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The Effect of Media Composition on The Change of Bag Log Waste Composition and Production of Oyster Mushroom

Sarono Sarono [1,✉], Yana Sukarya [2], Sri Astuti [3]

[1] Department of Agricultural Technology, Politeknik Negeri, Lampung, Bandar Lampung, INDONESIA.

[2] Master of Food Security, Politeknik Negeri Lampung, Bandar Lampung, INDONESIA.

[3] Department of Economics and Business, Politeknik Negeri Lampung, Bandar Lampung, INDONESIA.

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Corresponding Author:
✉sarono@polinela.ac.id

ABSTRACT

OPEFB has low biodegradability so its utilization is very limited. Oyster mushrooms are mushrooms that are capable of producing enzymes capable of degrading cellulose, hemicellulose, and lignin contained in) OPEFB. The research objective was to determine the effect of the composition of the growing media on the production of oyster mushrooms and changes in the composition of the resulting bag log waste. The study was carried out with the combination treatment of rubber wood and OPEFB, with levels: (a) 100% rubber wood powder and 0% OPEFB, (b) 75% rubber wood powder and 25% OPEFB, (c) 50% rubber wood powder and OPEFB 50%, (d) 25% rubber wood powder and 75% OPEFB, (e) 0% rubber wood powder and 100% OPEFB. Observation of raw material composition, mycelia growth, wet weight of the fungus, biological efficiency, changes in lignin content, cellulose, and hemicellulose before and after being used as a growing medium for oyster mushrooms. The results showed that the fastest mycelial growth occurred in the use of 100% OPEFB media. The highest oyster mushroom production occurred in the media 100% rubber sawdust. The higher the wet weight of the oyster mushrooms produced, the less the weight of the bag log waste produced. The oyster mushroom fermentation process for 60 days was able to reduce the content of 3.39% cellulose, 11.01% hemicellulose, and 1.98% lignin.

1. INTRODUCTION

The process of processing fresh palm oil fruit bunches produces by-products in the form of liquid waste, solid waste and gas waste. Solid waste which is equivalent to the amount of CPO produced and until now has not been utilized optimally is oil palm empty fruit bunches (OPEFB). According to Saelor *et al*., (2017) the largest solid waste is OPEFB of around 23% of the processed fresh fruit bunch (FFB), while according to Montoya *et al*., (2020) the amount of OPEFB is estimated at around 16.0% to 23.0. Research results in Indonesia show that the average number of OPEFB is around 21% (Yanti & Lestari, 2020).

So far, the OPEFB has been used only as a natural fertilizer by spreading it in gardens as mulch to cover the soil. Apart from producing methane gas emissions which is a greenhouse gas (GHG), the process of forming natural fertilizer also takes a long time, namely 60-90 days (Nurliyana *et al*., 2015). Methane gas emission from the decomposition of

OPEFB are a significant GHG with a strength of 20-30 times of CO2, and this is one of the reasons why palm oil products are claimed as not environmentally friendly (Amir & Manan, 2017).

According to Nurliana *et al*., (2015), the composition of empty oil palm fruit bunches is 95.64 ± 0.33% organic material; total carbon 41.97 ±1.42%; total nitrogen 0.664 ± 0.005%; lignin 20.34 ± 0.36%; cellulose 58.42 ± 0.01%; hemicellulose 21.29 ± 2.86%. Sarono *et al*., (2020) stated that OPEFB that just exited the factory has a cellulose content of 37.5%; hemicellulose 22.44%; lignin 23.03%; water 5.08%; and oil 5.63%. From this composition, it can be seen that OPEFB actually has a high organic component, but also has a high lignocellulose content. Lignocellulose is a component that is difficult to degrade naturally.

The research results of Mamimin *et al*., (2021), shows that EFB has low biodegradability so its use is very limited. Low biodegradability is caused by high lignocellulose content. According to Mamimin *et al*. (2021), the contents of hemicellulose, cellulose and lignin in empty oil palm fruit bunches are 22.0 ± 0.480; 45.5 ± 0.850; 24.5 ± 0.51. Research on the properties of lignocellulose has been carried out by Soh *et al.,* (2021), the results show that by using Van Krevelen Diagram Analysis the OPEFB components can undergo dehydration and decarboxylation reactions at a temperature of 240°C for 3h. This reaction implies the occurrence of partial decomposition of hemicellulose and cellulose at very high temperatures. The research results of Tukanghan *et al*., (2021) shows that the use of a consortium of microorganisms, especially the clostridium group, is very effective in accelerating the degradation of EFB. The consortium of microorganisms, especially the clostridium group, is able to produce the enzymes xylanase, lipase, endo-glucanase, and exo-glucanase which in their activities are capable of degrading EFB with an efficiency level of 57.5%.

