ISSN: 1410-8917
e-ISSN: 2597-9914

Jurnal Kimia Sains dan Aplikasi 25 (3) (2022): 130–136

Jurnal Kimia Sains dan Aplikasi
Journal of Scientific and Applied Chemistry

Journal homepage: http://ejournal.undip.ac.id/index.php/ksa

Effects of Additional Polyvinyl Alcohol (PVA) on the Physiochemical Properties of Chitosan-Glutaraldehyde-Gelatine Bioplastic

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https://doi.org/10.14710/jksa.25.3.130-136

Article Info

Article history:

Received: 27th December 2021
Revised: 11th March 2022
Accepted: 23rd March 2022
Online: 31st March 2022

Keywords:
PVA; bioplastic; physicochemical; biodegradable; antibacterial

Abstract

This study investigated the effects of additional PVA on the physicochemical properties of the chitosan-glutaraldehyde/gelatin bioplastic composite. The best results of the bioplastic film were obtained at a concentration of 3% PVA, with a tensile strength value of 3.3 MPa, flexibility reached 54%, a thickness value of 0.24 mm, percentage of inhibition against *E. coli* and *S. aureus* was 21.8% and 8.8% respectively. The FTIR spectrum results showed no change in the wavenumber of the chitosan and gelatin chitosan spectrum with OH, CO, and NH functional groups. The spectrum indicates that only physical interactions occurred. The bioplastics are similar in thermal stability and have slight differences in bioplastic morphological contours. The average thickness of the bioplastics is between 0.20–0.26 mm. Based on the Japanese Industrial Standard (JIS), all bioplastics meet the standard thickness, which is < 0.25 mm, excluding chitosan, which has a thickness of 0.26 mm. The addition of PVA into the bioplastics structure increased the hydrophobicity, pH resistance, and flexibility of bioplastics. Meanwhile, additional PVA decreased biodegradability, only degraded by 60% at eight weeks. Based on these data, not all bioplastics can meet the degradation time criteria set by the international bioplastic standard ASTM D-6002, that bioplastics must be 100% degraded within eight weeks. Bioplastics made from chitosan and chitosan-gelatin have been degraded by 90% for 48 weeks. Based on the antibacterial properties, the inclusion of PVA into the bioplastic structure enhances the antibacterial properties.

1. Introduction

Nowadays, various materials are used in food packagings, such as paper, fiberboard, glass, tinplate, aluminum, and different types of plastics. The intensity of the use of plastic materials increases, along with the advantages of plastic compared to other packaging materials. Plastic is light, flexible, strong, not easy to break, transparent, waterproof, and practical. On the other hand, the increasingly massive use of plastic causes various global problems. Non-biodegradable plastic or synthetic (inorganic) plastic is a polymer that is not easily degraded by microbes in the soil. The reason is that the main ingredients are derived from refined petroleum products (polyolefins) through a series of polymerization processes (combination) of ethylene and propylene with powerful chemical bonds [1]. The inability of microbes to degrade plastic materials creates a new problem in the form of environmental pollution. Efforts to handle plastic waste have been carried out by recycling, reusing, reducing use, and using renewable natural resources. However, this method requires high cost, limited use, and a decrease in the quality of the resulting product [2].

A new concept being developed is to make biodegradable plastic (bioplastic). Bioplastics are environmentally friendly plastics with properties and flexibility similar to conventional plastics but are easily decomposed into biomass in the soil by microorganisms [3]. Some materials that have been developed are chitosan, gelatin, starch, and other polymers derived from plant and animal cells [4]. Diversification is required

to make bioplastic materials more developed and readily available in the surrounding environment.

Chitosan (Cs) is a natural polymer that is regenerative, biodegradable, non-toxic, biocompatible, antimicrobial, and quickly swells to form thin bioplastics. Chitosan is a polymer derived from chitin with a molecular weight between 100–1,000 kDa (kg/mol), which can be obtained from the shells of shrimp, crabs, and flower crabs. Chitosan has several advantages, such as hemostatic, fungistatic, biocompatible, having two easily modified groups, having no side effects, and being easily decomposed by microbes. However, chitosan is insoluble in water, dense, slightly brittle, has weak mechanical strength, hence easily damaged [5]. Chitosan needs to combine with materials such as biopolymers or synthetic polymers to produce bioplastics with the desired properties.

