Utilization of *Pleurotus ostreatus* and *Lentinus squarrosulus* in The Manufacture of Mycelium-Based Biocomposites Using Sugarcane Bagasse and Cornstalk Media

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Abstract

Biocomposite is a composite consisting of a polymer matrix material and natural fiber reinforcement. Biocomposite technology, especially natural fibers, is currently in demand. The reasons are environmentally friendly, availability of many raw materials, low production costs, biodegradable, and recyclable. Natural fibers are used in the form of agricultural waste such as sugarcane bagasse and corn stalks while the mycelium of the fungus *Pleurotus ostreatus* and *Lentinus squarrosulus* are used as natural adhesives. The objectives of this research were to determine the effect of the type of white-rot fungus and the composition of the lignocellulosic material of agricultural waste that affects the quality of the resulting biocomposite board and to obtain the optimal type of white-rot fungus and the optimal composition of lignocellulosic material from agricultural waste to manufacture mycelium-based biocomposite board. The research method used a completely randomized design with ten treatments with three replications. The treatments used 2 types of mushrooms (*P. ostreatus* and *L. squarrosulus*) with 2 types of Lignocellulosic materials (Sugarcane bagasse and Cornstalk) and each lignocellulosic material has 4 types of compositions (100%, 75%, 50%, and 25%). Based on analytical test, the results showed that the treatment of the type of fungus and the lignocellulosic material used affects the quality of the resulting biocomposite board. *L. squarrosulus* in 100% sugarcane bagasse media is the optimal type of white-rot fungus and the optimal composition of lignocellulosic material to manufacture mycelium-based biocomposite board.

Key Words: *biocomposite, cornstalk, Lentinus squarrosulus, Pleurotus ostreatus, sugarcane bagasse*

INTRODUCTION

Biocomposite is a combination of two or more different materials into a microscopic unit form, consisting of a polymer matrix material and natural fiber reinforcement (Rusmiyatno, 2007; Davallo *et al.*, 2010). Natural fiber composites help to boost benefits such as being ecologically friendly materials throughout production, processing, and waste disposal with annual renewability (Balaji *et al.*, 2014).

Fungal mycelium acts as a natural adhesive for lignocellulosic fibers in the manufacture of biocomposite boards. Fungal mycelium will consume mostly organic matter or substrate, which can form tiny filaments. The better the ability of fungi to degrade lignin and utilize the cellulose content for mycelium growth, the stronger the fiber bonds, resulting in a better biocomposite board. According to Ghazvinian *et al.* (2019), mycelium-based composites are formed when mycelium growth on organic substrates is stopped during colonization by heating the mycelium. Heating the mycelium can kill and stop growth permanently. *P. ostreatus* and *L. squarrosulus* are members of the Phylum Basidiomycota, which can be used as inoculum fungi in the manufacture of biocomposites (Putra, 2020; Sari *et al.*, 2016). These two mushrooms were chosen because they have a high

growth rate of mycelium and also have a good ability to adapt to the environment and the level of productivity is relatively high.

The choice of media in the form of sugarcane bagasse and cornstalks is because these are natural materials categorized as agricultural waste that is commonly thrown away and can pollute the environment. Sugarcane bagasse is a lignocellulosic material, containing 52% cellulose, 25% hemicellulose, and 12% lignin (Fatriasari *et al.*, 2016), while cornstalks containing 35% cellulose, 6.8% hemicellulose, and 16% lignin (Yilmaz, 2015). The cellulose content found in sugarcane bagasse and cornstalks is the primary source of nutrition for the growth of fungal mycelium (Chang & Hayes, 1978). The bagasse fiber is water-insoluble, has good mechanical properties, is non-corrosive and has a density from 0.088 to 0.236 g/cm3 (Chen *et al.*, 2016) with a fiber length of 1.72 mm and a diameter of about 20 microns, making it suitable for processing into composite boards (Pramono *et al.*, 2019). Cornstalks have the potential to be used as a composite material to replace wood because it has a particle density of 0.210 g/cm^3 close to the wood bulk density of 0.23g/cm^3 (Astaria *et al.*, 2019). The two lignocellulosic materials have different lignocellulosic contents and fiber sizes. Mixing

lignocellulosic material in the form of sugarcane bagasse with cornstalks with a particular composition can affect the quality of the biocomposite to make the results more varied. The composition of the mixture will also affect the growth of fungal mycelium because the nutrients contained in the media are different (Sydor, et al, 2022).

