

Research Article

Degradation of Lignocelluloses Cocoa Shell (*Theobroma cacao* L.) by Various Types of Mould Treatments

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Lignocellulose can be degraded by lignocellulolytic microorganisms such as moulds. The purpose of the study was to obtain the right type of moulds in degrading lignocellulose on the cocoa shell powder. The study used a completely randomized design method using four treatments of different types of mould (*Trichoderma viride*, *Neurospora sitophila*, *Aspergillus niger*, and *Rhizopus oryzae*) towards cocoa shell powder fermentation. Solid fermentation of cocoa shell powder was carried out for 5 days in an incubator with a temperature of 30°C for *T. viride*, *N. sitophila*, and *R. oryzae*, while *A. niger* of 35°C. The fermented substrate was then dried in a cabinet oven with a temperature of 50°C for 4 days. Tests of lignin, cellulose, and hemicellulose were performed towards the treatments by the Chesson method, while the moisture content test was performed using the AOAC method. Degradation of fermented cocoa shell powder has shown a significant effect on moisture, lignin, cellulose, and hemicellulose contents. *Trichoderma viride* resulted in the highest lignocellulose degradation compared with the other treatments. The percentage decrease of lignin content is up to 46.69 wt%; while cellulose of 22.59 wt%; and hemicellulose is about 19.41 wt% from the initial lignin weight.

1. Introduction

Lignocellulosic residue has now become a concern along with the development of bioeconomy. The biomass with the basis of lignocellulose such as agricultural wastes was considered as a renewable resource that can be promoted for energy through biorefinery [1]. Valorisation of lignocellulose residue such as biorefinery has been recognized as a vital component to support arising bioeconomy by offering a diverse assortment of items from a broad range of biomass sources to fulfil the various requests of the society [2]. According to that, some utilization of various agricultural wastes needs to be addressed in order to enhance the economic, social, and environmental impacts through resources efficiency.

One of the potential agricultural wastes that still underutilized was cocoa shell. Cocoa fruit generally consists of 73.63% of shell (cocoa pod), 24.37% of seeds (generally,

there are 30–40 cocoa seeds in one fruit), and 2% of placenta (wrapping of cocoa seeds) [3]. On the processing of cocoa into chocolate products, cocoa shell needs to be separated from the cocoa. Cocoa shell is a thin, soft, and slightly slimy skin that has been separated from cocoa through the stripping process. Cocoa shell contains high fibre, so when the cocoa shell powder is dissolved, fibre and insoluble compound will settle after a few minute [4, 5].

Cocoa shell contains lignin and cellulose that amounted to 18.6% and 13.7% [6]. Crude fibre content such as lignocellulose is necessary to be degraded, so it can be digested. Lignocellulose can be degraded through degradation by chemical (basic, acid, solvent, and oxidation), physical (mechanical, thermal, and irradiation), and also biologically (enzymatic and biodegradation), which has been widely used so far [7–9]. While some combinations such as steam explosion, ammonia fibre extraction, and also several novel technologies, i.e., supercritical fluids and ionic liquids,

developed to make sure that the biocatalysts can access the complex polysaccharides and then convert them to simple sugar [9].

Several combinations of physicochemical and biochemical pretreatment in order for delignification also have been performed and aimed for the better process in resulting in simple sugar from the complex polysaccharides derived from the agricultural wastes. However, some physicochemical pretreatment still used high temperature that needed high energy and utilized acid or base compound that reacted corrosively towards the equipment that resulted high cost and not environment friendly [10, 11]. Biological pretreatments have been advanced as practical and harmless to the environment options to customary physicochemical treatments, which are costly and risky to the environment [12].

Biodelignification is an appealing methodology on account of its expense adequacy, low energy prerequisite, low climate effect, and low development of poisonous materials such as furfural and hydroxyl methyl furfural [13]. Microbiological delignification is selected as potential and environmentally safe methods for degrading lignocellulose. Mould has a better capability in decomposing lignin, hemicellulose, and cellulose than bacteria which only degrades lignin up to 50% [12, 14]. While, white rot mould has been known to have the ability to remove lignin up to 85% [15]. *Trichoderma viride* investigated as white rot mould, which is ideal microorganism for cellulose and hemicellulose hydrolysis, show delignification activity up to 56% that increased the biomass enzymatic degradability [16, 17]. Other moulds such as *A. niger*, *N. sitophila*, and *Rhizopus* sp. also have delignification activities through enzymatic releases such as CMCase, beta-glucosidases, endoglucanase, exoglucanase, pectinases, and xylanases [18].