The process of degrading lignocellulosic waste by microbial consortia has been widely carried out, apart from being environmentally friendly, it also produces beneficial mushrooms. Straw mushrooms and oyster mushrooms are food products produced from the lignocellulose degradation process. The results of observations in the community around the palm oil factory of the Bekri Business Unit of PTPN VII (Central Lampung) show that there is excellent innovation from the community (grassroots innovation), namely taking wild mushrooms that grow from piles of OPEFB as food. These mushrooms utilize food from OPEFB, including decomposing lignin (Krishnan *et al*., 2016).

So far, food mushroom cultivation has mostly used hardwood powder as raw materials such as rubber wood, sengon wood, and acacia wood (Wang & Zhao, 2023). The use of growing media and the inoculated mushroom species will produce different quality mushrooms, especially their protein content. The integration of oyster mushroom cultivation as a bio-pretreatment function in OPEFB to increase biodegradability needs further research. Therefore, the aim of this research is to determine the effect of the composition of the growing media on changes in the composition of bag log waste and the resulting oyster mushroom production.

2. Research Materials and Methods

The raw materials used are OPEFB obtained from the Bekri Business Unit of PTPN 7 in Central Lampung, rubber wood powder obtained from the straw mushroom business unit in Simbaringin Natar Village, South Lampung. The research was structured in a randomized complete block design with 3 replications. The research treatment was a combination of the portion of rubber wood sawdust and OPEFB, with the following levels:

(a) K100:T0 = rubber wood sawdust 100% and OPEFB 0%
(b) K75:T25 = Rubber wood sawdust 75% and OPEFB 25%
(c) K50:T50 = Rubber wood sawdust 50% and OPEFB 50%
(d) K25:T75 = Rubber wood sawdust 25% and OPEFB 75%
(e) K0:T100 = Rubber wood sawdust 0% and OPEFB 100%

The rubber wood and OPEFB were dried to a moisture content of around 12%. Other planting media that were added in fixed amounts were 10% of rice bran, 3% lime, and around 60% water. The transparent plastic with a size 15x30cm was used to make bag log. The weight of each bag log is made uniform, namely 1,200 g. The white oyster mushroom isolate (*Pleurotus ostreatus*) used was F2 seeds grown in sterilized corn media.

The number of bag logs in each replication was 6 bag logs of oyster mushrooms. Harvesting of oyster mushrooms was carried out at the age of 40 days after inoculation with the harvest criteria that the mushroom caps have bloomed. Harvesting was conducted by pulling out all the mushroom clumps. The research steps can be seen in Figure 1.