Based on the explanation above, the problems to be solved were: whether the type of white-rot fungus with lignocellulosic material composition affects the quality of the resulting biocomposite; and which type of white-rot fungus with lignocellulosic material composition is optimal for the manufacture of mycelium-based biocomposite.

The purposes of this research were (1) to determine the effect of the type of white-rot fungus and the composition of the lignocellulosic material of agricultural waste that affects the quality of the resulting biocomposite board, (2) to obtain the optimal type of white-rot fungus and the optimal composition of lignocellulosic material from agricultural waste to manufacture mycelium-based biocomposite board.

The benefits obtained from this research are expected to solve the problem of handling many agricultural wastes and wood processing industrial waste encountered, which has often resulted in environmental pollution and to improve the quality of the environment by producing composite boards that can be recycled (biodegradable) making it an environmentally friendly material. The results of this research can be used as the basis for developing biocomposite manufacture through the mycelium-based composite manufacturing process to get an optimal composite product.

MATERIAL AND METHODS

The materials used in this research are pure cultures of white-rot fungi *Pleurotus ostreatus* and *Lentinus squarrosulus* from the supervisor's collection, sugarcane bagasse, cornstalk, sawdust, lime (CACO$_3$), corn grind, bran, Potato Dextrose Agar (PDA), distilled water, cotton wool, 70% alcohol, acetic acid, sulfate acid, and NaOH.

The tools used in this research are autoclave, laminar air flow, mould box long 30 x wide 30 x height 5 cm, sprayer, plastic boxes, plastic containers, Petri dishes, test tubes, tweezers, measuring cups, pipette dropper, measuring pipette, porcelain dish, scales, glass bottle, plastic wrap, aluminium foil, needle loop, spiritus lamp, weighing scale, analytical balance, oven, Erlenmeyer flask, measuring cup, glass goblets, funnels, stirring rods, filter paper, cameras, desiccators, and furnaces.

The research was conducted at the Mycology and Phytopathology Laboratory, the Microbiology Laboratory, Faculty of Biology, Jenderal Soedirman University, Siloam Creative Workshop Rejasari Purwokerto, and the Forest Products Technology Laboratory, Faculty of Forestry, Gadjah Mada University.

The research was conducted in experimental method with Completely Randomized Design (CRD) consisting of ten treatments with three replications. The treatments used two types of mushrooms (*P. ostreatus* and *L. squarrosulus*) with two types of Lignocellulosic materials (Sugarcane bagasse and Cornstalk) and each lignocellulosic material has 4 types of compositions (100%, 75%, 50%, and 25%). The treatments used are: A1= *P. ostreatus* in 100% Sugarcane bagasse, A2= *P. ostreatus* in 75% Sugarcane bagasse:25% Cornstalk, A3= *P. ostreatus* in 50% Sugarcane bagasse:50% Cornstalk, A4= *P. ostreatus* in 25% Sugarcane bagasse:75% Cornstalk, A5= *P. ostreatus* in 100% Cornstalk, A6= *L. squarrosulus* in 100% Sugarcane bagasse, A7= *L. squarrosulus* in 75% Sugarcane bagasse:25% Cornstalk, A8= *L. squarrosulus* in 50% Sugarcane bagasse:50% Cornstalk, A9 = *L. squarrosulus* in 25% Sugarcane bagasse:75% Cornstalk, A10 = *L. squarrosulus* in 100% Cornstalk.Parameters measured in this research are the main parameters and supporting parameters. The main parameters are fungal growth in composite materials, composite density, composite water content, composite thickness development, modulus of elasticity and modulus of rupture. While the supporting parameters are composite biodegradability.

a. Preparation of PDA Media
The material used in making PDA media is 200 grams of potatoes, peeled and then thinly sliced. Potatoes are boiled with 1.000 ml of distilled water in the pot until potato become softer. Then 20 grams of dextrose was put into the boiled potato water while homogenized and then 20 grams of agar plain was added and then homogenized again while heated. After homogeneous, the media can be poured into an Erlenmeyer tube and then closed using a plug. Then the media can be sterilized using an autoclave at a temperature 121ºC, pressure of 2 atm for 15 minutes.