The use of chitosan as a plastic base material has been widely studied. It is reported that chitosan is excellent as a plastic base material though slightly brittle [6]. As a raw material for bioplastics, chitosan is rigid and fragile. Combined material is required to reduce these properties. Chitosan modification using chlorella increased tensile strength by 235%, increased degradation by 50%, and reduced water vapor permeability above 60% [6]. Chitosan-polyethylene (LDPE) composite produces bioplastics with better tensile strength and stretch. Water absorption, thermal degradation, increased, and the biodegradability of the mixture increased one hundred times compared to plastic made from pure LDPE [7]. The physicochemical properties of the chitosan-antioxidant composite were investigated by [8], and it was found that there was an increase in water absorption, solubility, and decreased swelling. A composite of chitosan/gelatin/chitosan modified silver nanoparticles (AgNP) grafted with catechol was increased plastic materials' tensile strength and water vapor resistance [9].

To increase the elasticity of the bioplastic, the researchers added other biomaterials such as gelatin, alginate [10], polyvinyl alcohol, and nano-hydroxyapatite [11]. Gelatin/chitosan composite, as has been done by [12, 13, 14, 15], produces bioplastics that are hydrophobic on the surface, have high mechanical strength, and thermal stability. A review of the degradability of water hyacinth PVA/gelatin-cellulose nanocomposite has also been studied [16]. Gelatin is translucent, transparent and non-toxic, odorless, solid, and can form a gel [17]. These properties are required to make plastic sheets with good physical characteristics. Combining chitosan with gelatin will make the resulting bioplastic more elastic and easier to shape in various forms of packaging.

Besides gelatin, polyvinyl alcohol (PVA) is also widely used as a mixture to manufacture plastics that have powerful properties. PVA is a synthetic polymer that is degradable, hydrophilic, forms bioplastics well, dissolves in water, is non-toxic, and has mechanical strength [18]. In many cases, PVA is always added to increase the film's elasticity because of its high mechanical strength [5]. The addition of PVA in making starch/chitosan composites has been investigated [19]. The resulting plastic has better mechanical strength, is non-porous, smooth, bright, and degrades up to 60% after 90 days [19]. Variations in the addition of gelatin to the chitosan/PVA composite were able to restrict water vapor permeability, reduce water solubility by up to 23% and increase mechanical strength. Therefore, the plastic obtained was following plastic properties, which are not hygroscopic and strong [20, 21]. A crosslinker is usually added to enhance the reaction between materials in bioplastic composites. The use of glutaraldehyde as a crosslinker has been widely used in chitosan [22, 23], chitosan and alginate [24], gelatin [25], and PVA/PPDOTS [26].

Glutaraldehyde has an aldehyde group at both ends of its structure, which can bind two different materials to form the desired material [19]. In addition, it was also reported that glutaraldehyde could reduce the degree of film swelling but at the same time increase its brittleness. The release of glutaraldehyde from the film to the simulant solution was not observed after incubation. The biodegradation test observed that although crosslinking did not prevent the degradability of the film, the film would still be degraded even though it took a long time. The qualitative analysis of the ecotoxicity of the film indicated the possibility of composting the developed film. Films present the potential for application as membranes and in packaging, and different formulations can be used according to the desired final characteristics. As an environmentally friendly synthetic polymer, modification with PVA on various bioplastic materials and increasing shelf life are also expected to increase the elasticity of bioplastics.

Therefore, this study aimed to examine the effect of the addition of PVA at various concentrations on the physical strength of the chitosan-gelatin-based bioplastics produced. In this study, three concentrations of added PVA were used. It is estimated that PVA, with its good properties, will provide added value to the bioplastics made.

2. Methodology

2.1. Materials

The materials used in this study were all pro-analytical, including: chitosan (Biotech Surindo Cirebon, molecular weight (MW) = 40,000 g/mol), fish gelatin (Buana Chem, MW = 100–150 kg/mol), PVA (Merck, MW = 72,000 g/mol), glutaraldehyde (Merck), NaOH (Merck, MW = 40 g/mol), acetic acid (Merck, MW = 60,052 g/mol), yeast extract (Merck), sodium agar (Merck), Peptone Bacto (Nitrakimia), HCl (Merck), bacteria *E. coli* and *S. aureus* obtained from Merck, distilled water was purchased from UPT Laboratorium Terpadu Undip.

