

## Research Article

# Characteristics of Biofoam Cups Made from Sugarcane Bagasse with *Rhizopus oligosporus* as Binding Agent

### Eti Indarti (),<sup>1,2</sup> Sri Muliani (),<sup>1</sup> and Dewi Yunita ()<sup>1</sup>

<sup>1</sup>Agricultural Product Technology Department, Faculty of Agriculture, Universitas Syiah Kuala, Banda Aceh 23111, Indonesia <sup>2</sup>Master Program of Agriculture Industrial Technology, Faculty of Agriculture, Universitas Syiah Kuala, Banda Aceh 23111, Indonesia

Correspondence should be addressed to Eti Indarti; eti\_indarti@unsyiah.ac.id

Received 8 December 2022; Revised 1 March 2023; Accepted 14 March 2023; Published 30 March 2023

Academic Editor: Kinga Pielichowska

Copyright © 2023 Eti Indarti et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

This study is aimed at producing a biofoam cup made from sugarcane bagasse with tempeh mold (*Rhizopus oligosporus*). Soybean flour (SF) was added to promote the growth of mycelia, which could bind the bagasse fiber matrix. The main materials were whole bagasse (B) and depithed bagasse (DB). The SF weight ratios to bagasse were 1:1 (SF1) and 1.5:1 (SF1.5). Therefore, the studied specimens were labeled B-SF1, DB-SF1, B-SF1.5, and DB-SF1.5. All biofoam cups were analyzed for their physical properties (water absorption and porosity), mechanical properties (puncture and compressive strengths), biodegradability, and thermal properties (thermogravimetric analysis). The lowest water absorption rates were obtained from the B biofoam cups ( $23\% \pm 2.45\%$ ) and the SF1.5 biofoam cups ( $25.83\% \pm 5.19\%$ ). Both B-SF1 and B-SF1.5 had lower porosity ( $8.72\% \pm 0.88\%$  and  $10.77\% \pm 1.54\%$ , respectively) than the DB biofoam cups. Moreover, the B biofoam cups showed the highest puncture strength ( $2.95 \pm 0.37$  kg cm<sup>-2</sup>) among all samples. Nevertheless, the B-SF1.5 biofoam cup had the highest compressive strength ( $3.98 \pm 0.39$  MPa) and the DB-SF1.5 exhibited the slowest degradation rate ( $27\% \pm 0.7\%$ ) after 14 days of soil burial. The highest thermal stability was obtained from B-SF1.5, which had a thermal degradation temperature of  $264^{\circ}$ C. Overall, B-SF1.5 had the smoothest surface, good thermal stability, and high compressive strength.

#### 1. Introduction

Online food and product ordering services are a source of ever-increasing plastic waste. According to a survey administered through social media on online food sellers in Wonomulyo, West Sulawesi Province, Indonesia, 65.3% of online food sellers use Styrofoam packaging [1]. A survey of 200 respondents in Banda Aceh and Aceh Besar, Aceh Province, Indonesia, in 2021 showed that 24.5% of the respondents ordered food online very often, and Styrofoam was the most widely used type of food packaging (68.5%) [2].

Styrofoam is made of polystyrene, which is a strong plastic material composed of styrene and benzene [3]. Styrofoam cannot be decomposed by microorganisms that exist in soil and nature, and the burning of Styrofoam waste produces dioxins, which are carcinogenic [4, 5]. Packaging made from biodegradable materials, such as sugarcane bagasse, can be developed to decrease the use of Styrofoam. Sugarcane bagasse is often scattered on roadsides, causing environmental pollution and degrading the overall appearance of places. Therefore, Styrofoam must be replaced with biodegradable foam (biofoam). This material is not carcinogenic and is thus safe for public health; it can also be decomposed by microorganisms and is therefore environmentally friendly.

The manufacture of biofoam using natural fibers has been extensively studied. Natural fibers from agricultural sources, including polysaccharides, are environmentally friendly polymers. Biomaterials from natural fibers are biodegradable, easy to obtain, nontoxic, and inexpensive.

The addition of wood cellulosic fibers to tannin-based foam (TBF) decreased the foam cell size and increased the foam cell density. With the addition of 2% natural fibers, the foam's compressive strength was 0.640 MPa. TBFs reinforced with 2 wt% wood fibers provided the best combination of mechanical and thermal properties and foam cell morphology [6]. Sawdust-based biofoam created using mushroom mycelium exhibited a compressive strength of 350-570 kPa [7]. Biofoam materials were prepared using 50% petroleum-based polyol and soy-oil-based polyol with bagasse in various amounts as a natural filler. The addition of 5 wt% bagasse increased the compressive strength of the biofoam from 3.924 to 4.274 kPa and slightly enhanced its thermal stability [8]. Other researchers studied the optimal properties of a cassava starch biofoam cup with the addition of cotton-fiber-reinforced rubber latex [9]. The synthetic polymer expanded polystyrene was binding with mycelia from macrofungus biofoams, Pycnoporus sanguineus and Lentinus velutinus [10]. In recent studies, synthetic substances are utilized as the main materials and natural sources serve as substitutes and reinforcements. Therefore, the novelty of this research is the development of a fully natural material from biomass (bagasse) and its reinforcement using mycelia produced by the tempeh mold, Rhizopus oligosporus.