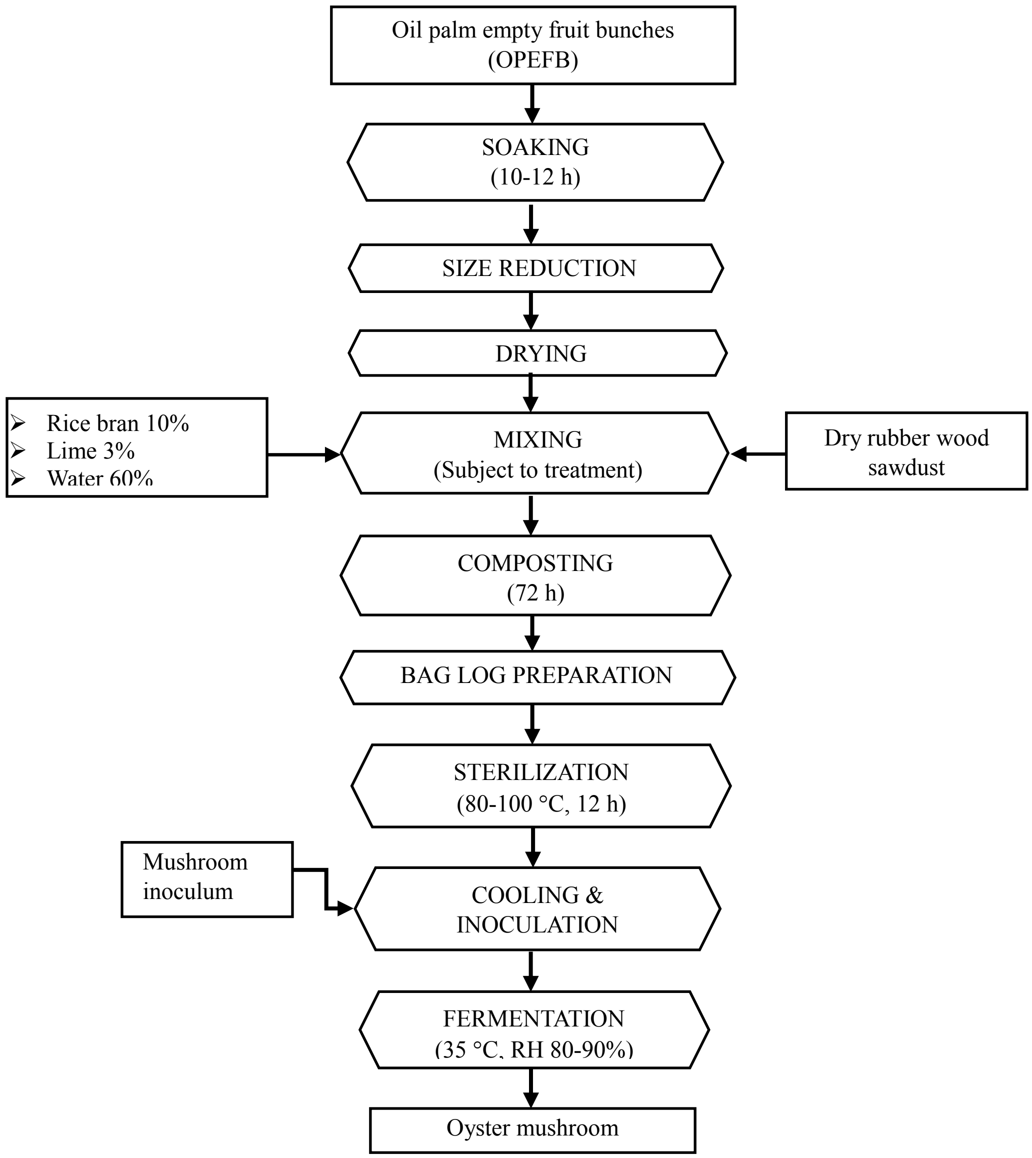

Figure 1. Stages of research implementation

Observations were made on the composition of raw materials, mycelial growth, wet weight of mushrooms, biological efficiency, changes in lignin, cellulose and hemicellulose content before and after being used as a growing medium for oyster mushrooms. Methods for analyzing the composition of raw materials are lignin, cellulose and hemicellulose, water and fat content (Chesson, 1981), mycelial growth (Kurniati *et al*., 2019), wet weight of mushrooms (Kurniati *et al*., 2019), and biological efficiency (Mamimin *et al*., 2021). The data was displayed in the form of graphs and tables.

3. RESULTS AND DISCUSSION

3.1. Raw Material Analysis

The physical appearance of rubber wood sawdust and OPEFB can be seen in Figure 2. Physically, rubber wood sawdust has a finer particle size, while OPEFB is very coarse in size and darker in color. OPEFB is physically in the form of fibers, so more effort is needed to smooth it. The composition of the OPEFB as compared with other studies is presented in Table 1, while the composition of the rubber wood sawdust can be seen in Table 2.

Figure 2. Physical appearance of rubber wood sawdust (a) and OPEFB fiber (b)

Table 1. Analysis results of OPEFB and its comparison from various sources

Characteristic	This work	Sarono *et al.* (2020)	Chan *et al.* (2021)	Saelor *et al.* (2017)
Cellulose (%)	40.52	37.50	23.70 – 65.00	43.30
Hemicelloluse (%)	22.75	22.44	20.58 – 33.52	26.20
Lignin (%)	21.79	23.03	14.10 – 30.45	30.50
Moisture (%)	7.21	5.08	2.40 – 14.28	ND
Fat (%)	4.59	5.63	ND	ND

Note: ND = Not determined

Table 2. Chemical properties of rubber wood sawdust from different sources

Characteristic	This work	Boerhendhy *et al.* (2010)	Safitri (2003)
Cellulose (%)	41.78	–	43.98
α-Cellulose (%)	–	–	37.71
Holocellulose (%)	–	67.38	70.06
Hemicellulose (%)	13.37	14.69	–
Lignin (%)	24.11	20.78	26.39
Pentose (%)	–	17.54	–
Moisture (%)	6.77	--	–
Ash (%)	1.06	0.779	–