2.2. Synthesis and Characterization

The manufacture of bioplastics was based on the phase inversion process, with the composition ratio of each material as shown in Table 1, with reaction conditions at pH 6–6.5, reaction temperature 45–50°C, and stirring time of 45 minutes [5]. In the initial stage,

1.5% gelatin solution (G) was made and followed by a crosslink reaction between gelatin and glutaraldehyde, with a mole ratio of 1:1 to produce a GG solution. Subsequently, prepare 1.5% chitosan solution in 2% acetic acid (Cs) and PVA 1.5% solution in warm water (P). After that, all the solutions were mixed and stirred according to the compositions written in Table 1 for 45 minutes. The solution mixture was put into an ultrasound bath at 50℃ for 10 minutes to complete the reaction. The mixture was allowed to cool and then poured into a petri dish to form a thin bioplastic and dried in an oven at 45℃ for 24 hours. The dried bioplastics were washed with distilled water, air-dried, and stored in dry containers. Furthermore, the bioplastics were characterized for their physicochemical properties, including the following tests: functional group (FTIR), surface morphology (SEM), thermal resistance (TGA), thickness, hydrophilicity, resistance to pH, tensile strength and percentage of elongation, biodegradability, and antibacterial using the turbidimetric method.

Table 1. Bioplastic Composition Ratio

Bioplastic	Cs (%)	G (%)	GG (%)	PVA (%)		
				1.5	3	4.5
C	100	-	-	-	-	-
CG	50	50	-	-	-	-
CGGP1.5	40	-	40	20	-	-
CGGP3	40	-	40	-	20	-
CGGP4.5	40	-	40	-	-	20

chitosan (Cs), gelatin (G), gelatin-glutaraldehyde (GG), polyvinyl alcohol (PVA)

3. Results and Discussion

3.1. Chemical Properties of Bioplastics

The resulting bioplastics are transparent, elastic, thin, hard to tear, and resemble plastic in a dry state, as shown in Figure 1. Almost all the bioplastics produced have the same physical appearance.

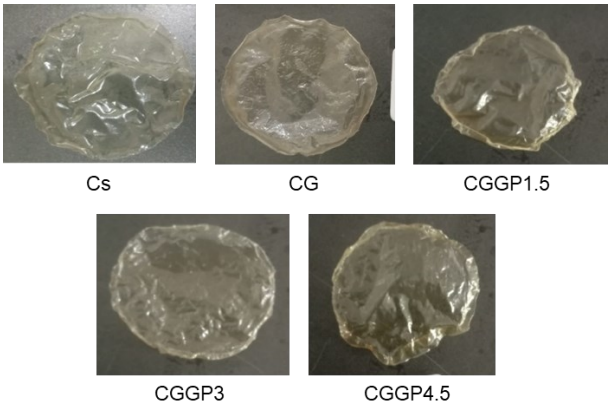

Figure 1. Bioplastics

Furthermore, the synthesized bioplastics were analyzed for their functional groups using FTIR to determine the success of the reaction (Figure 2). The functional group spectra show similarities (almost 90%) in the pure Cs and pure chitosan-gelatin (CG) spectra. This similarity is possible because both chitosan and gelatin are derived from natural polymers with similar functional groups, namely -OH, -CH, and -NH. Possible reactions between chitosan and gelatin are physical interactions such as hydrogen bonds. Spectral differences began to be seen in chitosan modified with glutaraldehyde and PVA. The wavenumber for the stretching vibration of the CO group appears to shift to a higher wavenumber. This shift in wavenumber is probably due to the crosslinks formed between gelatin-glutaraldehyde (GG) and chitosan-gelatin-glutaraldehyde (CGG). This crosslinking causes the movement of molecules to be more restricted and requires more energy to vibrate. This wavelength shifts following the energy rule, where the higher the vibration wavenumber of a functional group, the greater the energy produced/required (E=h.υ). The crosslinking between chitosan and glutaraldehyde should have appeared at a new peak at 1654.10 cm^{-1}, namely the C=N bond. Surprisingly, in the resulting spectra, chitosan also shows the same absorption in that area, and the C=N absorption becomes challenging to identify. The possibility is that only a few crosslinks formed between chitosan-glutaraldehyde.

The reaction between CG and PVA did not generate new groups because the additional PVA was only formed through hydrogen bonds [5]. The spectra of the composite only experienced a shift in the absorption region. Based on the spectra results, it can be concluded that four different materials produced the four FTIR spectra.