The use of mycelia as a binding agent in composite materials enhances the properties of biocomposites. Mycelia can produce strong, sturdy structures; the chitin and glucan protein contents of mycelia and the protein in the mycelium cell walls determine the properties of the resulting biocomposite [11]. Therefore, mycelia from tempeh mold are a potential reinforcing agent for bagasse to form fully natural biofoam.

Bagasse waste can be a raw material for biofoam because bagasse has high cellulose content (35.01%). This polymer is advantageous because it is available year-round, is renewable, and naturally deteriorates due to its lignocellulosic components [12]. In addition, bagasse can be utilized as a medium for the growth of *R. oligosporus* because it contains carbon (47%) and nitrogen (2.5%). In the manufacture of biofoam, mycelium growth is also influenced by the carbon source contained in the substrate used. The availability of carbohydrates (good carbon source) and protein (nitrogen source) fulfills the structural and energy growth needs of mycelium cells. Carbon compounds provide the energy mycelia's need to complete their life processes [13].

In this study, whole bagasse (B) and depithed bagasse (DB) were used as raw materials in the manufacture of biofoam cups with the addition of *R. oligosporus*, the starter culture used to make tempeh, a traditional Indonesian food made from fermented soybeans. The mycelium growth should strongly bind the bagasse fibers to existing soybean flour (SF) as a nutrient source. *R. oligosporus* mycelia can bind the matrix of bagasse fibers. However, *R. oligosporus* needs a protein source for growing and producing mycelia [13]. Soybeans are a healthy food that is rich in protein and minerals. The protein content of soybeans reaches 40%; in comparison, the protein content of other beans is only 20%–25% [14]. Therefore, in this study, SF was used as a protein source for the growth of *R. oligosporus* in the manufacture of biofoam cups from bagasse waste.

#### 2. Materials and Methods

2.1. Materials. Sugarcane bagasse was collected from a sugarcane juice producer near Universitas Syiah Kuala in Banda Aceh, Aceh Province, Indonesia. SF with an 80-mesh particle size (brand: Tani Kepyar; producer: CV Sidatani Sembada, Yogyakarta, Indonesia) and tempeh mold (brand: Raprima; producer: PT. Aneka Fermentasi Industri, Bandung, Indonesia) were purchased from a shop in Banda Aceh. Raprima's tempeh mold, preserved in its dry state, was about 80-mesh particle size flour and usually used as a starter in making tempeh. The tempeh mold contains *R. oligosporus*, whose spores can develop into mycelia. Plastic drinking cups were used for casting, and small holes were made around the outer side of each cast.

2.2. Preparation of Sugarcane Bagasse. Samples of B (Figure 1(a)) and DB (Figure 1(b)) were prepared by sundrying B for two days. For DB, the dried bagasse was combed with a wire brush to remove the pith and then cut using scissors into small pieces measuring approximately 0.5 cm (Figure 1(c)). All samples were kept in a plastic bag and placed in a refrigerator before the next step.

2.3. Fabrication of Biofoam Cups. The prepared bagasse was autoclaved at 121°C for 15 min for sterilization. The sterilized bagasse was cooled to room temperature for approximately 1 h. The bagasse in DB and that in B were manually mixed with SF in bowls at weight (g) ratios of 1:1 (SF1) and 1:1.5 (SF1.5). The tempeh mold (*R. oligosporus*; 60% of bagasse weight) was added to each treatment. Each mixture was cast between two stacked plastic cups and incubated for three days at room temperature (Figure 2). The produced biofoam cups were dried in an oven at 50°C for 48 h to stop mold growth.

2.4. Characterization of Biofoam Cups. The biofoam cups were analyzed for their physical properties, namely, water absorption, porosity, and density. Their structural and mechanical properties (puncture and compressive strengths), biodegradability, and thermal properties were also assessed. All analyses except the morphological and thermal analyses were conducted in triplicate on all samples (a total of 12 specimens).

2.4.1. Water Absorption and Porosity. The standard test method reference TAPPI T 441 om-09 was followed to analyze the water absorption properties of the biofoam cups. Each sample was weighed, and its weight was recorded as its initial weight  $(m_0)$ . Then, each sample was immersed in 200 mL of water for 900 s, dried using a tissue to remove the remaining water adhering to its surface, and then weighed; this weight was denoted as  $m_b$ . Water absorption (%) was calculated as follows:

Water absorption = 
$$\frac{m_{\rm b} - m_{\rm o}}{m_{\rm o}} \times 100\%.$$
 (1)

Porosity was analyzed by subtracting the dry membrane volume  $(v_2)$  from the wet membrane volume  $(v_1)$  and then dividing the quotient by the dry mass volume (v) [15].

$$Porosity = \frac{v_1 - v_2}{v} \times 100\%.$$
 (2)



FIGURE 1: (a) Bagasse, (b) depithed bagasse, and (c) bagasse after cutting into small pieces.



FIGURE 2: Polypropylene plastic cup for biofoam cup casting: (a) before use and (b) after use.

2.4.2. Morphology. Surface morphologies were characterized using a digital microscope endoscope (Lolovi) that was connected to a computer. The fracture and surface of each sample were placed in front of the microscope lens vertically at 1600x magnification.