The hydrolysis of complex components in lignocellulose is closely related to some enzymes involved in the hydrolysis such as cellulose, lignin peroxidase (LiP), manganese peroxidase (MnP), and laccase [19]. Cellulose enzyme resulted by mould is a mixture consisting of three enzymes, namely, endo β -glucanase, cellobiohydrolase, and β -glucosidase, which work synergistically in hydrolyze crystallized cellulose to glucose [20]. *Trichoderma viride* can produce large amounts of endoglucanase, while *A. niger* produces a cellulose enzyme containing β -glucosidase, yet low in exo and endoglucanase [21]. In addition to *T. viride* and *A. niger*, *N. sitophila* has high cellulolytic and hemicellulose activities in fermentation to produce simple sugars [22]. *N. sitophila* is strong amylolytic by α -amylase and glucoamylase enzymes production, yet inadequate in its lipolytic and proteolytic properties, while *Rhizopus oryzae* produce carbohydrate

degrading enzymes such as amylase, cellulase, xylanase, and glucoamylase [23]. Endoglucanase is an enzyme that attacks amorphous part of crude fibre, whereas β -glucosidase is an enzyme that converts cellobiohydrolase and oligosaccharide into glucose [24]. Amorphous portion of crude fibre hydrolyzed by endoglucanase will pave the way for cellobiohydrolase; however when cellobiose is accumulated in substrate, endoglucanase activity will be inhibited [25]. The purpose of this study is to obtain the right mould in degrading cocoa shells lignocellulose.

2. Materials and Methods

Dried cocoa (*Theobroma cacao* L.) obtained from a private plantation in West Java, *Aspergillus niger* and *Trichoderma viride* obtained from Microbiology Laboratory, Faculty of Mathematic and Natural Science, Padjadjaran University, and *Rhizopus oryzae* and *Neurospora sitophila* obtained from Microbiology Laboratory, School of Life Science and Technology, Institut Teknologi Bandung. Mould isolates were propagated for 3 days at optimum temperature of mould on in potato dextrose broth/PDB (Oxoid). The temperature used for propagation and fermentation was the optimum temperature of each mould, i.e., 35°C for *A. niger* and 30°C for *T. viride*, *R. oryzae*, and *N. sitophila*. After propagation, the mould isolates were then enriched for 5 (five) days on PDB until it reached 10^8 colonies/ml.

Cocoa shell was ground into cocoa shell powder, sieved with a 60-mesh sieve, and then sterilized using an autoclave at 121°C for 15 minutes. The sterile substrates were then inoculated with the mould isolates (*A. niger*, *T. viride*, *R. oryzae*, and *N. sitophila*) and incubated for 5 days at the temperature of 30°C (*T. viride*, *R. oryzae*, and *N. sitophila*) and 35°C (*A. niger*). After fermentation, the result was then dried at 50°C for 4 days, and the dried cocoa shell powder was milled and tested. Moisture contents were tested by the AOAC method; meanwhile, lignin, cellulose, and hemicellulose were tested by the Chesson method; then, the microstructure was tested with a scanning electron microscope.

2.1. Moisture Content Tests. Two empty dishes dried on an oven with the temperature of 105°C for 1 hour, cooled for 15 minutes, and then weighed until constant (Mi). Two grams of samples were prepared and put into the dish and dried at the temperature of 105°C for 1 hour, cooled for 15 minutes, then weighed until constant (Mf), and calculated using the following equation:

$$\% \text{moisture} = \frac{M_i - M_f}{M_i} \times 100 \text{weight B} - \text{weight C} \times 100\% \text{weight A.} \quad (1)$$

2.2. Lignin, Cellulose, and Hemicellulose Tests. Eight sheets of 90 mm Whatman filter paper were placed in an oven at 105°C for 1 hour and then cooled in a desiccator for 15

minutes. The filter paper was weighed until a constant weight was obtained. Two porcelain dishes were put into the furnace at a temperature of 600°C for 60 minutes and then

cooled in a desiccator for 30 minutes. The porcelain dish was weighed until a constant weight was obtained. Two sheets of constant filter paper were folded and put into a constant porcelain dish and then ashed in a furnace at 600°C for 3 hours. The weight of the filter paper ash was weighed and recorded, and then, the constant filter paper ash content was calculated.