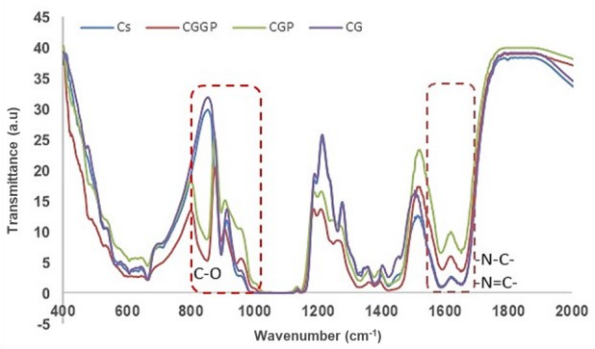

Figure 2. FTIR Spectra of Cs (chitosan), CGGP (chitosan-glutaraldehyde-gelatin/PVA), CGP (chitosan/gelatin/PVA), and CG (chitosan/gelatin)

To further clarify FTIR results, a thermal analysis was conducted using TG/DTA to determine the influence of temperature on the substance stability. The results of thermal stability characterization are presented in Figure 3. Based on TGA and DTG, there are two stages of weight loss. The first stage occurred at around 70–150℃, related to water evaporation in the bioplastic. At this stage, the weight loss of chitosan was 17%, while the other two bioplastics were nearly the same at 13%. This evaporation does not occur simultaneously but in the range of time and temperature. The water is bound to the surface until the water in the pore lattice in the bioplastic evaporates slowly. The evaporation is endothermic, as indicated by the DTG thermogram. Based on the weight loss results, chitosan had a higher water content than GG, and GGP modified chitosan.

The second stage occurred at temperatures between 240–500℃, with a weight loss of 55%. This significant

weight reduction is probably not covered by the evaporation of CO, CO_2, and NH_3 gases due to depolymerization. This causes bond breaking and ring opening due to continuous heating to produce smaller fractions released as a gas until the final compound is formed, namely carbon (C). This weight loss is exothermic by releasing heat. These results are in agreement with studies [27, 28, 29, 30].

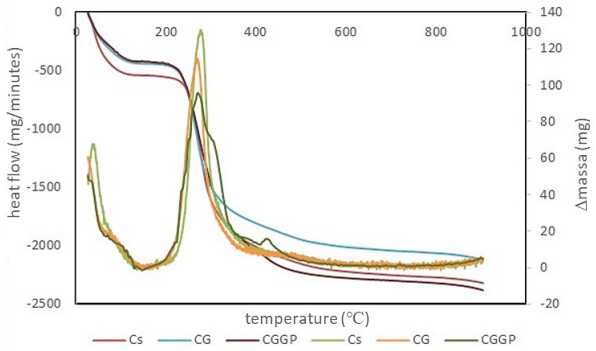

Figure 3. TGA/DTG chromatogram of Cs, CG, CGGP bioplastics

The morphological characterization of the side of the bioplastic (cross-section) was performed. The SEM images (Figure 4) showed that the surface morphology of bioplastics, both pure Cs and CG bioplastics, were smooth and not contoured (a and b). A slight difference was obtained from image c; a bioplastic from CGGP had a rough surface. There were several voids in bioplastics. This result followed the results obtained by a previous study [31], which stated that the addition of glutaraldehyde and PVA effectively made the composite coarser and revealed clearer voids formed as carried out by [31].

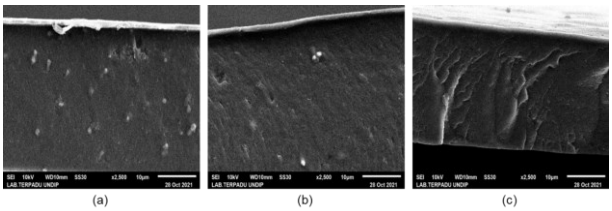

Figure 4. Cross-sectional sections of (a) Cs (pure chitosan), (b) CG (chitosan/gelatin), and (c) CGGP (chitosan-glutaraldehyde-gelatin/PVA3)

3.2. Physical Properties of Bioplastics

According to the Japanese Industrial Standard (JIS), the thickness of the bioplastic produced must be below the maximum standard for the thickness of the bioplastic packaging, which is 0.25 mm. If the bioplastic exceeds the maximum thickness limit, it will affect the packaged material by changing the aroma and taste. Based on Table 2, the addition of glutaraldehyde and PVA significantly reduced the thickness of the bioplastic, unlike Cs and CG, which did not reduce significantly. The results of this thickness test follow the study by [27]. Based on JIS standards, all bioplastics meet the thickness standard, which is < 0.25 mm.