2.4.3. Puncture Test and Compressive Strength Analysis. Puncture strength analysis was performed using a texture analyzer according to SNI:8058:2014. The analysis settings were arranged so that the probe was used, and the load range was selected so that failure occurred between 10% and 90% of the full-scale load. The specimen was placed in the middle and between the clamping plates. The test was performed at a machine speed of  $300 \pm 10 \text{ mm min}^{-1}$  until the pressure rod caused the test object to collapse. Then, the average puncture strength was calculated, and the standard deviation of all test results was determined.

Compressive strength analysis was performed using the texture analyzer with a TA 18 probe at a speed of  $1 \text{ mm s}^{-1}$ . The distance between the probe and the material was 0.5 cm. The average compressive strength was determined from three measurements [16].

2.4.4. Biodegradability. Biofoam biodegradability analysis was conducted according to the soil burial test method [9]. The biofoam samples were cut into  $2.5 \text{ cm} \times 4 \text{ cm}$  pieces and soaked in water for 1 min. Then, their initial weights  $(w_o)$  were measured. A 20 cm tall box containing soil was prepared. Each biofoam sample was wrapped in gauze and then buried in soil for 14 days. After immersion, the sample

was cleaned of soil debris, and its final weight  $(w_1)$  was determined. The percentage loss in weight was calculated as follows:

Weight lost = 
$$\frac{(w_o - w_1)}{w_o} \times 100\%$$
. (3)

2.4.5. Thermogravimetric Analysis (TGA). TGA was conducted using a Mettler TG-50 module attached to a Mettler TC-11 4000 thermal analyzer (USA). The specimen was analyzed in a nitrogen atmosphere at a flow rate of 50 mL min<sup>-1</sup>. The temperature was set from 30°C to 600°C at a rate of 20°C/min [17].

*2.5. Statistical Analysis.* Data from the analysis of physical properties, mechanical properties, and biodegradability were analyzed using analysis of variance (ANOVA) tests.

#### 3. Results and Discussion

3.1. Water Absorption. The water absorption of the biofoam cups is directly affected by the mycelium condition on the surface and the internal opening of the biofoam. The ANOVA test suggests that the water absorption of the biofoam cups is significantly affected by the bagasse composition ( $P \le 0.01$ ) and the amount of SF added to the bagasse  $(P \le 0.05)$ . Figure 3 shows that the B biofoam cups have lower water absorption (23%) than the DB biofoam cups (31.17%). This finding is related to the pith consistency in B (DB has no pith). Bagasse pith consists of suberin (wax) and cutin, which are water-resistant in a biofoam matrix. Suberin is a lipophilic macromolecule found in plant cell walls, and it has lipids as a hydrophobic component [18]. Cutin, which has the same structure and function as suberin, forms the cuticle skeleton [19]. Therefore, both cutin and suberin play an important role as an impermeable matter in the biofoam cups, and the B biofoam cups have lower water absorption rates than the DB biofoam cups.

Figure 4 shows that the SF1.5 biofoam cups have lower water absorption than the SF1 samples. The protein content (20%) of the SF is responsible for the water absorption of the biofoam cups. Protein consists of several amino acids, such as valine, glutamic acid, and isoleucine, which are hydrophobic; the higher the protein content, the lower the water absorption [20]. Thus, the SF1.5 biofoam cups have lower water absorption (25.83%) than the SF1 samples (28.33%).

Biofoam performance before (a) and after (b) immersion in water is seen in Figure 5. The biofoam samples show clearer bagasse sticks after water absorption than before the immersion process. Such performance of the biofoam cups is related to their water absorption ability. The DB biofoam cups have higher water absorption due to the absence of bagasse pith. During immersion, water enters the surface of the biofoam cup, thus shrinking the mycelium structure and causing bagasse sticks to appear on the specimen surface. The DB biofoams show clearer bagasse sticks than the B biofoams. Therefore, pith cells prevent water from entering the biofoam cup structure.



FIGURE 3: Effect of raw material composition on water absorption on depithed bagasse (DB) and bagasse (B) (values followed by the same letter indicate nonsignificant differences).

Another study tested biofoam made from corn husk, starch, and glycerol via a heating process, and it exhibited water absorption rates of 34.4%–35.07%, which are higher than those of our biofoam cups [21]. The differences in the composition and raw materials of the current and previous biofoams correlate with their absorption. However, commercial, synthetic Styrofoam still has a lower absorption rate (approximately 7.14%) compared with recently reported biofoams [21].

The surfaces of the SF1 and SF1.5 biofoam cups before (a) and after (b) water absorption are shown in Figure 5. The protein in SF contains a large amount of nitrogen and thus can be an energy source for forming mycelia, which are needed to bind the bagasse fibers [22]. The higher SF content of the SF1.5 biofoam cups results in thicker mycelium layers compared with that of the SF1 biofoam cups (Figure 5(a)). Consequently, the SF1.5 biofoam cups have thinner mycelium layers than the SF1 biofoam cups. This confirms that the mycelium layer contributes to water absorption in the biofoam cups.