One gram of sample (weight A) was put into an Erlenmeyer flask and added 75 ml of H₂O and then refluxed at 100°C in a waterbath for 1 hour. The reflux result was filtered with constant filter paper, and the residue was washed with 300 ml of hot water. The residue was dried in an oven at 105°C for 1 hour, cooled in a desiccator for 15 minutes, and then weighed until a constant weight (weight B) was obtained. The residue was put into an erlenmeyer and added 75 ml of H₂SO₄ 1N (Merck), and refluxed in a waterbath for 1 hour at 100°C. The results were filtered and washed until neutral with 300 ml of hot water. The residue was dried in an oven at 105°C for 1 hour, cooled in a desiccator for 15 minutes, and then weighed until a constant weight (weight C) was obtained. The dry residue was added 50 ml of H₂SO₄ 72% and soaked at room temperature for 4 hours. The residue was added with 75 ml of 1N H₂SO₄ and refluxed at 100°C in a waterbath for 1 hour. The residue was filtered and washed with 400 ml hot water until neutral. The residue was heated in an oven at 100°C for 1 hour and cooled in a desiccator for 15 minutes, repeated until the weight was constant and reweighed (weight D). The residue was ashed along with the filter paper in a furnace at 600°C for 5 hours. The weight of residual ash and filter paper was weighed, reduced by the constant weight of filter paper (weight E) and recorded, and then, the ash content was calculated using the following equations:

$$\text{Hemicellulose (wt\%)} = \frac{(\text{weight B} - \text{weight C}) \times 100\%}{\text{weight A}}, \quad (2)$$

$$\text{Cellulose (wt\%)} = \frac{(\text{weight C} - \text{weight D}) \times 100\%}{\text{weight A}}, \quad (3)$$

$$\text{Lignin (wt\%)} = \frac{(\text{weight D} - \text{weight E}) \times 100\%}{\text{weight A}}. \quad (4)$$

2.3. Microstructure Tests. Samples were coated by aluminium-palladium and affixed by carbon tape and then mounted. Sample surface height and the holder were adjusted and then tightened. The electron microscope (JEOL JSM-6360LA) was used to visualize at 10 kV of acceleration voltage [26].

2.4. Statistical Analysis. The study was done using experimental methods using completely randomized design (CRD) with 4 (four) treatments of different kinds of mould (*Trichoderma viride*, *Neurospora sitophila*, *Rhizopus oryzae*, and *Aspergillus niger*) with 4 (four) replicates. The data were analysed with analysis of variance (ANOVA) to determine

the significance of the treatment applied and followed by the Duncan test to identify the difference among the treatments.

3. Results and Discussion

3.1. Moisture Contents Identification. Moisture contents have a significant different effect due to the mechanism of respiration and hydrolysis of lignocellulose by mould. The high moisture content produced may be due to accumulation of water from the cellular respiration mechanism and the low heat transfer on substrate, so evaporation rate tends to be low and results in water contained in substrate as well metabolite yield accumulated in the system and increasing moisture content of material [27]. Low moisture content is caused by hydrolysis of lignocellulose component by enzyme that require large amounts of water, although it produced water as a byproduct of respiration, yet it required more water to degrade lignocellulose compared to water produced by respiratory of byproduct [28].

A. niger, *N. sitophila*, and *T. viride* used water to produce secondary metabolite which is enzyme, whereas *R. oryzae* used water to supply the need of primary metabolite respiratory, so that water needs of *R. oryzae* was much more than other moulds. Decrease in moisture content can be due to changes of catabolic heat on substrate surface that causes water evaporation from inside to outside of the system. Overall moisture content of degraded cocoa shell was 4.11%–4.52%. According to the Indonesian National Standard, water content of cocoa shell is maximum of 5%. So, in this study, cocoa shell powder has fulfilled the moisture content requirement; on the other hand, it showed that cocoa shell powder was dried enough, so it can be stored in long term and does not require particular treatment. It can be stored in sealed container at room temperature to avoid mechanical damage and moisture absorption from surrounding environment [29]. If the moisture content is known, then it can be adjusted to storage condition to minimize the capability of microorganism growth after the fermentation process stopped.