b. Preparation of Mushroom Inoculum
The fungal inoculums of *P. ostreatus* and *L. squarrosulus* were rejuvenated on PDA medium in Petri dishes, and incubated at room temperature for 5-7 days. The fungal inoculum was then transmitted to sorghum medium, sorghum media consists of sorghum with the addition of 1% lime (CaCO$_3$) and sufficient water then put into a 300 ml glass bottle. The media in the bottle was then sterilized using an autoclave with a pressure of 1 atm for 30 minutes, and incubated at room temperature for 10-15 days.

c. Preparation of Lignocellulosic Materals (Elsacker et al., 2019)

According The lignocellulosic materials to be used are sugarcane bagasse and cornstalk, each material is crushed using a grinding machine and then filtered using a sieve then air-dried until completely dry and sterilized using an autoclave at a pressure of 2 atm for 30 minutes.

d. Mushroom Inoculation and Incubation

Mushroom were inoculated on logs containing lignocellulosic material (Widiastuti & Panji, 2008), then incubated at room temperature until fungal colonization was 75%. The results were transfered and cultured into the mould box by crushed and homogenized by hands aseptically, three logs to fill two mould boxes. The mould box covered with plastic and incubated until the mycelium had covered the entire material. After these conditions were achieved, all samples were taken out from the mold dan dried in an oven at 70°C for 5 to 10 hours, until the weight stabilized and all the water was evaporated (Elsacker *et al*., 2019).

e. Parameter Examination

1) Fungal Mycelia Growth On Composite Materials (Sudarma *et al*., 2013)

The mycelium growth in each baglog is measured on four sides of the baglog, so the average mycelium growth rate of each baglog was obtained. Mycelium length from the growing point was measured every seven days, until the mycelium growth reached 75% part of the baglog, it is considered that growth has reached maximum.

2) Composite Density

The test sample with length (l) 10 cm, width (b) 10 cm and thickness (d) 1 cm were prepared. Composite boards that have been made in a dry state is weighed. The length, width and thickness of composite board is measured.
The density of the composite board is calculated using the following equation (Ruhendi & Putra, 2011):

$$\rho = \frac{m}{V}$$

Information:
ρ : Composite board density (g/cm3)
m : Mass of composite board (g)
V : Particleboard volume (length (l)×width (w)×thickness (t)) (cm3)

3) Composite Water Content

The test sample with length (l) 10 cm, width (b) 10 cm and thickness (d) 1 cm were prepared. Composite board is weighed, the aim is the composite board is in a stable state. After weighing and the dry mass value is obtained, then the composite board is dried in an oven for 6 hours at a temperature of ±100ºC so that the water contained in the composite board undergoes evaporation and reaches a constant mass.

After drying, the composite board was weighed again, to obtain the dry mass value of the board after being baked. The amount of water content is calculated using the following equation (Ruhendi & Putra, 2011):

$$KA = \frac{ma\text{-}mk}{mk} \text{ x } 100\%$$

Information:
KA : Composite board water content (%)
ma : Initial mass of composite board (gr)
mk : Absolute dry mass of composite board (gr)

4) Composite Thickness Development

The test sample with length (l) 5 cm, width (b) 5 cm and thickness (d) 1 cm were prepared. The thickness of the dry composite board is measured, then the composite board is immersed in cold water for 24 hours. After soaking, then the composite board is measured again. Determination of the thickness expansion value can be calculated by using the following equation (Ruhendi & Putra, 2011):

$$PT = \frac{t2\text{-}t1}{t1} \text{ x } 100\%$$

Information:
PT : The thickness expansion of composite board (%)
t1 : Thickness of composite board before immersion (cm)
t2 : Thickness of composite board after soaking (cm)

5) Composite Modulus of Elasticity (MoE)

The test sample with length (l) 15 cm, width (b) 5 cm and thickness (d) 1 cm were prepared. The test sample dimensions of width (b) and thickness (d) was measured. The test sample is laid on the host machine of Universal Testing Machine (UTM) with a support distance of 15 cm (L). Then in the middle of the support distance a load is given and the loading is carried out until the elastic point limit of the test sample and observed. Moe and MoR values have units of MPa, where 1 MPa = 1 N/mm^2. The MoE value is calculated using the following equation (Wahyuni & Lapanporo, 2014):

$$MoE = (\Delta P \text{ x } L3) / (4 \text{ x } \Delta Y \text{ x } b \text{ x } d3)$$

Information:
MoE : Modulus of Elasticity (MPa)
ΔP : Change in the load used (kg)
L : Support distance (cm)
ΔY : Change in deflection at each change in load (cm)
b : Width of particle board test sample (cm)
d : Thickness of particle board test sample (cm)