3.2. Porosity. Porosity is the amount of free space (void) in the biofoam cup matrix. This area greatly affects the biofoam strength. Structural density prevents the entry of water and results in a compact structure of the biofoam. The ANOVA results indicate that the compositions of the raw materials (B and DB) and their interaction with the amounts of added SF (SF1 and SF1.5) have a significant effect ( $P \le 0.01$ ) on the porosity of the biofoam cups (Figure 6). The DB-SF1 biofoam cup has a higher porosity than DB-SF1.5, and their difference is significant. This is due to the amount of SF in the DB medium, which affects the growth of mycelia. Thus, the higher ratio of the amount of SF in SF1.5 is sufficient for *R. oligosporus*.

Compared with the B biofoams, the DB biofoams contain short bagasse fibers, which can form many pores in the mixed material (bagasse and SF). In B, the pith in hollow spaces causes the mixed material to appear stable and compact. During the fermentation process, mycelium growth spreads along the main structure of the bagasse. Therefore,



FIGURE 4: The effect of weight ratio of soybean flour to bagasse (SF1) and (SF1.5) on the water absorption of the biofoam cup (values followed by the same letter indicate nonsignificant differences).



FIGURE 5: The appearance of the surface of the biofoam cup with 1x magnitude before soaking (a) and after soaking (b) where DB is depithed bagasse, B is bagasse, SF1 is the ratio of soybean flour to bagasse = 1, and SF1.5 is the ratio of soybean flour to bagasse = 1.5.

the initial structure of bagasse affects the porosity of biofoam; in this case, pith plays a major role in maintaining a compact biofoam structure.

Mycelia mostly grow in the spaces in the bagasse mixture, and mycelium growth also influences the porosity of the biofoam cups. As for the ratio of the SF as a nutrient source to the tempeh mold (R. oligosporus) concentration, 65% has been reported to be the best concentration of tempeh mold [2]. In this study, we focus on the optimal mycelium growth in bagasse with and without pith and on the effect of the amount of SF on the bagasse medium. SF fills the cavities between the bagasse fibers; thus, the greater the quantity of bagasse air cavities, the better the mycelium growth. Hence, DB-SF1 has a higher total pore volume (23.08%) than DB-SF1.5 (20%) because it has more air cavities. Moreover, the SF ratios to bagasse (B-SF1 and B-SF1.5) have no significant difference (8.72%–10.77%), and below that DB medium, the same trend was observed in TBFs reinforced with wood cellulosic fibers; foam porosity decreased as the quantity of wood fibers increased [6].

*3.3. Morphology.* The cross-sectional morphologies of the biofoam cups are presented in Figure 7. B-SF1 and B-SF1.5 are smoother than DB-SF1 and DB-SF1.5. This is due to the better mycelium growth between the bagasse fibers and pith, causing fewer holes and crevices on the biofoam cups. The mold and mycelium growths form strong bonds between the bagasse fibers. Therefore, the compactness and strength of the biofoam cups. The DB biofoam cup image (red arrow) shows voids and cracks that are not filled by pith or mycelia.



FIGURE 6: The interaction effect of the bagasse composition of bagasse fiber (DB) and whole bagasse (B) and addition of soybean flour bagasse ratio 1:1 (SF1) and 1.5:1 (SF1.5) to the porosity of biofoam cup (values followed by the same letter indicate nonsignificant differences).

The amount of added SF affects the layer of mycelium growth, so SF1.5 has a smoother surface than SF1. R. oligos*porus* plays a role in producing amylase, protease, and lipase enzymes during tempeh fermentation, where these enzymes break down proteins. The mycelium growth of *R. oligosporus* can reach a height of approximately 10 mm [23]. The crosssectional morphologies of the biofoam cups are closely related to their water absorbance and porosity. The B-SF1.5 biofoam cup shows low porosity, which agrees with the sample image in Figure 7. The surface of the B-SF1.5 biofoam is shown in Figure 8. All surfaces are covered by mycelia, but B-SF1.5 exhibits the smoothest surface among all specimens. The DB samples have more indents and crevices compared with the B biofoams. The pith fills the gaps in the cavities, thus allowing the mycelia to grow easily between the bagasse fibers and evenly cover the entire biofoam cup surfaces.

Mycelium growth depends on the existence of substrates in the media. The high protein content in SF plays a role in the formation of hyphae [24]. In addition, the calcium content in SF (0.14%) is required for hyphal growth by releasing calcium-containing vesicles [25]. Usually, hyphae grow at angles of  $42^{\circ}$ – $47^{\circ}$  to the long axes of existing hyphae and form mycelium networks [26]. As seen in Figure 8(d), B-SF1.5 is dense and smooth, and the entire bagasse surface is covered by mycelia. This is because mycelia grow on both sides of, inside, and around individual particles during colony formation. These networks attach to discrete particles to form a compact composite. As a colony stretches, vegetative hyphal fusion connects discrete hyphae and forms a leaf-like lattice structure [27].

The main constituents of fungus cell walls are chitin,  $\beta$ glucan, and glycoprotein. The outer surface of a fungus's cell wall is rich in glucan, and its inside contains chitin microfibrils, which covalently form a network of cross-links with glucans [28]. This network plays a role in spreading mycelium growth, filling cavities and valley part, and covering the outer surface of the bagasse matrix. In addition to maintaining the integrity of a cell wall, chitin is responsible for the formation of a network between the cell wall and the capsule for epithelial adhesion [29, 30].