From the table it can be seen that the three largest important components in OPEFB and rubber wood sawdust are cellulose, hemicellulose and lignin. In Table 1 and Table 2 it can also be seen that the cellulose, hemicellulose and lignin components of OPEFB between the research results compared to various previous research results are not the same. The composition of cellulose, hemicellulose, and lignin in the OPEFB is influenced by plant age, FFB maturity level, plant variety, and location of oil palm fruit planting (Chen *et al*., 2022). The use or processing of TKKS must pay attention to the nature of these three components, because they are the main components in TKKS.

Cellulose is a plant constituent substance found in cell structures. The cellulose content in plant cell walls is always higher than lignin and hemicellulose in plant dry weight (Chen *et al.*, 2022). Cellulose is a glucose polymer with ß-1,4 glucoside bonds in a straight chain. The chemical structure of cellulose can be seen in Figure 3. The crystalline structure of cellulose makes it difficult for cellulose to be degraded either by acids or bases. For enzymatic degradation, lignocellulose must be delignified first to facilitate the work of the cellulase enzyme in degrading cellulose (Mosier *et al*., 2005).

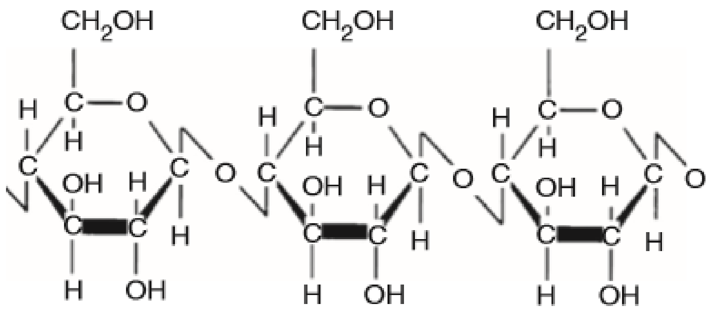

Figure 3. Chemical structure of cellulose (Chen *et al*., 2015)

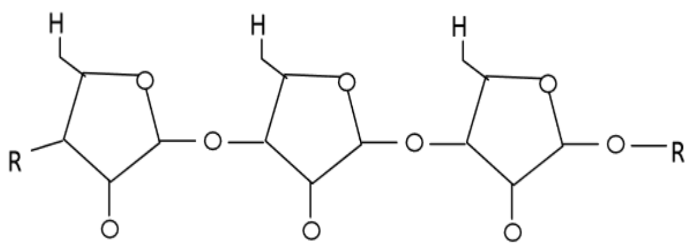

Figure 4. Chemical structure of the hemicellulose unit (Chen *et al*., 2022)

Table 3. Chemical characteristic of substrate for different treatments

Characteristic	Treatment				
	K100:T0	K75:T25	K50:T50	K25:T75	K0:T100
Cellulose (%)	41,78	41,46	41,15	40,84	40,52
Hemicellulose (%)	13,37	15,72	18,06	20,40	22,75
Lignin (%)	24,11	23,53	22,95	22,37	21,79
Moisture (%)	6,77	6,88	6,99	7,10	7,21
Ash (%)	1,06	1,94	2,83	3,71	4,59

Hemicellulose is a heterogeneous group of polysaccharides with a lower molecular weight than cellulose. The amount of hemicellulose is usually between 15 and 30 percent of the dry weight of the lignocellulosic material. Hemicellulose is relatively easier to hydrolyze with acids into monomers containing glucose, mannose, galactose, xylose and arabinose. Hemicellulose also cross-links with lignin to form a complex network and provides a strong structure (Suparjo, 2010). Hemicellulose consists of D-glucose, D-galactose, D-mannose, D-xylose, and L-arabinose units which are formed together in various combinations and glycosidic bonds (Xia *et al*., 2021). The formula for building hemicellulose can be seen in Figure 4. The composition of the mixture between OPEFB and rubber wood powder according to the treatment can be seen in Table 3.