6) Composite Modulus of Rupture (MoR)

The Modulus of Rupture was tested using the Universal Testing Machine (UTM) with the same sample as in MoE test. the Modulus of Rupture can be calculated using the formula (Allison, 1923):

$$MoR = (3 \text{ x } P \text{ x } L) / (2 \text{ x } b \text{ x } d2)$$

Information:
MoR : Maximum bending stress (broken modulus) (MPa)
P : Maximum load (kg)
L : Support distance (cm)
b : Width of particle board test sample (cm)
d : Thickness of particle board test sample (cm)

7) Composite Biodegradability Measurement

The test sample with length (l) 5 cm, width (b) 5 cm and thickness (d) 1 cm were prepared and composted on a pile of powder hardwood saw mixed with compost and added composting starter (EM4). Composting is carried out in the open air and every 10 days interval for 1 month is observed for biodegradability by measuring the ash content. The working procedure of this test is as follows (Saleh *et al.*, 2009):

Composite material samples were weighed as much as 5 grams in a porcelain dish. Then the sample is put in the furnace and heated to a temperature of 575°C for 4 hours. Then the sample was cooled in a desiccator and weighed until the weight remained constant. The equation calculates ash content:

$$\text{Ash content} = \frac{\text{ash weight}}{\text{sample weight}} \times 100\%$$

The research data from the main parameter were analyzed using Analysis of Variance (ANOVA) with an error rate of 5%, then further tested with post hoc Duncan at 95% confidence level to compare the effect between treatment.

The biocomposite boards scoring is made to determine the type of the best board. This scoring technique refers to Tarigan (2017), where particle board scoring is made by involving the main parameters test as well as the fulfillment of these parameters based on the standard used is JIS A 5908-2003 (Japanese Standard Association, 2003). The average score is divided into 5 with a detailed score of 1 for very bad, 2 for bad, 3 for moderate, 4 for good, and 5 for very good. The best board is determined based on the highest total score.

RESULT AND DISCUSSION

a. Fungal Mycelia Growth on Composite Materials

Based on Figure 1, variation in the growing media composition affects the average value of mycelium growth. The fiber content influences the growth rate of fungal mycelium in the media. The lignin content in cornstalks is greater than in bagasse, while the cellulose content in bagasse is greater than in cornstalks. Fungi use cellulose as a source of nutrition for mycelium growth. The higher the cellulose content in the media, the faster the mycelium will grow. High levels of lignin will make the mycelium slower to grow because the fungus must spend more energy and a longer time to degrade lignin in the media. Based on the statistical test, the treatment of fungus type and the lignocellulosic material used had a significant effect on the growth of the fungal mycelium. The result of Duncan's test (Figure 1) showed that treatment A6, A7, and A8 had a significant higher in the mycelium growth compared to other treatments.

According to Zuniar & Purnomo (2016), the difference in mycelium growth rate occurs because the levels of cellulose, lignin, pentosan and other substances in the lignocellulosic materials are different, so the lower the lignin content and the greater ability of fungi to decompose the lignin content, the faster the mycelium will grow. If the lignin content is high and the ability of fungi to degrade lignin is low, the mycelium will grow slowly. However, if the ability of fungi to degrade lignin is great, even though the lignin content is high, the mycelium can grow fast.

