

Journal Bionature

p-ISSN 1411-4720 e-ISSN 2654-5160 Vol. 24. No. 1. April 2023, p. 180-187 http://ojs.unm.ac.id/bionature

The Effect of the Addition of Mealworm Frass (*Tenebrio molitor*) and Molasses on the Increasing of the Proximate Value of White Oyster Mushroom (*Pleurotus ostreatus*)

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Received: 08- 03 - 2023 Published: 30 - 04 - 2023

Abstract

This study aims to determine the effect of the addition of mealworm frass and molasses on the proximate value of white oyster mushrooms. This type of research was an experiment using a completely randomized design (CRD) consisting of four treatments with three replications each. Test parameters include water content, ash content, crude protein, crude fat and crude fiber. Proximate data on water content, ash content, crude protein, crude fat and crude fiber were analyzed using analysis of variance (ANOVA) and Tukey's follow-up test with a 95% confidence level. The results showed that treatment with frass and frass molasses had an effect on water content, ash content, crude protein, crude fat, and a decrease in fiber content. Therefore, it can be concluded that the addition of mealworm frass and molasses affects the proximate value of the resulting oyster mushrooms.

Keywords: White Oyster Mushroom, Flour Caterpillar Frass, Molasses, Proximate.

INTRODUCTION

White oyster mushroom is a type of wood mushroom that grows in weathered tree. This mushroom grows in subtropical, temperate, and tropical areas, with suitable environments. The content of white oyster mushrooms, including protein, phosphorus, fat, iron, riboflavin, and lovastatin (cholesterol-lowering), which is beneficial to our bodies. Synytsya (2002) reported that white oyster mushrooms contain a total dietary fiber content of 38.9-64.8%, soluble fiber content of 2.0-4.9% and insoluble fiber content of 31.8-61.4%.

Oyster mushrooms have a delicious taste and a high nutritional value. It is more than two times higher than the protein content in asparagus, cabbage, and potatoes. It is four times that of tomatoes and carrots and six times that of oranges. In addition, it contains 35-58 mg of vitamin C and 4.7-4.9 mg of vitamin B per 100 g of dry weight. Other ingredients in oyster mushrooms include mineral salts, iron (Fe), phosphorus (P), potassium (K), sodium (Na), and calcium (Ca) (Alex, 2011).

As mushroom production is not comparable to mushroom consumption in Indonesia, efforts are needed to increase mushroom cultivation. Mushroom production is largely determined by the quality of the growth medium. Suitable planting media consist of sawdust, lime, carbon, nitrogen, and molasses. Sawdust, which is used as a planting medium, contains a large amount of cellulose, which is required in large quantities by the fungus. Carbon was used as the main energy source, and nitrogen was used to build the mycelium. Lime maintains the

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acidity of media and mineral sources. Molasses is a nutritional supplement for white oyster mushrooms (Cahyana et al., 2005).

Mealworm (*Tenebrio molitor*) are insect larvae that are very easy to breed for housewives, both as their main livelihood and as a sideline. Mealworms are a commodity that is used as food for birds, fish, and reptiles. Besides that, it is also used as a raw material for cosmetics. During their lifetime, mealworms undergo several cycles, including eggs, larvae, cocoons (pupae), and ladybug/insect compatriots (Astuti et al., 2017).

Mealworm waste (frass) is leftover food and dirt used to raise mealworms. In general, mealworm shell waste contains 9.52% protein, 3.3% Mg minerals, 2.88% K minerals, 12.8% chitin, and other components, such as dissolved substances, 13.43% fat, and 14% crude fiber (Aziz et al., 2013). The shell is the waste in the cultivation of mealworms and will still be rich in chitin content. Furthermore, it can be used as an additive in the manufacture of white oyster mushroom media. Mealworm waste contains 11.20% lignin (Akhir, 2012).

Molasses is a waste product of sugar factory which can no longer crystallize. It contains K, Ca, and Cl (Armini et al., 2010), which function in the growth of white oyster mushrooms, as well as sugar, as a source of energy for cell metabolism which stimulates mycelium growth. Molasses also contains nitrogen elements ranging from 2-6% which function to build mycelia. The selection of additional media for molasses at different doses is expected to increase the production of white oyster mushrooms (fulfillment of mycelia, number of mushroom fruiting bodies, and fruit weight of white oyster mushrooms).