Lignin is a component of plants which, together with cellulose and other fiber materials, forms the structural parts and cells of plants. In plant stems, lignin functions as a binding material for other components, so that a tree can stand upright. Lignin is formed from aromatic groups which are linked to each other by aliphatic chains, which consist of 2-3 carbons. The chemical structure of the lignin unit can be seen in Figure 5. During pyrolysis process, lignin is the source of aromatic chemical compounds such as phenol, especially cresol (Chen *et al*., 2022). The lignin structure is very resistant to chemical and enzymatic degradation. Lignin, cellulose and hemicellulose are always present in plant cell walls, so they are often classified as carbohydrates, even though lignin is not a carbohydrate. This is evidenced by the presence of a higher proportion of carbon in lignin (Xia *et al*., 2021). The lignin content differs between one plant and another, for example in pine straw 22.65%; wheat straw 20.40%; and hemp fiber 14.88% (Xia *et al*., 2021).

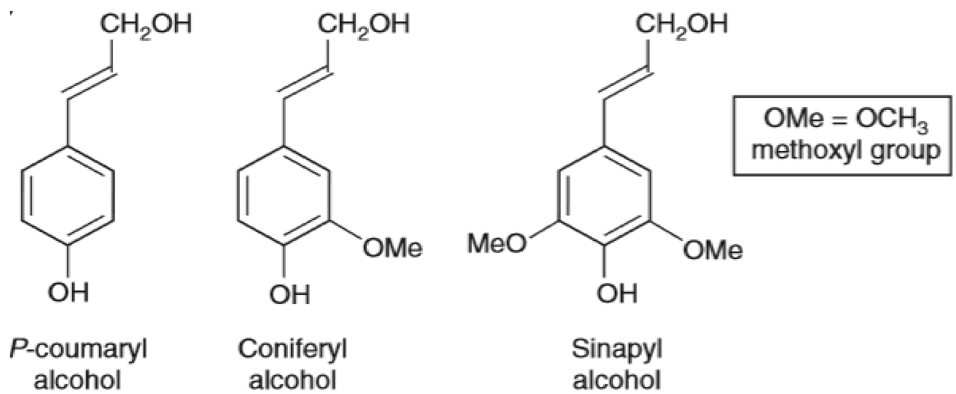

Figure 5. Chemical structure lignin unit (Chen *et al*., 2022)

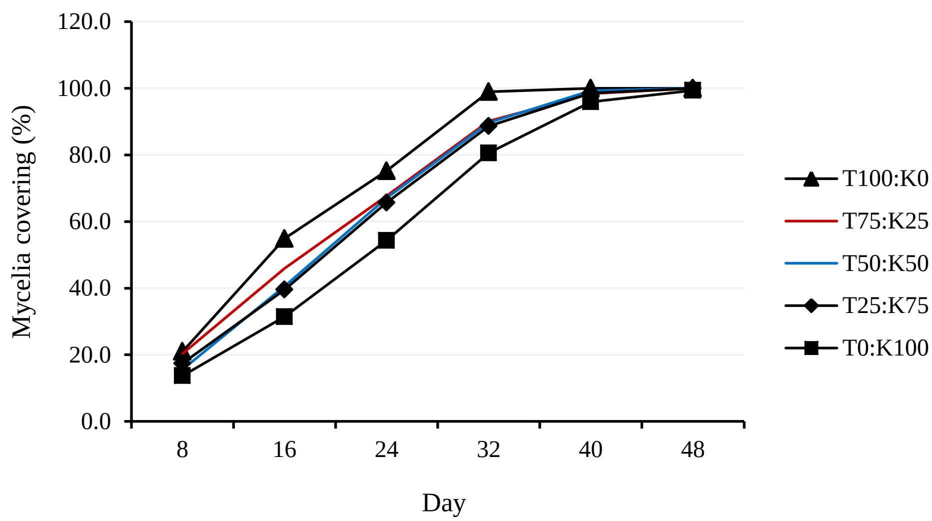

Figure 6. Growth of mycelia covering for different planting media

3.2. Mycelia Growth

The results of the research showed that the fastest media coverage by mycelia occurred in the treatment of 100% OPEFB powder and 0% rubber wood powder. On media made from EFB, 100% full media coverage occurred on day 32 after inoculation. Figure 6 shows the development of mycelial growth in each treatment. The average coverage of the media by mycelia occurred on day 40. These results indicate that OPEB are very suitable for use as a growing medium for oyster mushrooms. The speed of mycelial growth on bag log can reflect the suitability of the media used. The higher the suitability of the media, the faster the mycelial growth.