b. Composite Density

The density test results in Figure 2 showed that the density value of the biocomposite board varies between 0.120-0.177 g/cm^3. Based on the JIS A 5908-2003, all the biocomposite boards from this research did not meet the referenced standard, which is 0.40-0.90 g/cm^3 (Japanese Standard Association, 2003). Based on the statistical test on density of biocomposite board showed that treatment of the type of fungus and lignocellulosic material used had a significant effect on the density of composite. The results of Duncan's test (Figure 2) showed that treatment A7, A8, and A9 had a significant higher density value compared to other treatments. The difference in density is caused by variations in the thickness of the biocomposite board. This thickness difference can be caused by the spring back effect, which means the action of particles experiencing internal stress or the particles in the composite returning to their original state after the compression pressure is removed during the conditioning period (Eggertsen & Mattiasson, 2009). In addition, the uneven density can be caused by human error during the process of forming the biocomposite board, such as fungal colonization that has not been fully formed on the baglog but has been transferred to the muhsroom and uneven placement of the material on the mushroom during the transfer of culture. Haygreen & Bowyer (1989) explains that the higher the density of particle board, the higher the persistence properties of the resulting fracture of the board besides, the particle size also affects the value of strength broken a particle board.
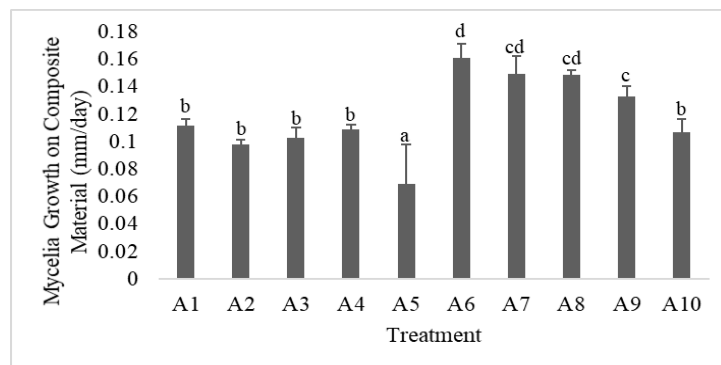
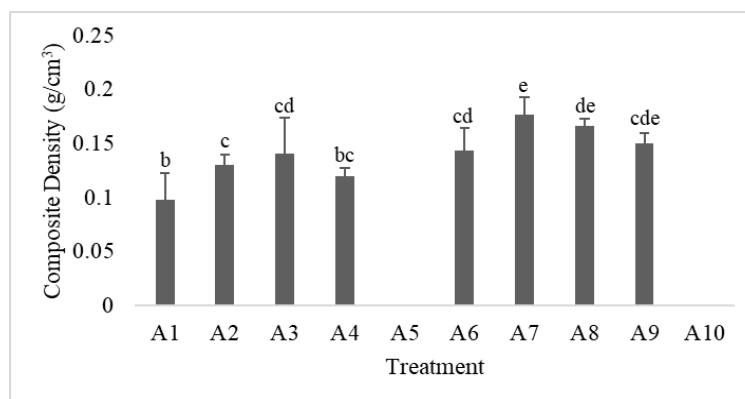**Figure 1.** Histogram of fungal mycelium growth on composite

Figure 2. Histogram of the average value of biocomposite density
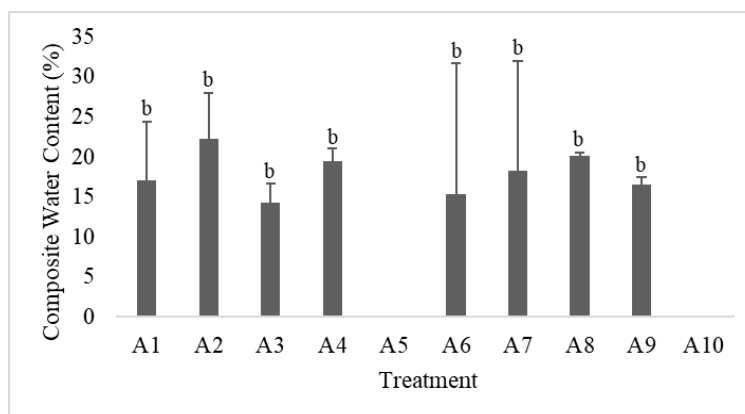

Figure 3. Histogram of the average value of composite water content

c. Composite Water Content

The water content value shows how much water is contained in the biocomposite board when it is in equilibrium with its environment. In this research, the water content of the biocomposite board was in the range of 14-22% (Figure 3). The water content value in this research did not meet the range accepted by the JIS A 5908-2003 standard, which is 5-13% (Japanese Standard Association, 2003). Based on the statistical test showed that the treatment of the type of fungus and the lignocellulosic material used had a significant effect on the water content of the composite. The results of Duncan's test (Figure 3) showed that all treatments exclude treatment A5 and A10 had a significant higher water content value.