Proximate analysis is the analysis of major components of food and other agricultural products. Quantitative analysis was conducted on the content of substances such as water, ash, lipids, proteins, and carbohydrates. Qualitative analysis to determine the nutritional content of foodstuffs is generally carried out using proximate analysis. A proximate analysis is performed to determine the main chemical components of the food. The proximate analysis carried out in this study included moisture content, ash content, crude protein, crude fat, and crude fiber. The study aimed to determine the approximate value of the addition of mealworms frass and molasses, because there is still limited information regarding the role of frass in mealworms on the proximate value of white oyster mushrooms. Therefore, it is necessary to investigate the effect of adding mealworm frass and molasses on the proximate value of white oyster mushrooms.

RESEARCH METHODS

Time and place of implementation

This study was conducted on CV Surya Muda Mandiri oyster mushroom cultivation. Proximate analysis test was performed at the Feed Laboratory for Quality Testing of Animal Husbandry Products at the Animal Husbandry and Animal Health Services Office of the Province of South Sulawesi. This research was conducted from September 2021 to December 2021.

Growth media

Growth medium with four treatments and three replications were used to obtain 12 treatment combinations. Each replicate consisted of 50 bag logs; thus, the total treatment consisted of 600 bag logs. The basic media used additional nutrients in the form of frass, sugar, flour, and molasses at different percentages: K = Control (planting medium: sawdust 95%, bran 2%, dolomite 1%), F = Frass (planting medium + Frass 1%), M = Molasses (Planting medium + Molasses 0.05%), FM = Frass Molasses (Growing medium + Frass 1% + Molasses 0.05%). Furthermore, the ingredients of the mushroom-based media were mixed and sufficient water was added to each treatment to reach the total moisture content. The mixed material was placed into a mushroom bag log plastic weighing 1.5 kg/plastic with a total weight of 1 kg/plastic

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planting medium, and then compacted using a log bag press. After the media was compacted, a ring was attached to the plastic neck such that it resembled a bottle cap and then closed. The growth medium was sterilized using a steamer at 100–150°C for 3 h.

Inoculation and incubation

Inoculation was carried out by opening the bag log cover and bringing the end of the bag log closer to the bunsen burners then the F2 mushroom seeds (Muslimin et al., 2021) were inserted into it through the middle of the Parallon ring in the media. Incubation of the oyster mushroom bag log was carried out until all white (mycelium-grown) media (bag log) filled the bag log evenly and was then transferred to the mushroom house which was installed with the temperature and humidity controlled (Desnanjaya & Sugiartawan, 2022). The observed parameters were the growth of oyster mushrooms. Incubation ends after 5-6 weeks which is indicated by the presence of mycelia, which appear uniformly white, covering the entire surface of the growing medium. After growing the fruiting body, white oyster mushrooms were harvested by cutting the limbs of the fruiting body. A proximate analysis test was conducted, which included the following:

a) Water Content Test

The water content test was obtained using the Gravimetric method (AOAC, 2005) where the determination of the water content was based on the following formula:

Moisture content (%) = $(CD)/(BA) \times 100\%$

Information:

A: Empty cup weight (g)

B: Weight of cup + initial sample (g)

C: Weight of cup + dry sample (g)

b) Ash Content Test

Ash content was determined using the oven method (AOAC, 2005). Determination of ash content is calculated based on the following formula:

Ash content (%) = $(CD)/(BA) \times 100\%$

Information:

A: Empty cup weight (g)

B: Weight of cup + initial sample (g)

C: Weight of cup + dry sample (g)

c) Crude Protein Test

Protein levels were determined using the kjeldahl method (AOAC, 2005).