Kurniati *et al*., (2019) reported that media composition greatly influences the growth of oyster mushrooms using planting media a combination of rice husks and wood dust. The results showed that the number of fruit bodies and wet weight of white oyster mushrooms was better in the composition of 15% husk media +85% wood powder, namely 38.70 fruit bodies and a wet weight of 493.98 g.

3.3. Oyster Mushroom Wet Weight

The wet weight of white oyster mushrooms is the accumulation of four harvests during the production period. In general, the weight of the mushrooms produced tends to decrease from harvest to next harvests. The decrease in mushroom yields is due to the decreasing amount of nutrients in the bag log or because good quality mushroom seeds that growing at the beginning.

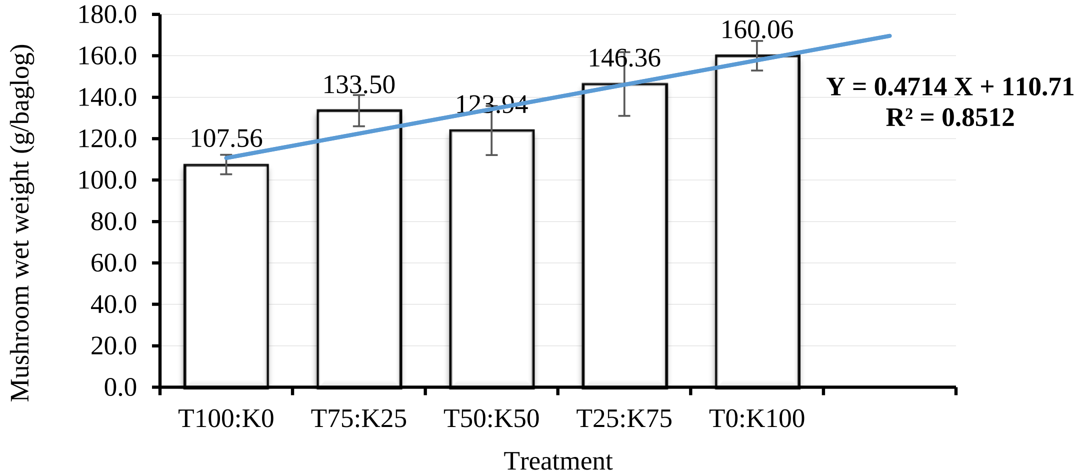

Figure 7. Effect of media composition on the mushroom wet weight

Figure 7 shows that mushroom yield (Y) increase with rubberwood sawdust (X) portion. The relation can be presented linearly as Y = 0.4714X + 110.71 with R^2 = 0.8512. The highest wet weight of oyster mushrooms was obtained from cultivating oyster mushrooms using 100% rubberwood sawdust growing media, which reached 160.06 g/bag log (48.8% higher than that of using 100% OPEFB). Mamimin *et al*., (2021) stated that OPEFB has the potential as a food source, namely as a substrate for producing straw mushrooms. The research results show that 1 ton of OPEFB can produce 47.3 kg of mushrooms and 82% or 820 kg of used OPEFB. The research results of Kurniati *et al*., (2019) shows that the maximum substitution of husk for wood sawdust is 15%. The addition of excess wood sawdust will reduce the wet weight of the oyster mushrooms produced.

3.4. Biological Efficiency

Biological efficiency (BE) is the percentage of mushroom in using the substrate to form fruiting bodies. A high BE indicates the mushroom's good ability to use its production media. The BE value is directly correlated with the wet weight of the straw mushrooms produced. Therefore, the highest BE value was produced by the 100% rubber sawdust treatment, namely 13.34% and the lowest BE value was produced by the 100% OPEFB treatment, namely 8.96% (Figure 8). The relation of BE and rubberwood sawdust portion (X) can be presented linearly as BE = 0.0393X + 9.226 with R^2 = 0.8507. The trend of BE is quite similar to that of mushroom yield. This further strengthens that the main part of the substrate that decomposes and produces mushrooms is rubberwood sawdust.