3.2. Lignin Degradation of Cocoa Shell. Based on the result of ANOVA, mould addition gave a significant effect towards lignin content of cocoa shell (Table 1). Lignin prevents the entry of enzyme in hydrolyze polysaccharides into monosaccharides. Lignin binds hemicellulose and cellulose and forms a layer as anchoring from penetration of solution [14]. In this study, lignin content has a significantly different effect because each mould has different abilities to degrade lignin depending on the production of extracellular enzyme.

Cocoa shell powder fermented by *T. viride* has the lowest lignin content than other treatments. *T. viride* could reduce lignin content from 17.20% to 9.17%, which means a decrease in lignin content by *T. viride* reached 46.59% on cocoa shell powder. *T. viride* is white decaying fungi that can degrade lignin of in wheat up to 56% by producing lignin-degrading enzyme [16]. According to Saili et al. (2014), most of *Trichoderma*-type moulds produce polyphenol oxidase to degrade lignin.

TABLE 1: Characteristics of cocoa shell.

Treatment	Moisture (wt%)	Lignin (wt%)	Cellulose (wt%)	Hemicellulose (wt%)
Cocoa shell	13.67	17.20	31.87	48.26
<i>Trichoderma viride</i>	4.52 ^b	9.17 ^a	24.67 ^a	38.89 ^a
<i>Neurospora sitophila</i>	4.46 ^b	12.24 ^b	25.46 ^b	40.27 ^b
<i>Aspergillus niger</i>	4.42 ^b	15.02 ^c	27.60 ^c	42.38 ^c
<i>Rhizopus oryzae</i>	4.11 ^a	16.47 ^d	29.77 ^d	45.24 ^d

Treatment marked with the same letter showed no significant difference at 5% level according to the Duncan test.

A decrease of lignin with fermentation treatment of *N. sitophila* was lower than *T. viride* which is about 28.84%. *N. sitophila* can reduce lignin level up to 25% by producing laccase enzyme [30]. In the lignin degradation process, laccase oxidizes phenol to be phenoxyl. A decrease in lignin level by *A. niger* was 15.02%, which means only 12.67% from the initial lignin content. In accordance with Silva et al. (2010), *Aspergillus* sp. can degrade lignin up to 26.5% by producing high number of laccase.

Based on Table 1, *R. oryzae* could reduce lignin level about 4.24% from initial lignin content, where decrease by this mould showed lower percentage than other three moulds. According to Viji and Neelanarayanan (2015), lignin degradation by *R. oryzae* can reach 23.8% in rice straw, which is the lowest if compared with other moulds such as *A. oryzae* that can reduce lignin level to 52%, *A. fumigatus* of 58%, and mix culture of *R. oryzae*, *A. oryzae*, and *A. fumigatus* of 93.8%. It showed that treatment with *R. oryzae* and *Aspergillus* can reduce lignin level. However, the mix culture can result in higher decreasing percentage of lignin level.

R. oryzae type moulds generally produce more cellulose enzymes, hemicellulose, pectinase, amylase, protease, and lipase, but some of them can produce tyrosinase and peroxidase to hydrolyze lignin such as ENHE type of *R. oryzae* [31]. Highest lignin degradation was resulted using Basidiomycetes types such as white rot fungi, brown rot fungi, and soft rot fungi. *A. niger*, *N. sitophila*, and *T. viride* are included in soft rot fungi class because *A. niger* and *N. sitophila* are microfungi from phylum Ascomycotina and *T. viride* is microfungi from subphylum Deuteromycota [32].

3.3. Cellulose Degradation of Cocoa Shell. Based on the result of ANOVA, mould addition treatment gave a significant effect towards cellulose content of cocoa shell. In this study, cellulose content presented a significantly different effect because the capability of mould in degrading cellulose is varying. *T. viride* succeeded in decreasing cellulose level of cocoa shell to 22.59% of total initial cellulose. *Trichoderma* produces endoglucanase to degrade cellulose up to 20% [33]. *T. viride* is a type of mould that can destroy high level of cellulose and capable to synthesize several essential factors to dissolve a strongly bonded cellulose with hydrogen bond [34]. The bonded cellulose is converted to glucose and simple sugars by using cellulose enzyme produced by the mould [21].