Determination of protein content is calculated based on the following formula:

Protein (%) = $((TB) \times N \times 1.4 \times F)/W \times 100\%$

Information:

Q: Volume for sample titration (mL)

B: Volume for blank titration (mL)

N: HCL

W: Sample weight (g)

F: Protein correction factor

d) Crude Fat Test

Fat content was determined using the Sokhlet method (AOAC, 2005). Determination of fat content is calculated based on the following formula:

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Total fat (%) =
$$(CA)/B \times 100\%$$

Keteragan:

A: Weight of empty round fat squash (g)

B: Sample weight (g)

C: weight of the extracted boiling flask in the oven (g)

e) Crude Fiber Test

Crude fiber content was carried out with reference to the method (AOAC, 2005). The formula for obtaining crude fiber is as follows:

Crude fiber (%) =
$$(W1 - W2 - B)/(W) \times 100\%$$

Information:

B: Weight of paper after oven (g)

W: Sample weight (g)

W1: Weight of sample + paper + after oven (g) W2: Sample weight + paper + after tenure (g)

RESULTS AND DISCUSSION

Results

Based on the research results, the average proximate content of white oyster mushrooms from various treatment media is listed in Table 1.

Table 1 Average proximate levels of white oyster mushrooms

Treatment	Water (%) ±SD	Ash (%) ±SD	Crude Protein (%) ±SD	Crude Fat (%) ±SD	Crude Fiber (%) ±SD
K	$91.85c \pm 0.01$	5.786bc± 0.334	22.723b± 0.488	0.813a± 0.075	21.903b± 2,141
F	$92.033d \pm 0.005$	$6.216c \pm 0.102$	23.433b± 0.542	1.743d±0.015	$15.2a \pm 0.4084$
M	$91.73b \pm 0.01$	5.123a± 0.136	18.616a± 0.427	1.44c±0.01	13.81a± 1.005
FM	90.81a ±0.01	5.583ab±0.112	23.273b±0.921	1.213b±0.144	13,593a±0.607

Notes: superscript accompanied by the same letter in the column does not show a significant difference

Information:

K = Control

F = Frass

M = Molasses

FM = Frass Molasses

Based on the results of proximate analysis data on the treatment of water, ash, crude protein, crude fat, and crude fiber content, the planting medium had a significant effect on each treatment. This shows significant results for each treatment with the growing media. The test was continued with the Tukey test, which showed that various treatments on each planting medium yielded significantly different results, including moisture, ash, crude protein, crude fat, and crude fiber contents.

In accordance with the results shown in Table 1, the highest average water content was found in treatment F, which was significantly different from all treatments, whereas the lowest water content was found in the FM treatment, which was significantly different from all treatments. The highest average ash content was found in treatment F, which was significantly different from all other treatments, and the lowest ash content was found in treatment M, which was significantly different from all other treatments. The highest mean

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crude protein content was in treatment F, and the average was the lowest in treatment M, both of which were significantly different from all treatments. The highest average crude fat content was found in treatment F and the lowest in treatment K, both of which were significantly different from the other treatments. The highest average crude fiber content was found in the K treatment and the lowest in the FM treatment.

Discussion

Water content

The water content in the growth medium greatly affects the growth of the mycelium and the development of fruiting bodies. Oyster mushrooms require approximately 90% water content (Sutarja, 2010). Optimal water content can stimulate rapid mycelial growth (Mufarrihah, 2009). With optimal water content, it can meet its water needs through the substrate as a growth medium for white oyster mushrooms, so that hyphae cells in white oyster mushrooms quickly absorb existing nutrients (Suryaningrum, 2012). The water regulatory system in fungi was found to increase (absorption increases) in mature primordia, which form fruiting bodies until the final stages of fruiting body development. Apart from water being used to fill the volume of cell growth, some are used up because of evaporation or exudation (Herman & Bleichrodt, 2021).

Treatment F has a high-water content, presumably because it contains a high-water content (Hapsari et al., 2018), which provides a high percentage of water content and does not comply with the optimal nutritional standards for white oyster mushrooms. Moisture content that is too low can interfere with the rate of growth and development of the mycelium; conversely, if it is too high, it will cause the mycelium to rot and die (Zamroji, 2020).

Ash content

The results of the Tukey test showed that the addition of mealworm frass and molasses to the growth medium resulted in significantly different ash content. Treatment F (6, 216%) had the highest ash content and was also the optimal standard value for the nutritional content of existing white oyster mushrooms (Sumarsih, 2015). Treatment F gave the highest yield because mealworm frass contains Mg, K, Ca, Na, S, and C minerals (Senthilkumar et al., 2016). Frass mealworms contribute a lot of minerals so that they can supply more minerals for the needs of the growth and development of white oyster mushrooms.