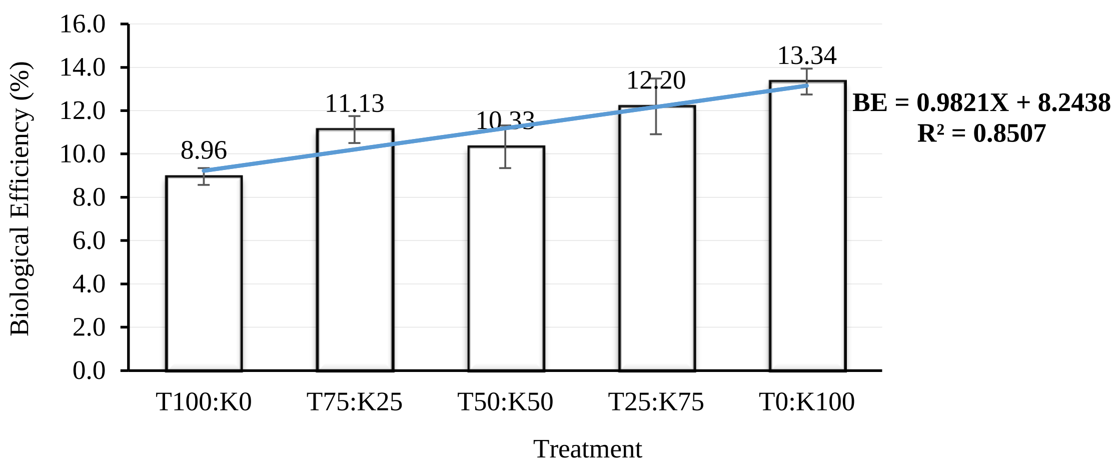

Figure 8. Effect of growing media on the biological efficiency of white oyster mushrooms

The results of this study show that the BE value is still low as compared to the other studies. Hultberg *et al*., (2023) have conducted research on the use of high concentration household waste fertilizer produced anaerobically for cultivating oyster mushrooms using sawdust as a growing medium. The results showed that the addition of 0.5 liters of fertilizer per kg of sawdust was able to produce biological efficiency (79.5 ± 5.4%). Yang *et al*., (2022) conducted research on the composting process and proper preparation of sawdust in an effort to increase the biological efficiency value of oyster mushroom production. The results showed that a composting duration of 4–5 days at temperatures above 58 °C was suitable for mushroom cultivation based on a BE range of 69.76–73.41% and a contamination level of 0%. The total carbon content (TC) continues to decrease during composting, while the total nitrogen content (TN) continues to increase. The final total nitrogen and C/N were 1.89% and 28/1, respectively, which were within the optimal range of nutritional requirements for oyster mushroom cultivation. According to Yang *et al*., (2022), composting bacteria are more diverse than fungal species. *Caldibacillus*, *Thermobispora*, *Thermopolyspora*, *Thermobacillus* and *Ureibacillus* are the dominant bacterial genera during the thermophilic stage.

3.5. Substrate Waste Weight

The results of the research showed that the weight of bag log waste produced was inversely proportional to the fresh weight of oyster mushroom products and decreased with the proportion of rubberwood sawdust (Figure 9). In the mushroom growing media treatment, 100% rubber wood powder produced the highest fresh weight of mushrooms and produced the lowest medium waste weight. The high fresh weight of mushrooms will also consume high levels of nutrients from the bag log, so that the weight of the bag log waste decreases. The relation of rubberwood sawdust portion (X) and the bag log waste (W) can be presented linearly as W = -0.172X + 1033.6 with reasonable high R^2 = 0.797.