3.4. Puncture Test. The puncture strength analysis is aimed at determining the surface tension of the biofoam cups against puncture. The ANOVA findings suggest that the bagasse composition and amount of added SF have a significant effect ( $P \le 0.01$ ) on the puncture strength of the biofoam cups.

As shown in Figure 9, the DB biofoam cups have higher puncture strength  $(2.95 \text{ kg cm}^{-2})$  than the B biofoam cups  $(1.80 \text{ kg cm}^{-2})$ . The pith in the bagasse medium weakens the surface tension. We assume that the layer of bagasse fibers can promote surface strength and structural sturdiness. Thus, the pith in bagasse may create gaps, and incompatibility regions may appear between the bagasse fibers and their pith.

Interestingly, although the DB biofoams have higher porosity than the B biofoams, the DB biofoams have better surface tension than the B biofoams, which have lower porosity. This is due to the homogeneity of the bagasse material and the strong binding created by the mycelia. Figure 5 shows that DB-SF1 (a) and DB-SF1.5 (a) have thicker mycelium layers than B-SF1 (a) and B-SF1.5 (a).

The SF1.5 biofoam cup has a puncture strength of  $2.65 \text{ kg cm}^{-2}$ , which exceeds that of SF1 ( $2.1 \text{ kg cm}^{-2}$ ; Figure 10). The amount of added SF affects the puncture strength of the biofoam cup; the addition of SF enhances the structural density of the biofoam cup and reduces air; meaning the biofoam has a tighter structure [31]. With a larger amount of added SF, the mold has sufficient nutrition and can grow more robustly, leading to a thicker mycelium layer.

3.5. Compressive Strength. The compressive strength of the biofoam cups ranges from 1.74 to 3.98 MPa. The B-SF1.5 (3.98 MPa) biofoam cup has the highest compressive strength among all samples, and on average, the B biofoam cups have higher compressive strengths than the DB biofoam cups (Table 1). This is because the bagasse pith fills the hollow areas, thus promoting the compactness of the biofoam structure. This finding is in line with the porosity analysis results (Section 3.2), which indicate that the B biofoams have lower porosity than the DB biofoams. The lower porosity results in a denser matrix, as the hollow areas are filled by pith.

Apart from the presence of pith, the thickness of the mycelium layer supports the bond strength between sugarcane fibers between strands. Mycelium growth depends on the amount of substrate in the medium. The greater substrate amounts in SF1.5 accelerate the multiplication and growth of hyphae in the bagasse media. The SF1.5 biofoam cups exhibit more mycelium layers compared with the SF1 biofoam cups (Figure 8), hence strengthening the bonds between the fibers. The compressive strength or mechanical resistance of fungal composites is affected by the length of the culture period, depending on the fungal species used



(c)

(d)

 $\mu m$ 

FIGURE 7: Morphology of cross section of biofoam cup with 1600x magnitude: DB-SF1 (a), DB-SF1.5 (b), B-SF1 (c), and B-SF1.5 (d).



FIGURE 8: Morphology of surface of biofoam cup with 1600x magnitude: DB-SF1 (a), DB-SF1.5 (b), B-SF1 (c), and B-SF1.5 (d).



FIGURE 9: Effect of raw material composition of depithed bagasse (DB) and bagasse (B) on the puncture strength of biofoam cup (values followed by the same letter indicate nonsignificant differences).



Amount of soybean flour

FIGURE 10: Effect of soybean flour addition ratio to bagasse 1:1 (SF1) and 1.5:1 (SF1.5) on the puncture strength of the biofoam cup (values followed by the same letter indicate nonsignificant differences).

TABLE 1: Compressive strength of biofoam cup.

Bagasse	Soybean flour to bagasse ratio	Compressive strength (MPa)	
Depithed bagasse (DB)	1:1 (SF1)	$1.74 \pm 0.27$	
	1:1.5 (SF1.5)	$2.07\pm0.73$	
Bagasse (B)	1:1 (SF1)	3.85 ± 1.35	
	1:1.5 (SF1.5)	$3.98\pm0.39$	

[7, 22, 32]. Likewise, mycelium density, substrate type, and composite moisture can influence the mechanical properties of fungal composites [32].

A mycelium cell wall contains chitin, glucans, proteins, and lipids, whose concentrations depend on the available substrate. The outer layer consists of glucans, whereas the inner layer contains chitin microfibrils cross-linked with other polysaccharides in the form of strong covalent bonds [28]. This structure plays an important role in the formation of mycelium networks in biofoam, which affects the mechanical properties of the final material [11]. In the process of hyphal growth, the formation of branches and networks is called tip extension. In the initial phase of hyphal growth, hyphae form a network on the surface of a substrate [33] and continue to form a tubular structure [25]. The protein content in SF is important for septal formation in filamentous fungi. In this study, the formed colonies create a dense composite on the substrate surface, thus covering the entire surfaces of the bagasse fibers.