These minerals are absorbed during the mycelium growth process and then transported to mushroom fruiting bodies during the formation process (Chang & Miles, 1989). According to Sutarja (2010), the addition of nutrients in the form of frass waste causes an increase in mineral content. This is thought to cause the fulfillment of the ash content in white oyster mushrooms.

The difference in the ash content of the white oyster mushroom may be caused by the conditions in which it grows or the planting medium used. This is in accordance with Nafitri's research (2013), who stated that the difference in higher ash content was thought to be due to differences in the types of growing media where it was grown, as well as differences in the concentration of inorganic materials between one type of growing medium and another, hence the nutrient absorption by plants is also different.

Crude protein content

Protein is one of the macronutrient components which plays an important role in the formation of biomolecules because it make up half of the cell. Thus, proteins determine the size and structure of cells, which are the main components of enzymes that act as biocatalysts in various metabolic reactions in the body (Nuryanti et al., 2012).

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The F treatment (23.433%) and FM treatment (23.273%) produced the highest protein content because these two additional ingredients (frass and molasses) increased the concentration of protein that could be absorbed by the fungus. The additional material for mealworm frass in the planting medium contained a crude protein content of 19.32% (Hairani, 2006). Meanwhile, molasses contributed only a small amount of crude protein, which was approximately 3.1% (Trisnadewi et al., 2017). It can also be seen from the results that the weight of the mushrooms in the F and FM treatments increased compared to the weight of the K mushrooms (not published). This is because the F and FM media obtain more nutrients from molasses and frass. Mushrooms require a source of nutrients, such as nitrogen, phosphorus, sulfur, carbon, and several other elements. According to (Apriani, 2007), with increasing protein levels, it will later be degraded by fungal hyphae cells through digestion. This digestion runs extracellularly outside fungal body using protease enzymes, which break the protein down into micro-molecules in the form of amino acids. After digestion, these simple molecules were immediately absorbed by hyphae. The cells were then circulated using a transport system.

Proteins are also used as a part of enzyme formation process, a component of cell membrane formation, and a source of nitrogen and carbon that is used in many processes for the formation of new cell materials (Suryani & Cahyanto, 2022). Increasing protein levels help to meet the nutritional needs and standards of white oyster mushrooms, as outlined in (Sumarsih, 2015).

Crude fat content

Treatment F resulted in a high crude fat content compared to treatment K. The total fat content in widely cultivated mushrooms generally varies between 1.26-2.66% of the dry weight (Sumarsih, 2015). The crude fat content is relatively small for the needs of mushrooms, but its presence is essential because it is used in growth and other processes that are important in their life (Mshandete, 2012). The need for fat during the growth and development of mushrooms is achieved by expanding the mycelium to penetrate the substrate and absorbing nutrients by the white oyster mushroom hyphae (Angelia, 2016).

Crude fiber content

The results of the Tukey test showed that the addition of mealworm frass and molasses to the growth medium resulted in a significantly different crude fiber content. The control treatment had the highest fiber content because sawdust is a medium rich in fiber. Replacing the media with molasses, which is low in fiber (0.6%) (Trisnadewi et al., 2017), will certainly reduce the fiber content of the fungus. The addition of mealworm frass also had lower yields than the K treatment because, according to (Shifriyah et al., 2012), the crude fiber content in mealworm frass was 13.5%, so the fiber content in oyster mushrooms, even in the FM treatment, was lower.

Closing Statment

Conducting a proximate test on molasses (M), frass (F), and frass molasses (FM) media at different concentrations aims to determine the correct concentration for growing media/baglogs of white oyster mushroom (Pleurotus ostreatus). Conduct further tests by analyzing the content of each treatment in baglog to obtain better validation of growth factors and productivity.

CONCLUSION

The additional of mealworm frass alone enhance the proximate quality and suitable for growing white oyster mushroom.

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ACKNOWLEDGEMENT

This study was supported by the ministry of education, Culture, Research, and Technology, Indonesia 2021 year.

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