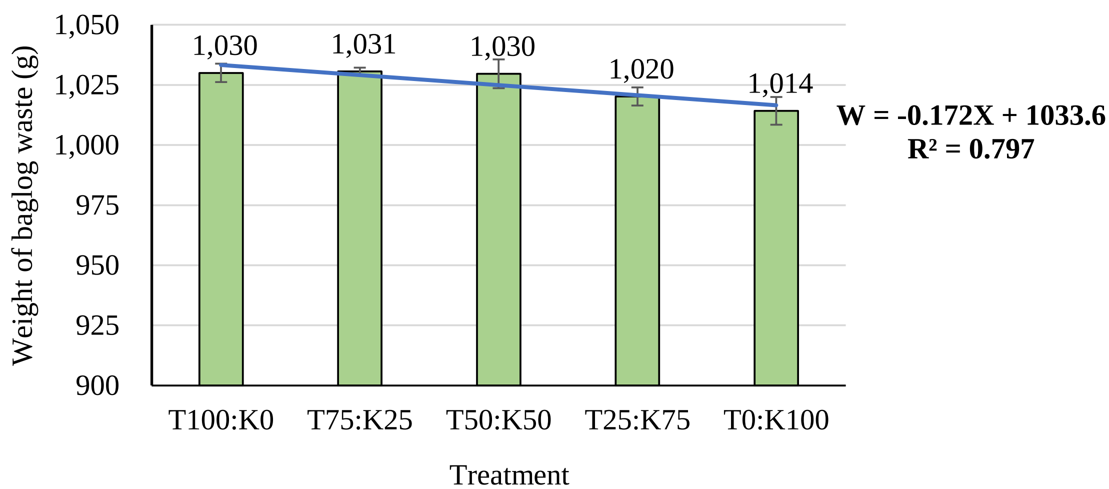

Figure 9. Effect of treatment on the weight of substrate (bag log) waste

3.6. Changes in OPEFB Composition

The results of the research showed that cultivating oyster mushrooms using mixed medium (rubber sawdust and EFB) was able to reduce the cellulose, hemicellulose and lignin content. The amount of reduction was 3.39% cellulose, 11.01% hemicellulose, and 1.98% lignin (Figure 10). If we pay attention to the very small decrease in cellulose, hemicellulose and lignin content, this shows that the cellulose, hemicellulose and lignin components are components that are very difficult to decompose. Research on the properties of lignocellulose has been carried out by Soh *et al*., (2021). The results of his research showed that by using Van Krevelen Diagram Analysis the OPEFB components could undergo dehydration and decarboxylation reactions at a temperature of 240°C for 3 hours. This indings imply that the partial decomposition of hemicellulose and cellulose occurs at very high temperatures and very long times.

Mamimin *et al*., (2021) stated that EFB has low biodegradability, which limits its commercial use. The use of EFB as a cultivation medium for straw mushrooms (*Volvariella volvacea*) can increase biodegradability so that reduce the content of lignin, cellulose and hemicellulose. The results showed that there was a decrease in OPEFB cellulose, hemicellulose and lignin of 3.3%, 21.3% and 17.6% respectively. Öztürk & Atila (2021) conducted research on the relationship between the level of degradation of lignocellulosic waste grown by white rot fungus (*Hypsizygus ulmarius*) for 90 days. The results showed that there was a much lower reduction in lignin levels compared to hemicellulose and cellulose. This study also revealed that the key factor that dominates the success of *Hypsizygus ulmarius* mushroom cultivation is a substrate that has a moderate N content, low lignin and high cellulose content.

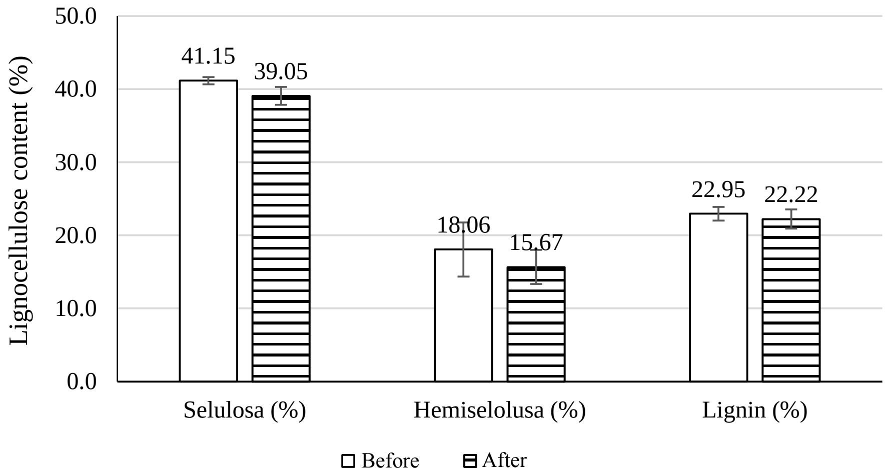

Figure 10. Changes in lignocellulose content of the substrate after used as a media for growing oyster mushrooms

Furthermore, Wang & Zhao (2023), stated that the process of using OPEFB as a medium for growing mushrooms is an environmentally friendly and economical process. It is said to be environmentally friendly because the process is carried out biologically and the by-product is compost. It is said to be economical, because the products produced have very promising market potential.

4. CONCLUSIONS

The fermentation process in making oyster mushrooms for 60 days was able to reduce the cellulose content to 3.39%, hemicellulose 11.01%, and lignin 1.98%. The fastest mycelial growth occurred on 100% EFB media, but the highest oyster mushroom production occurred on 100% rubber wood dust media. The higher the wet weight of oyster mushrooms produced, the lower the weight of bag log waste produced.

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