The amount of available nutrients plays an important role in the formation of mycelia, which enhances network density and strength, thereby increasing the strength of the biofoam [11, 27]. As for the B-SF1.5 biofoam cup, the low porosity and high density of the biofoam lead to an increase in the compressive strength of the sample. In a past study on TBFs reinforced with wood fibers, the biocomposite density increased the compressive strength of the TBFs; at densities of 70 and 150 g/cm<sup>3</sup>, the compressive strengths were 0.095 and 0.400 MPa, respectively [6].

3.6. Biodegradability. The biodegradability test results of biofoams with different bagasse compositions and SF ratios in the soil burial tests are shown in Figure 11. Biodegradability analysis is aimed at determining the rates of natural decomposition of the biofoam cups in soil via hydrolysis, microbial degradation, or both. After being buried in soil for 14 days, the biofoam cups exhibit weight losses ranging from 27%





FIGURE 11: Biofoam sample with 1x magnitude: before the biodegradability test (a) and after the biodegradability test (b).

Bagasse	Soybean flour	Biodegradability (%)
DB	SF1	$32.33 \pm 20.82$
	SF1.5	$27.00 \pm 21.66$
В	SF1	$56.67 \pm 13.01$
	SF1.5	$33.67 \pm 8.96$

to 56.67% (Table 2). The biodegradation of a biofoam cup is directly related to the biodegradation of its molecular chains. The B biofoam cups have higher biodegradation rates than the DB biofoam cups. This is due to the incompatibility between the bagasse fibers and pith, which makes them easy to separate and promotes microphase disintegration. This condition aids in enzymatic hydrolysis and enhances the biodegradability of the biofoam cups. The SF1 biofoam cups have higher biodegradation rates than the SF1.5 biofoam cups because they have thicker mycelium layers, which hinder water penetration and delay microorganism attacks.

*3.7. TGA.* This section discusses the thermal stability of the biofoam properties, specifically the effect of using B or DB to make the biofoam cups and the effect of various SF ratios on the thermal behavior of the cups. Figure 12 shows the



FIGURE 12: Thermogram analysis results of TGA biofoam cup bagasse, where DB is depithed bagasse, B is bagasse, SF1 is the ratio of soybean flour to bagasse 1, and SF1.5 is the ratio of soybean flour to bagasse 1.5.

TABLE 3: Thermal properties of biofoam cup.

Sample	Degradation temperature (°C)	Residue w $T_{400}$ (°C)	veight (%) T <sub>600</sub> (°C)
DB-SF1	258	35	12
DB-SF1.5	257	39	28
B-SF1	248	36	22
B-SF1.5	264	37	27

Description: DB: depithed bagasse; B: bagasse; SF1: ratio of soybean flour to bagasse 1; SF1.5: ratio of soybean flour to bagasse 1.5.

TGA and derivative thermogravimetric curves and summarizes on set temperatures of each sample in Table 3, namely,  $T_{400}$  (%) and  $T_{600}$  (%). All samples exhibit almost the same thermal degradation trend, undergoing three degradation phases.

In stage I, weight loss occurs in each sample beginning at  $30^{\circ}$ C-100°C. In this phase, an endothermic process occurs, namely, the evaporation of water vapor in the biofoam cup during heating [34]. Thermal degradation does not occur, and the weight loss is 90%–85%.

In stage II, the biofoam cups begin to decompose after the initial degradation temperature. The B-SF1.5 biofoam cup has the highest initial degradation temperature (264°C), indicating that its biofoam properties are more thermally stable than those of the other samples. The SF1.5 biofoam cups have thicker mycelium layers than the other samples. This contradicts previous studies stating that mycelia start to decompose at approximately 225°C [35] lower than cellulose-based materials, including bagasse. However, the higher the degradation temperature of a sample, the more stable it is as a heat retardant. In this stage, the glycosidic bonds in cellulose are broken, which produces  $CO_2$ ,  $H_2O$ , volatile compounds, and various hydrocarbon derivatives [17]. These results are in agreement with previous findings indicating that the thermal degradation temperature of mycelium-based biofoam made using sawdust as the fiber matrix was between 220°C and 310°C [10]. This result was associated with the cellulosic content of the sawdust; the thermal decomposition of cellulose typically starts at temperatures greater than 315°C because of the predominantly crystalline nature of cellulosic chains [36].

In stage III, residue is produced after thermal degradation at 400°C-600°C. The compounds formed during thermal degradation evaporate, resulting in a weight loss of 12%-38%, and each sample is reduced to ash.

As seen in Table 3, the DB-SF1 biofoam cup has a small amount of residue (12%) at  $T_{600}$ . This is because the largest component in bagasse is cellulose (35%). Cellulose has a high rate of decomposition, resulting in volatile components and small carbonation residue [37]. While the hemicellulose and lignin contents further increase the carbonation residue, the lignin content influences the decomposition temperature; lignin decomposes at a minimum temperature of 500°C.