The value of the water content of the biocomposite board is strongly influenced by water content of raw material. Therefore, the raw material's water content must be very low before being processed into boards. To make raw materials' water content low can be done by heating using an oven or air drying in an open space. The hygroscopic nature characteristic of the fiber also influences the value of the water content of the board due to its lignin and
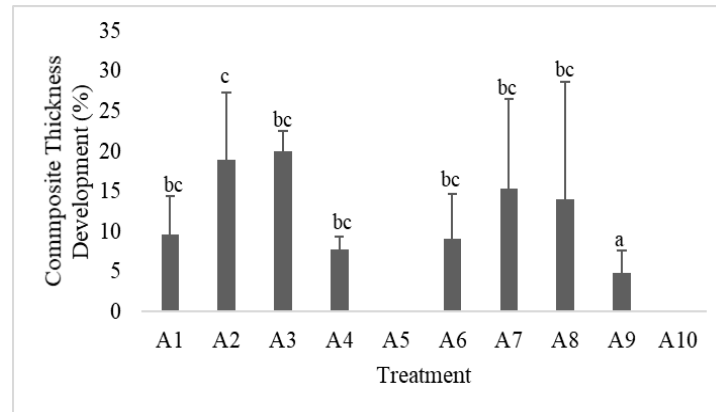
Figure 4. Histogram of the average value of composite thickness development

cellulose content. The cellulose content in sugarcane bagasse is higher than in cornstalks. In the dry state, cellulose is hygroscopic, hard, and brittle. The higher the value of the water content of the board, the not good the quality because the board becomes easy to absorb water and increases the risk of decay and damage. This is in accordance with Haygreen & Bowyer (1989), wood that has been subjected to temperatures above 100ºC for a long time becomes less hygroscopic because the higher the water content of the raw material makes the water content of the particle board higher.

d. Composite Thickness Development

The expansion value of the board thickness after soaking for 24 hours ranged from 4-19% (Figure 4). In the JIS A 5908, the thickness development board maximum is 12% (Japanese Standard Association, 2003), so only treatments A1, A4, A6, and A9 that met the standar of JIS A 5908. Based on the statistical test on the development of composite thickness showed that the treatment of the type of fungus and the lignocellulosic material used had a significant effect on the thickness development of the composite. The results of Duncan's test (Figure 4) showed that A2 had a significant higher composite thickness development compared to other treatments. Compared to the JIS A 5908 standard, the board thickness expansion values produced by these ten treatments were moderate to moderately high. The higher the thickness development value, the worse the board quality because the board will not stand being placed in humid conditions, which causes product damage to occur faster.

Besides being related to water absorption, the thickness of the board is also affected by the density of the board. In this research, the ten boards had a low density. This is due to the cavities formed on the board due to the unevenness of the adhesive and the material used containing pith. Sugarcane bagasse has a lot of pith, which causes water to be easily absorbed. According to Alghiffari (2008), pith is a water-absorbing material whose weight can increase seven times after immersion from its original weight.

e. Composite Modulus of Elasticity (MoE)

In Figure 5, it can be seen that the average value of the MoE ranges from 3,917,0000 - 42,923,6667 MPa. The MoE value is influenced by the type of raw material used. The length of sugarcane bagasse fiber and cornstalks affects the elasticity of the board. The longer the fiber from the raw material and the stronger the adhesive that binds the material, the higher the MoE value. This is in accordance with the statement of Muhdi *et al.* (2013) on testing the quality of particleboard using acacia wood harvesting waste (*Acacia mangium* L.). with urea-formaldehyde and phenol-formaldehyde adhesives, boards with a higher density have a higher number of bonds between particles than those with a lower density. All the biocomposite boards produced from this research have met the standard MoE value in JIS A 5908, a minimum MoE value of 2000 MPa (Japanese Standard Association, 2003). Based on the statistical test on the development of composite thickness showed that the treatment of the type of fungus and the lignocellulosic material used had a significant effect on the Modulus of Elasticity (MoE) of the composite. The results of Duncan's test (Figure 4.5) showed that A7 had a significant higher Modulus of Elasticity (MoE) compared to other treatments.