The superiority of *T. viride* as a producer of cellulose enzyme is due to its capability to produce complete cellulose

required to hydrolyze crystalline cellulose [35]. The mycelium of the mould can produce variety of enzymes, including cellulose and chitinase enzymes. Mould generally has cellulose because of its habitat in organic materials containing cellulose. Cellulose consists of endo- β -1,4-glucanase, exo- β -1,4- glucanase, and β -1,4 glucosidase (cellobiase), which can be produced by *T. viride* [36].

All of these enzymes are hydrolytic and work either consecutively or simultaneously. Cellobiohydrolase is an enzyme that has affinity towards high level of cellulose capable of breaking crystalline cellulose, whereas endoglucanase acts on amorphous cellulose [37]. Cellobiohydrolase shatters cellulose by cutting the hydrogen bonds that cause the glucose chains to be readily hydrolyzed. Further hydrolysis is carried out by β -glucanase and β -glucosidase, resulting in glucose.

N. sitophila could degrade cellulose to 20.11%, whereas *A. niger* degrade cellulose up to 13.39% of initial cellulose. *Neurospora* sp. utilizes existing carbon compound in rice and wheat husk to produce cellulose enzyme, while *Aspergillus* sp. utilizes ammonium salt to produce cellulose enzyme [38, 39]. *A. niger* produce cellulose enzyme containing many β -glucosidase, yet low exo and endoglucanase [40]. *N. sitophila* under optimum condition can produce cellulose enzyme (endoglucanase, exoglucanase, and β -glucosidase) at 35°C and pH 5 on banana substrates [41].

Treatment of *R. oryzae* results in a low decrease of cellulose level since it can only reduce cellulose level from 31.87% to 29.77%, which means only 6.59% of initial cellulose. *R. oryzae* provides endoglucanase and exoglucanase on certain substrates to degrade cellulose (Ghosh and Rani Ray, 2011). *R. oryzae* can produce a relative high level of exoglucanase within a short time from dried flower and lemon peel by utilizing the substrate as a carbon source [42]. This statement is supported by Viji and Neelanarayanan (2015), which states *R. oryzae* can degrade cellulose up to 16.74% in rice husk. Cellulose degradation by *R. oryzae* is the lowest, since *R. oryzae* can only produce endoglucanase and exoglucanase, while other moulds may also produce β -glucosidase.

3.4. Hemicellulose Degradation of Cocoa Shell. Based on the result of ANOVA, the moulds treatment significantly affected hemicellulose content of cocoa shell. Hemicellulose content gave a significantly different effect due to the varying abilities of mould to degrade hemicellulose and cellulose hydrolysis. One of the most widely studied hemicellulose-degrading enzymes is xylanase [34, 43]. Xylanase is an