The hydrophobicity of plastic is also a necessary standard, and this is related to the use of bioplastics as packaging containers. Hydrophobicity will make the packaged material is not easily damaged and does not change the aroma and taste of the material. Based on SNI standard plastic criteria, the hydrophilicity of bioplastics should not exceed 1%. The hydrophilicity test was carried out using the sessile drops method in this study. The liquid was dripped on the surface of the bioplastic, waited for a few seconds, and measured the contact angle. All bioplastics generally have hydrophilic properties, as seen from the small contact angle. A material is called hydrophilic if the contact angle is < 90°. The addition of glutaraldehyde and PVA increased the contact angle value, causing the bioplastic to be more hydrophobic than Cs bioplastic. From the results in Table 2, the bioplastics produced generally did not meet the standard criteria for hydrophobic values from SNI.

Bioplastics must possess tensile strength and flexibility because these two properties indicate the ability of bioplastics to be produced in various forms of packaging. The tensile strength test method was performed in a wet state. The sample was immersed in distilled water for ± 1 hour, then, with particular energy, was pulled with a load considering the breaking ability of the amount of energy offered. Chitosan modification using gelatin reduced the flexibility of bioplastics (Table 2). This result is related to the highly hydrophilic character of gelatin. Bioplastic absorbs water more easily due to its hydrophilic structure, causing the bioplastic to swell. The continuously applied force makes the bioplastic unable to withstand the force to break. Modifications using glutaraldehyde and PVA at various concentrations produced more vigorous and more flexible bioplastics than Cs and CG bioplastics. The results of this study were in line with research conducted by [27]. Based on the test results, it was concluded that the tensile strength of all bioplastics did not meet Indonesian National Standard (SNI). According to SNI, the tensile strength and elongation values are 24.7–302 MPa and 21–24, respectively, and the result did not meet the standard. Based on SNI data, bioplastic for packaging material requires the flexibility to be easily assembled in various shapes [27].

Table 2. Thickness, Contact Angle and Mechanical Strength Measurement Results of Bioplastics

Bioplastic	Thickness (mm)	Contact angle (°)	Tensile strength (MPa)	Elongation (%)
C	0.26	45	2.041	21.349
CG	0.24	38	2.806	16.907
CGGP1.5	0.18	53	2.829	30.419
CGGP3	0.24	40	3.300	54.772
CGGP4.5	0.24	40	2.214	37.050

The stability of bioplastics over a wide pH range indicates chemical resistance. In other words, bioplastics can be utilized in various pH conditions. The pH resistance test was conducted by measured bioplastic mass before and after immersion in a solution of pH 3, 5, 7, 9, and 11. A significant decrease in the mass of bioplastics indicated the instability of bioplastics at certain pH conditions, which could be caused by the decomposition of the components of bioplastics under

acidic or alkaline conditions. Based on Table 3, at pH 3, all bioplastics experienced a very significant decrease/damage. At pH 5–11, the mass of all types of bioplastics did not reduce. It can be concluded that all bioplastics can be used from a pH range of 5–11 without being damaged [27].

Table 3. Average Percentage of Mass Reduction of Bioplastics at Various pH Conditions

Type of Bioplastic	Mass Reduction (%)				
	pH				
	3	5	7	9	11
Cs	15	3	0	0	0
CG	47	9	0	0	0
CGGP1.5	100	3.0	0	0	0
CGGP3	100	2.8	0	0	0
CGGP4.5	100	2.6	0	0	0

3.3. Biodegradability of Bioplastics

Biodegradability testing of bioplastics aimed to determine the degree of damage to bioplastics due to the influence of microorganisms in the soil. This test used the soil burial test—burying bioplastics into the soil.