Besides being influenced by the type of raw material used, the MoE value is also influenced by the spread of the adhesive on the biocomposite board. Biocomposite boards produced from *P. ostreatus* adhesive had lower MoE values rather than *L. squarrosulus* adhesive. This is related to the growth of mycelium spread on the media in the mould box. The uneven spread of adhesive on the material in the muold box causes the resulting biocomposite board to be less strong so that the modulus of elasticity test results in a low value. It is appropriate and related to the parameters in this research's fungal mycelia growth on composite
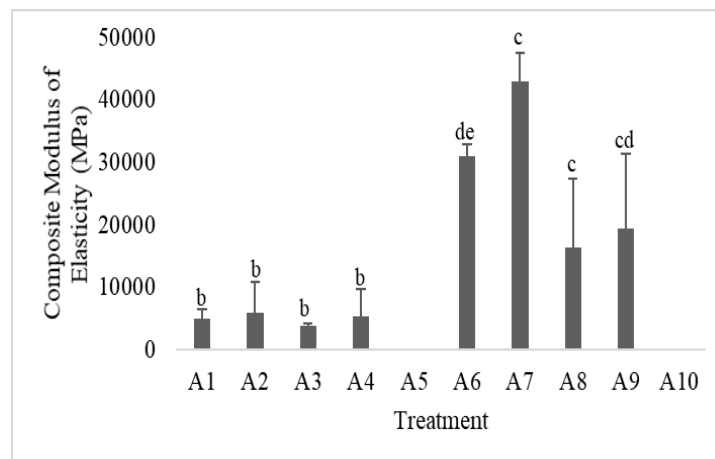
Figure 5. Histogram of the average value of composite Modulus of Elasticity
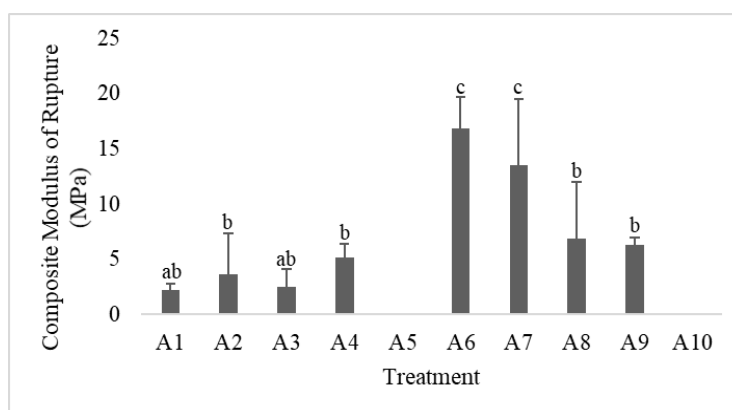

Figure 6. Histogram of the average value of composite Modulus of Rupture

materials that the development value of *P. ostreatus* mycelium was slow. This causes the lignocellulosic material not to be fully bonded, resulting in low board strength.

f.   Composite Modulus of Rupture (MoR)

Based on the Figure 6 The average value of the Modulus of Rupture (MoR) was between 2.2367 - 16.8800 MPa. Only biocomposite boards treated with A6 with an MoR value of 16.88 MPa and A7 with an MoR value of 13.57 MPa have met the standard MoR value in JIS A 5908, which is at least 8 MPa (Japanese Standard Association, 2003). Based on the statistical test on the development of composite thickness showed that the treatment of the type of fungus and the lignocellulosic material used had a significant effect on the Modulus of Rupture (MoR) of the composite. The results of Duncan's test (Figure 6) showed that A6 and A7 had a significant higher Modulus of Rupture (MoR) compared to other treatments.

The high MoR value is due to the uniform distribution of the adhesive on the raw material, causing the strength between the particles to increase so that a high rupture strength is obtained. In

addition, density also affects the MoR value, so the higher the density, the higher the MoR value. According to Maloney (1993), a density-increasing board will increase the properties of the particle board. In general, it can be said that the higher the density of particle board produced, the higher its strength. In Figure 6, the MoR value of biocomposite boards made from media with sugarcane bagasse composition has a higher value than cornstalks, and this is because sugarcane bagasse fiber density is greater than cornstalks. The higher cellulose content of sugarcane bagasse than cornstalks also gives strong properties to sugarcane bagasse fiber. This is in accordance with the statement of Yuanisa *et al.* (2015), that cellulose is a compound that makes up wood in the form of microfibrils. This compound has strong and rigid properties, so it is thought to be the main factor in increasing the strength of bagasse fiber.