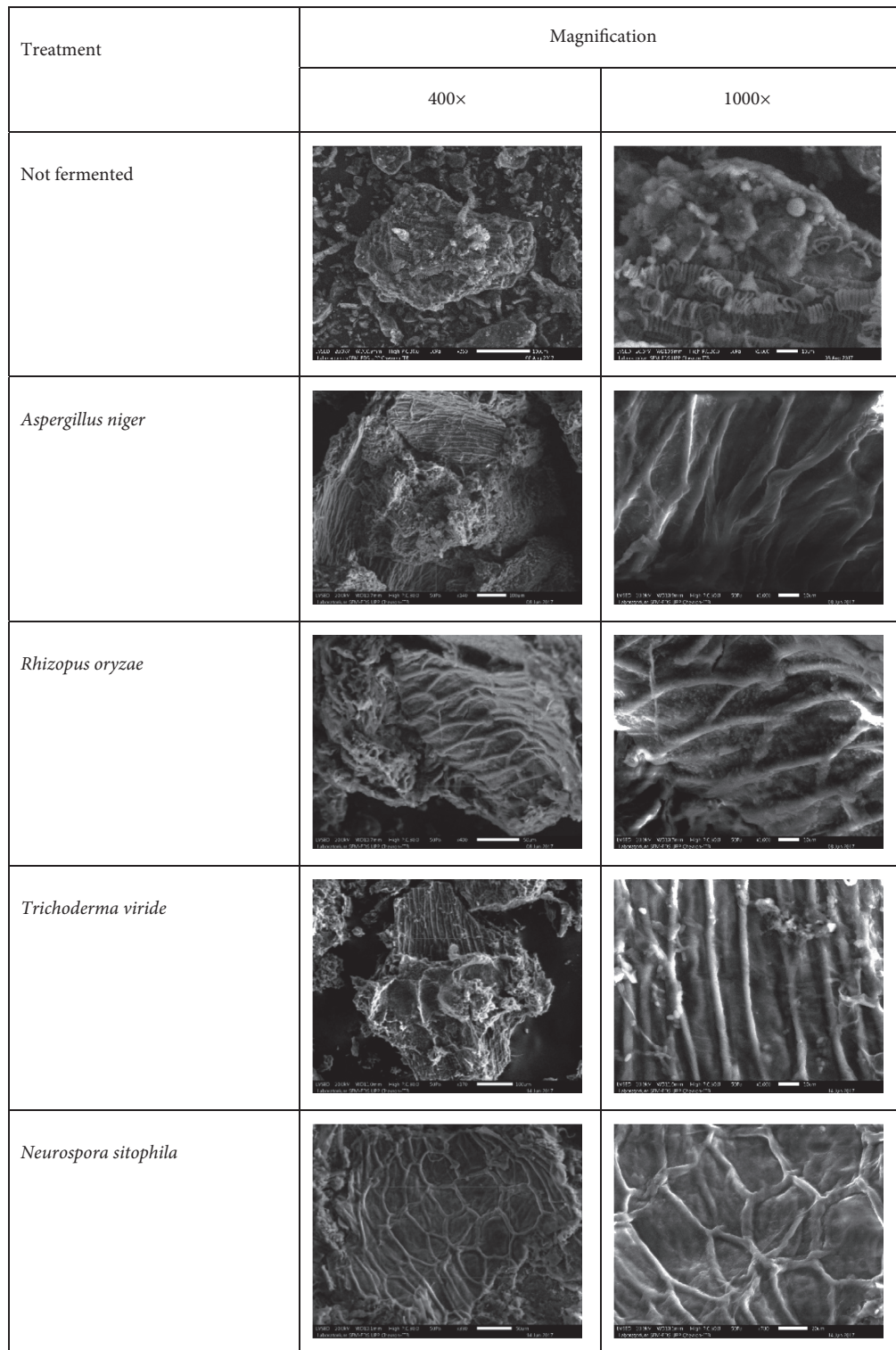


FIGURE 1: Microstructure of cocoa shell powder.

enzyme that catalyzes the hydrolysis bond on xylan which is one of the principal constituents in hemicellulose [44]. Endo-1,4- β -xylanase cuts the glycosidic bond on xylan backbone and reduces the degree of substrate polymerization. Xylan is not attacked randomly; however, the bond chosen for hydrolysis depends on the nature of substrate

molecule such as length of the chain, degree of branching, and the presence of substituents [45].

Xylanase is widely produced by microorganism such as *Trichoderma* sp. and *Aspergillus* sp. [46]. In addition, *T. viride* also produces all xylanolytic enzymes [47]. Based on results, the treatments of *T. viride* and *A. niger* reduced

hemicellulose contents, respectively, 19.42% and 12.18% from initial hemicellulose of cocoa shell. According to [48], *N. sitophila* has capability of cellulolytic and hemicellulolytic activities on fermentation process to produce simple sugars. *N. sitophila* grows normally on substrates containing only carbon sources, organic salts, and biotin. The carbon sources are mono-, di-, or polysaccharides. According to the statement, hemicellulose contents of *N. sitophila* treatment of 40.27% decreased 16.56% from initial hemicellulose.

In accordance with [49], *Rhizopus* sp. produces endo and exopolysaccharonase enzyme to degrade hemicellulose. Based on the results of hemicellulose contents known, *R. oryzae* treatment reduced hemicellulose level from 48.26% to 45.24%, which is 6.26% decreased. The low degradation of hemicellulose may be caused by hemicellulose content in cocoa shell that contains low galacturonan. Polysaccharonase, one of pectin class enzymes, plays a role in the process of ripening fruit naturally [50].

3.5. Microstructure of Treated Cocoa Shell Powder. Based on Figure 1, there was a difference on surface of initial cocoa shell that has not been fermented by mould, where it still had dense and complex lignocellulose structure, while on the surface of fermented cocoa shell was more tenuous in the structure. Dense arrangement of structure indicates cocoa shell has high lignocellulosic level. Surface of cocoa shell is a nonuniform particle which exhibits lignocellulose composed of various compounds, namely, lignin, cellulose, hemicellulose, and other compounds such as pectin and polyphenol [51].

