have more branches of hyphae compared to 10 % w/w spawn loading in Figure 2(a). The mycelium in 20 % w/w of spawn loading was highly compact compared to the 10 % w/w of spawn loading. The enzyme produced during the fungal foam formation indirectly affects the number of hyphae attach to the substrate. Therefore, the substrate rich in polysaccharides alter the morphology of the fungal foam [2].

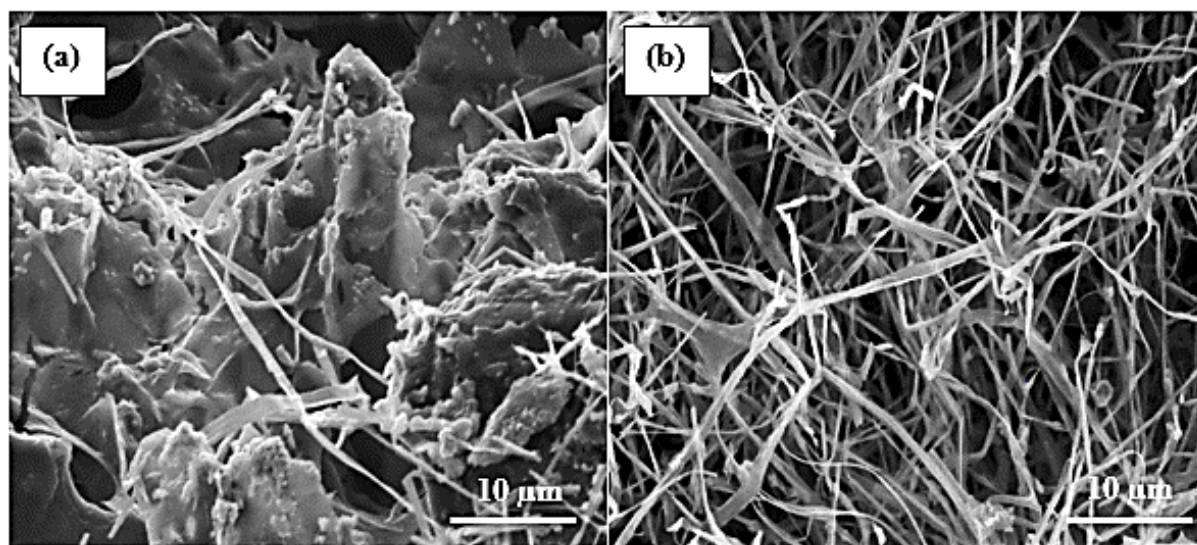


Figure 2: Growth SEM fracture of micrograph of (a) Fungal foam of 10 % spawn loading and (b) Fungal foam with 20 % spawn loading

Chemical Structure of Fungal Foam

The FTIR spectrum of *P. florida* fungal foams from 20 % w/w of spawn loading was shown in Figure 3. The result shows six regions peak of absorbent. The absorption band appeared around 3342 cm^{-1} with strong and broad wavelength represent the O-H group. In the other hand, the broad band in the range of 2440- 3460 cm^{-1} attribute from the water present in the substrate. Therefore, the band at 2923.2 cm^{-1} shows medium wavelength corresponding to the C-H stretching. The band at 1651.2 cm^{-1} shows the -C=H stretching attribute to the lignin

aromatic group. Then, 1320.6 cm^{-1} shows the represent of medium O-H bending. The large peak at 1041.09 cm^{-1} shows the CO-O-CO stretching. Lastly, the peak at 779.09 shows the C-H bending of the mode of vibration aromatic component. This may have indicated the aromatic skeletal vibration [14].

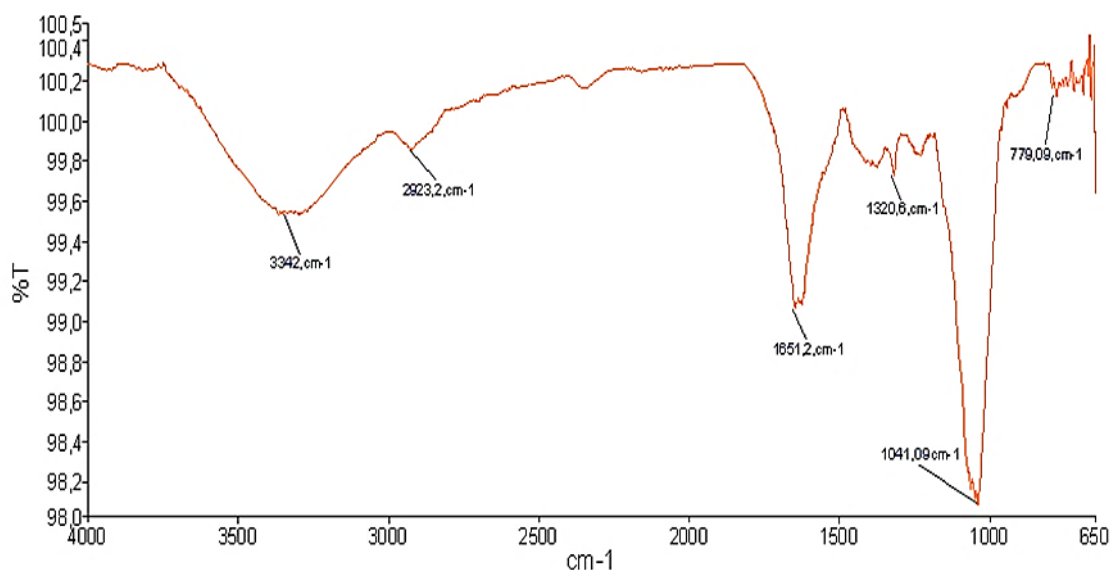


Figure 3: The wavelength of *P. florida* sample in sawdust using FTIR

Conclusions

In conclusion, the *P. florida* mushroom is one of the edible mushrooms that can quickly grow in any medium. The cultivation process is easy and consists of high nutritional values. By using different spawn loading, the growth of fungal foam has been observed and evaluated referring to the linear regression model. According to this, higher spawn loading shows the fastest growth rate at 2.415 cm/day within 24 days of cultivation of mycelium to become foam. Morphology of both fungal foams from 10 % w/w and 20 % w/w of spawn loading from scanning electron microscope (SEM) showed a massive difference between the both samples were indicating the numbers of branches mycelium foam indicates the characteristic of fungal foam. The mass production of *P. florida* and accessible growth of mushroom brought a great potential to replace the plastic industry because it may not biodegrade for thousands of years, and the process may take even longer.

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Author Contributions

All authors contributed toward data analysis, drafting and critically revising the paper and agree to be accountable for all aspects of the work.

Disclosure of Conflict of Interest

The authors have no disclosures to declare

Compliance with Ethical Standards

The work is compliant with ethical standards

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