g.   Composite Biodegradability Measurement

Based on the Figure 4.7, the ash content at a longer incubation time showed a lower value, this indicates that the composite board was successfully degraded by the microorganisms present in the
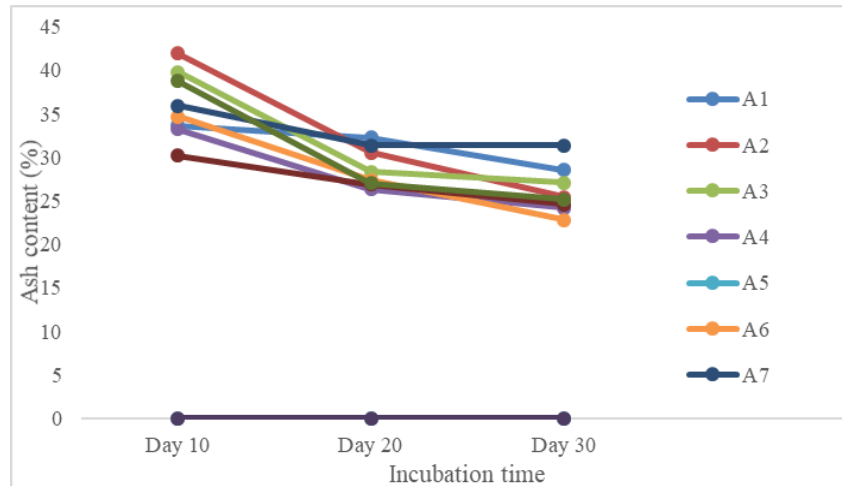
Figure 7. Graph of the percentage of ash content based on different incubation times

Tabel 1. Scoring biocomposite boards

Parameter	Treatment									
	A1	A2	A3	A4	A5	A6	A7	A8	A9	A10
Mycelium growth (mm/day)	0.11	0.09	0.10	0.11	0.06	0.16	0.14	0.14	0.13	0.10
▪ Average score	3	2	2	3	1	5	4	4	3	2
Density (g/cm³)	0.09	0.13	0.14	0.12	0.00	0.14	0.17	0.16	0.15	0.00
▪ Average score	2	3	3	2	1	3	5	4	4	1
▪ JIS	0	0	0	0	0	0	0	0	0	0
Water Content (%)	17.04	22.19	14.27	19.48	0.00	15.30	18.29	20.10	16.51	0.00
▪ Average score	4	2	5	3	1	5	3	2	4	1
▪ JIS	0	0	0	0	0	0	0	0	0	0
Thickness Development (%)	9.63	18.97	19.92	7.79	0.00	9.02	15.27	13.96	4.79	0.00
▪ Average score	4	2	2	5	1	4	3	3	5	1
▪ JIS	1	0	0	1	0	1	0	0	1	0
MoE (MPa)	4964.67	5915.33	3917.00	5383.33	0.00	30968.00	42923.67	16339	19418	0.00
▪ Average score	2	3	2	3	1	5	5	4	4	1
▪ JIS	1	1	1	1	0	1	1	1	1	0
MoR (MPa)	2.24	3.60	2.53	5.20	0.00	16.88	13.54	6.84	6.34	0.00
▪ Average score	2	3	2	3	1	5	5	4	4	1
▪ JIS	0	0	0	0	0	1	1	0	0	0
Total Score	17	15	16	19	5	27	25	21	24	6

Details:
Standar JIS A 5908 (2003): Average score: 1-5
Fulfill = 1, Unfulfill = 0   1 = Very bad, 2 = Bad, 3 = Moderate, 4 = Good, 5 = Very Good

sawdust media as well as the EM4 starter given periodically to the wood sawdust media effectively degraded the composite board sample. The highest ash content value was obtained from treatment A2 (*P. ostreatus* in 75% sugarcane bagasse: 25% cornstalk media) and the lowest from treatment A8 (*L. squarrosulus* in 50% sugarcane bagasse: 50% cornstalk) media. The results of the ash content test in the sample do not show a completely grayish-white color, but there is still several black color samples which means that there are still organic substances in the sample. This is in accordance with Sudarmaji *et al.* (2003) statement that the measured ash content is useful for knowing the mineral content in the sample material. The higher the ash content, the worse the quality of the resulting product because it indicates the high content of metal elements in the product.

The water content in treatment A2 was the highest among the other treatments (Figure 7), this also affects the value of the ash content because the wetter condition of the sample, the longer it will burn to evaporate the water content in it. This is in accordance with Danarti's (2006) statement that ash content is influenced by the water content of a material in the combustion process. This happens because it takes some of the energy produced by a product in combustion to evaporate the water contained in the product.

The results of the recapitulation of the assessment of the biocomposite boards produced in this research are presented in Table 1.

CONCLUSION

Based on the discussion, it can be concluded that the treatment of the type of fungus and the lignocellulosic material affect the quality of the resulting biocomposite board. *Lentinus squarrosulus* in 100% sugarcane bagasse media is the optimal type of white-rot fungus and the optimal composition of lignocellulosic material to manufacture mycelium-based biocomposite board

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