In the FTIR test, the absorption of 1079 cm^{-1} is a particular region of C–O ester. The presence of an ester group indicates the ability of bioplastics to decompose. Due to hydrophilic C–O esters, water molecules can cause microorganisms in the soil to enter the bioplastic matrix and damage or degrade the bioplastic. The measurement of bioplastic degradation results can be seen in Figure 5. Based on these data, it can be seen that the degradation rate increases in plastics with a CG composition. The increase in gelatin mass is in line with the degradation rate by biomass in the soil. Based on the mechanism of degradation in the soil, it was argued that the more hydrophilic material is, the easier it will be for microbes to enter the material because water is an appropriate medium for the entry of microbes into the material [28]. In the gelatin, PVA and glutaraldehyde composite experience a slow degradation rate. This is because the entry of synthetic PVA elements into the bioplastic structure causes the biomass to require a slightly longer to recognize the elements in the material. Hence, it decelerates the degradation process. Based on the data above, not all membranes follow the degradation time criteria of international bioplastic standard ASTM D-6002, namely that bioplastics must be 100% degraded within eight weeks.

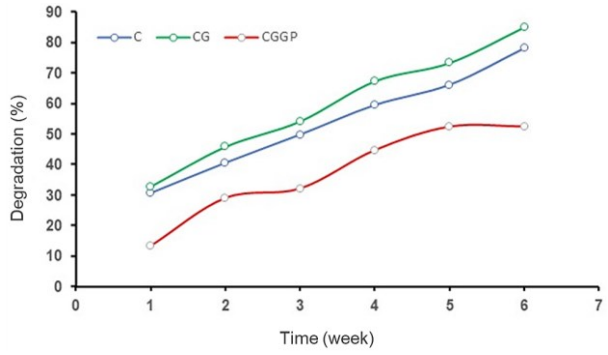

Figure 5. Average time of degradation by bacteria in soil

3.4. Antibacterial Activity

Antibacterial activity testing aimed to evaluate the level of inhibition of *E. coli* and *S. aureus* growth in bioplastics using the turbidimetric method. This turbidimetric method is the same as that conducted by [29]. The test results are presented in Table 4.

Table 4. Antibacterial activity by turbidimetric method for 12 hours

Type of bioplastic	*E. coli*		*S. aureus*	
	Inhibition (%)	Bacterial Count ($\times 10^8$ CFU/mL)	Inhibition (%)	Bacterial Count ($\times 10^8$ CFU/mL)
C	14.199	1.287	2.632	1.461
CG	20.510	1.192	6.184	1.407
CGGP1.5	20.601	1.180	7.005	1.387
CGGP3	21.845	1.172	8.816	1.368
CGGP4.5	21.000	1.182	7.100	1.398
Ciprofloxaxin (+)	96.602	0.051	94.605	0.081

The active group $-NH_3^+$ in bioplastics has the opportunity to inhibit the growth of bacteria because this group will bind to the negatively charged groups in bacteria. This is evidenced by calculating the percentage of inhibition of these bacteria. The percentage of inhibition obtained on C bioplastic was 14.199% in *E. coli* and 2.632% in *S. aureus*. The inhibition percentage of CG bioplastic was 20.510% in *E. coli* and 6.184% in *S. aureus*, and bioplastics varied from 20.6–21.8% for *E. coli* and 7–8.8% for *S. aureus*. In comparison, the addition of PVA3 had the highest inhibitory power of 21.845% on *E. coli* bacteria and 8.816% on *S. aureus*. The highest inhibition with the addition of PVA 3 was possibly due to the increasing positively charged functional groups in bioplastics that could bind gram-positive and gram-negative bacteria. Research indicating the presence of antibacterial properties in PVA material is in line with research conducted by [31]. It was reported that the addition of PVA into a thin film increased the antibacterial ability.

4. Conclusion

Chitosan/gelatin-glutaraldehyde/PVA composite has been successfully synthesized and made into thin plastic bioplastics. In general, the resulting plastic was transparent and not easily torn. The best results of the bioplastic film were obtained at a concentration of 3% PVA, with a tensile strength value of 3.3 MPa, the flexibility of 54%, a thickness value of 0.24 mm, percentage inhibition against *E. coli* and *S. aureus* of 21.8% and 8.8%, respectively. The resulting bioplastics were similar in thermal stability and had slight differences in bioplastic morphological contours. The average thickness of the resulting bioplastics was between 0.20 and 0.26 mm. The addition of PVA increased the hydrophobicity, pH resistance, and flexibility of the bioplastic. Meanwhile, biodegradability decreased with the addition of PVA, which was only 60% degraded at eight weeks. Based on the antibacterial properties, the addition of PVA into the bioplastic structure enhanced the antibacterial properties.

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