#### 4. Conclusions

The B biofoam cups have a lower water absorption rate (23%) than the DB biofoam cups, with the B-SF1.5 biofoam cup having a lower water absorption rate (25.83%) than B-SF1. Both SF1 and SF1.5 have lower porosity (8.72% and 10.77%, respectively) than the DB biofoam cups. These findings agree with the morphology analysis results; the B-SF1.5 biofoam cup has a smoother biofoam surface, smaller voids, and lower porosity than the other samples. However, the DB biofoam cups have a higher puncture strength (2.95 kg cm<sup>-2</sup>) than the B biofoam cups, with the SF1.5 samples having a higher puncture strength (2.65 kg cm<sup>-2</sup>) than the SF1

biofoam cups. The samples exhibit no significant differences in the compression and degradability tests. In particular, the B-SF1.5 biofoam cup shows a higher compressive strength (3.98 MPa), but the DB-SF1.5 biofoam cup has the slowest degradation rate (27%) after 14 days of soil burial. The B-SF1.5 biofoam cup has the highest thermal stability, having a thermal degradation temperature of 264°C. Overall, the best biofoam cup is the one made from B with SF (1.5 weight ratio to bagasse), which has good thermal stability and compressive strength.

#### **Data Availability**

The data used to support the findings of this study are available from the corresponding author upon request.

#### **Conflicts of Interest**

The authors declare no conflicts of interest regarding this article.

#### Acknowledgments

The authors would like to acknowledge and thank Universitas Syiah Kuala for financing this work under the Lektor Kepala Research Scheme with grant number 191/ UN11.2.1/PT.01.03/PNBP/2022.

#### References

- E. Indarti, Marwan, R. Rohaizu, and W. D. Wanrosli, "Silylation of TEMPO oxidized nanocellulose from oil palm empty fruit bunch by 3-aminopropyltriethoxysilane," *International Journal of Biological Macromolecules*, vol. 135, pp. 106–112, 2019.
- [2] E. Indarti, S. Muliani, S. Wulya, R. Rafiqah, I. Sulaiman, and D. Yunita, "Development of environmental-friendly biofoam cup made from sugarcane bagasse and coconut fiber," *IOP Conference Series: Earth and Environmental Science*, vol. 711, article 012011, 2021.
- [3] Y. A. Yudanto and I. Pudjihastuti, "Characterization of physical and mechanical properties of Biodegradable foam from maizena flour and paper waste for Sustainable packaging material," *International Journal of Engineering Applied Sciences and Technology*, vol. 5, pp. 1–8, 2020.
- [4] C. Pamilia, M.'i. Roosdiana, S. H. Wijaya, R. A. M. Andre, and R. Robinsyah, "Pengaruh konsentrasi NaOH serta rasio serat daun nanas dan ampas tebu pada pembuatan biofoam," *Jurnal Teknik Kimia*, vol. 24, pp. 1–7, 2018.
- [5] N. Hendrawati, E. N. Dewi, and S. Santosa, "Karakterisasi biodegradable foam dari pati sagu termodifikasi dengan kitosan sebagai aditif," *Jurnal Teknik Kimia dan Lingkungan*, vol. 3, p. 47, 2019.
- [6] X. Wu, W. Yan, Y. Zhou et al., "Thermal, morphological, and mechanical characteristics of sustainable tannin bio-based foams reinforced with wood cellulosic fibers," *Industrial Crops and Products*, vol. 158, article 113029, 2020.
- [7] Z. J. Yang, F. Zhang, B. Still, M. White, and P. Amstislavski, "Physical and mechanical properties of fungal myceliumbased biofoam," *Journal of Materials in Civil Engineering*, vol. 29, no. 7, 2017.

- [8] J. Zhao, Z. Yu, H. Tian, S. Liu, and X. Luo, "Effect of bagasse content on low frequency acoustic performance of soy oilbased biodegradable foams filled with bagasse and regulation mechanism analysis," *Journal of Applied Polymer Science*, vol. 138, no. 48, article 51457, 2021.
- [9] W. Sanhawong, P. Banhalee, S. Boonsang, and S. Kaewpirom, "Effect of concentrated natural rubber latex on the properties and degradation behavior of cotton-fiber-reinforced cassava starch biofoam," *Industrial Crops and Products*, vol. 108, pp. 756–766, 2017.
- [10] C. Bruscato, E. Malvessi, R. N. Brandalise, and M. Camassola, "High performance of macrofungi in the production of mycelium-based biofoams using sawdust — sustainable technology for waste reduction," *Journal of Cleaner Production*, vol. 234, pp. 225–232, 2019.
- [11] S. Manan, M. W. Ullah, M. Ul-Islam, O. M. Atta, and G. Yang, "Synthesis and applications of fungal mycelium-based advanced functional materials," *Journal of Bioresources and Bioproducts*, vol. 6, no. 1, pp. 1–10, 2021.
- [12] J. Yang, Y. C. Ching, and C. H. Chuah, "Applications of lignocellulosic fibers and lignin in bioplastics: a review," *Polymers*, vol. 11, p. 751, 2019.
- [13] Y. H. Arifin and Y. Yusuf, "Mycelium fibers as new resource for environmental sustainability," *Procedia Engineering*, vol. 53, pp. 504–508, 2013.
- [14] A. Wahyudi, "Pengaruh penambahan arenga saccharifera terhadap kualitas produk sabun transparan," *Jurnal Redoks*, vol. 3, p. 30, 2018.
- [15] S. Saiful, N. Nurfitriana, M. Ramli, and I. Maulana, "Pengembangan membran magnesol untuk pemurnian biodiesel," *Jurnal Rekayasa Kimia & Lingkungan*, vol. 9, pp. 118–125, 2013.
- [16] J. H. Etikaningrum, E. S. Iriani, R. Syarief, and A. W. Permana, "Pengaruh penambahan berbagai modifikasi serat tandan kosong sawit pada sifat fungsional biodegradable foam," *Jurnal Penelitian Pascapanen Pertanian*, vol. 13, pp. 146–155, 2016.
- [17] E. Indarti and W. D. Wanrosli, "Thermal stability of oil palm empty fruit bunch (OPEFB) nanocrystalline cellulose: effects of post-treatment of oven drying and solvent exchange techniques012025," *Journal of Physics Conference Series*, vol. 622, 2015.
- [18] J. Graça, "Suberin: the biopolyester at the frontier of plants," *Frontiers in Chemistry*, vol. 3, article 00062, 2015.
- [19] Y. Li-Beisson, "Cutin and Suberin," in *Encyclopedia of Life Sciences*, vol. 2011, John Wiley & Sons, Ltd.
- [20] Y. W. Widyarani, E. Sari, J. P. M. Ratnaningsih, M. E. Sanders, and M. E. Bruins, "Production of hydrophobic amino acids from biobased resources: wheat gluten and rubber seed proteins," *Applied Microbiology and Biotechnology*, vol. 100, no. 18, pp. 7909–7920, 2016.
- [21] S. S. Gani and H. Kusumayanti, "The optimization of additional of glycerol on the biodegradable foam from corn husk," *Journal of Vocational Studies on Applied Research*, vol. 4, pp. 18–26, 2022.
- [22] M. Haneef, L. Caseracciu, C. Canale, I. S. Bayer, J. A. H. Guerrero, and A. Athanassiou, "Advanced materials from fungal mycelium: fabrication and tuning of physical properties," *Scientific Reports*, vol. 7, no. 1, article 41292, 2017.
- [23] D. A. Mitchell, H. W. Doelle, and P. F. Greenfield, "Agar plate growth studies of *Rhizopus oligosporus* and *Aspergillus oryzae* to determine their suitability for solid-state fermentation,"