Each treatment showed different degradation effects, as shown in Figure 1. Lignocellulose structure of cocoa shell which has been delignified by *N. sitophila* and *R. oryzae* has similar structure; however, structure of cocoa shell treated by *N. sitophila* tends to remain mutually binding with tenuous distance, while the structure of cocoa shell treated by *T. viride* has similar structure of cocoa shell treated by *A. niger*. Lignocellulose structure of cocoa shell treated by *T. viride* tends not to bind and has tenuous distance compared to *A. niger* which tends to be linked to each other. It may be due to the difference in the capability of extracellular enzyme produced by mould to break lignin bonds against cellulose and hemicellulose.

The tenuous and discontinuous lignocellulose structure of *T. viride* treatment is caused by enzymes activity either successively or at the same time degrading lignin, cellulose, and hemicellulose bonds. Cellobiohydrolase enzyme produced by *T. viride* is able to break down part of crystalline cellulose, whereas endoglucanase acts on cellulose amorphous [24]. Cellobiohydrolase breaks down cellulose by cutting the hydrogen bonds that cause glucose chains to be readily hydrolyzed. Further hydrolysis is carried out by β -glucanase and β -glucosidase, thus producing glucose.

Lignocellulose structure that tends to bind to treatment of *A. niger* and *N. sitophila* is caused by the work of less optimal laccase enzyme. Laccase has ability to break α and

β -carbon bonds in lignin (Janusz et al., 2017). Dense structure is due to low endoglucosidase enzyme produced by *A. niger* so that amorphous portion of cellulose is still formed, in contrast to lignocellulose structure treated by *N. sitophila* that has tenuous structure because *N. sitophila* produce higher endoglucosidase, so amorphous part of cellulose was split [52]. The lignocellulose structure of *R. oryzae* treatment that binds and tends to be denser than lignocellulose structure of *N. sitophila* treatment caused by enzymes produced by *R. oryzae* is less able to degrade lignin, cellulose, and hemicellulose.

In accordance with [53], cellulose and hemicellulose cannot be converted directly because it is associated with lignin, so the structure is tight. When mould produces extracellular enzyme, cellulose and hemicellulose bonds with lignin can be disconnected within lignin itself. The disconnected bond was hydrogen bond between cellulose and lignin or hemicellulose and bonds between lignin and hemicellulose in the form of an ether or ester group. The extracellular enzyme produced may also break the bonds in lignin compound comprising the ether group and carbon bonds into lignin fragments [54]. The activity of lignocellulosic enzyme also leads to bond breaking in lignin compound that causes lignin to be degraded. Lignin degradation is started by attacking hydrogen atom attached on the phenolic group by hydroxyl ion.

4. Conclusions

Fermentation of cocoa shell powder by *Aspergillus niger*, *Rhizopus oryzae*, *Trichoderma viride*, and *Neurospora sitophila* has a significantly different effect on lignin, cellulose, and hemicellulose contents and then no significant difference at moisture content on *T. viride*, *N. sitophila*, and *A. niger*. *Trichoderma viride* were mould which show highest cocoa shell-degrading ability with percentage of decrease of lignin, cellulose, and hemicellulose level of 46.69%, 22.59%, and 19.42% respectively. *Neurospora sitophila* degraded lignin of 28.84%, cellulose of 20.11%, and hemicellulose of 16.56%, *A. niger* degraded lignin of 12.67%, cellulose of 13.39%, and hemicellulose of 12.18%, while *R. oryzae* degraded lignin, cellulose, and hemicellulose of 4.24%, 6.59%, and 6.26%, respectively.

Cocoa shell powder fermented by *Trichoderma viride* is potent as alternative source that is possible to be mixed with the main raw material for cocoa powder production. High number percentage of lignin, cellulose, and hemicellulose that degraded by fermentation processes resulting in potential cocoa shell powder that shown digestible characteristics that can be identified further. Delignification of cocoa shell powder by *Trichoderma viride* has shown the effectiveness of lignocellulose waste pretreatments which can be a source of new raw materials that can maximize the utilization of cocoa shell.

Data Availability

The data used to support this study are included within the article.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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