Applied Microbiology and Biotechnology, vol. 28, no. 6, pp. 598–602, 1988.

- [24] M. Raudaskoski, "The central role of septa in the basidiomycete Schizophyllum commune hyphal morphogenesis," *Fungal Biology*, vol. 123, no. 9, pp. 638–649, 2019.
- [25] N. N. Levina and R. R. Lew, "The role of tip-localized mitochondria in hyphal growth," *Fungal Genetics and Biology*, vol. 43, no. 2, pp. 65–74, 2006.
- [26] N. P. Money, The Fungi: Third Edition, Elsevier Ltd., 2016, 00002-5.
- [27] H. Du and P. Perré, "A novel lattice-based model for investigating three-dimensional fungal growth on solid media," *Phy*sica A: Statistical Mechanics and its Applications, vol. 541, article 123536, 2020.
- [28] J. R. Herrera and L. O. Castellanos, "Cell wall glucans of fungi. A review," *The Cell Surface*, vol. 5, article 100022, 2019.
- [29] D. L. Goldman and A. G. Vicencio, "The chitin connection," *MBio*, vol. 3, article 100022, 2021.
- [30] S. J. Free, Advances in Genetics, vol. 81, Elsevier Inc., 2013, 00002-6.
- [31] W. Fitri and M. Mora, "Pengaruh persentase serbuk ampas tebu terhadap sifat fisik dan mekanik papan semen partikel," *Jurnal Fisika Unand*, vol. 7, no. 4, pp. 367–373, 2018.
- [32] J. L. Teixeira, M. P. Matos, B. L. Nascimento, S. Griza, F. S. R. Holanda, and R. H. Marino, "Production and mechanical evaluation of biodegradable composites by white rot fungi," *Ciência e Agrotecnologia*, vol. 42, no. 6, pp. 676–684, 2018.
- [33] M. Nopharatana, D. A. Mitchell, and T. Howes, "Use of confocal scanning laser microscopy to measure the concentrations of aerial and penetrative hyphae during growth of Rhizopus oligosporus on a solid surface," *Biotechnology and Bioengineering*, vol. 84, no. 1, pp. 71–77, 2003.
- [34] D. Salbeti and I. S. Harlia, "Sintesis dan karakterisasi termal plastik ramah lingkungan polyblend selulosa ampas tebu dan limbah botol plastik polietilen tereftalat," *Jurnal Kita Khatulistiwa*, vol. 7, p. 60, 2018.
- [35] M. Jones, T. Bhat, T. Huynh et al., "Waste-derived low-cost mycelium composite construction materials with improved fire safety," *Fire and Materials*, vol. 42, no. 7, pp. 816–825, 2018.
- [36] A. Edhirej, S. M. Sapuan, M. Jawaid, and N. I. Zahari, "Cassava/sugar palm fiber reinforced cassava starch hybrid composites: physical, thermal and structural properties," *International Journal of Biological Macromolecules*, vol. 101, pp. 75–83, 2017.
- [37] A. D. Hidayati, S. Kurniawan, N. W. Restu, and B. Ismuyanto, "Potential of sugar cane as an alternative raw material for making activated carbons," *Natural B, Journal of Health and Environmental Sciences*, vol. 3, no. 4, pp. 318–322, 2016.