Science and Technology Indonesia
e-ISSN:2580-4391 p-ISSN:2580-4405
Vol. 8, No. 2, April 2023

Research Paper

The Characterization of the Hydrophobic Surface of Nanocomposites *Aloe vera*/PS for Antimicrobial Pathogens

Lathifa Zonesya Putri[1], Ratnawulan Ratnawulan[1]*, Ramli Ramli[1], Hamdi Rifai[1], Faridah Lisa Supian[2], Irna Humairah[3]

[1]*Departement of Physics, Universitas Negeri Padang, Padang, 25132, Indonesia*
[2]*Departement of Physics, Sultan Idris Education University, Tanjung Malim, Perak Darul Ridzuan, 35900, Malaysia*
[3]*Laboratorium Lembaga Layanan Pendidikan Tinggi Wilayah X, Sumatera Barat, 25173, Indonesia*
***Corresponding author**: ratnawulan@fmipa.unp.ac.id

Abstract
The spread of pathogenic microorganisms on a large scale creates a health disaster for the world. Modifying the surface of the fabric so that it is antimicrobial pathogens with hydrophobic properties is one solution to inhibit the spread of microorganisms. Surface modification was carried out using *Aloe vera* powder as filler and polystyrene matrix with *Aloe vera*:PS composition variations of 1:5, 2:5, and 3:5 using the dip coating method and drying temperatures of 10°C, 50°C, and 90°C. The characterization results that have been carried out with the best composition variations are 3:5 and 50°C with a layered morphology using Scanning Electron Microscopy (SEM), resulting in a surface particle size of 21.243 nm, large contact angle/WCA using a Canon D350 camera with a size of 150.23°, Fourier Transmission Infrared Analysis (FTIR) with the dominant functional group in the vibration band 2629 cm^{-1} indicates the presence of a long hydroxyl chain, the vibration band 1500 cm^{-1} with C-H stretching of the aromatic ring is caused by the characteristics of the polystyrene polymer and the wave number is 1700 cm^{-1} with stretching C=O which shows the characteristics of the carbonyl group in the *Aloe vera* sample is a flavonoid compound. The resistance of *Aloe vera*/PS solution to Klebsiella pneumonia resulted in a 20.18 mm diameter barrier showing strong resistance against bacteria and X-Ray Diffraction analysis showed that the AV powder was amorphous. Thus, the AV/PS 3:5 composition with a calcination temperature of 50°C represents the greatest variation in the synthesis and characterization of hydrophobic thin films for pathogenic antimicrobial organisms.

Keywords
Aloe vera, Composition, Hydrophobic, Anti-Microorganisms, Polystyrene

Received: 1 December 2022, Accepted: 26 February 2023
https://doi.org/10.26554/sti.2023.8.2.212-218

1. INTRODUCTION

Aloe vera is a herbal plant that contains active compounds, including polysaccharides, vitamins, minerals, amino acids, denolic compounds, organic acids, acemannan, and saponins (Heng et al., 2018; Rani et al., 2021). The structures of *Aloe vera* compounds were identified as Anthrone-C-Glucosyl, Barbaloin (Aloin A), and Homonataloin. Structure of aloesin, barbaloin and isobarbaloin (aloin B), aloenin, 2'-O-feruloylaloesin, aloe-emodin, aloeresin A (p-coumaric acid ester of aloesin), 8-C-Glucosyl -7-O-methyl-(S)-aloesol, isoaloeresin D and aloeresin E (Ibrahim et al., 2018). The content of *Aloe vera* compounds has the ability to clean and is antiseptic as well as antiviral, antibacterial, and antifungal (El Fawal et al., 2019; Nizam et al., 2021; Mpiana et al., 2020).

Hydrophobic thin films sourced from *Aloe vera* are one of the efforts to produce coating materials for anti-pathogenic microorganisms by adding polystyrene polymers as a matrix (reinforcer) (Seifunnisha et al., 2021). The hydrophobic coating has the characteristics of being effective in self-cleaning and having a contact angle of >90° (Ratnawulan et al., 2020; Fauzi, 2019; Fauzi and Putri, 2020). *Aloe vera* leaves have a gel that can be prepared into flour/powder to have better physical properties and be more resistant to damage (Maan et al., 2021; Wang and Dai, 2016; Hendrawati, 2015). *Aloe vera* powder or flour can be prepared using the foam mat drying method, which is a drying process by making foam from liquid material added to the foam stabilizer by drying at 60°C-75°C using an oven for 6 hours, this method is proven to be simpler and cheaper (Javed et al., 2018; Varhan et al., 2019; Hardy and Jideani, 2017; Mounir, 2017).

Based on previous research, a layer of *Aloe vera*/Polystyrene (AV/PS) has been successfully prepared by producing a diameter of 113 mm inhibition area for pathogenic microorganisms and a contact angle (WCA) of 112° applied to cotton using the dip coating method (Seifunnisha et al., 2021; Dastan et al., 2016; Tang and Yan, 2017; Ho et al., 2018). However, this study has a drawback where the value of the contact angle can still be increased. One effort that can be done is to conduct research on the effect of composition variations on AV/PS hydrophobic thin films for pathogenic antimicrobials with variations of 1:5, 2:5, and 3:5. It has been studied that the composition of AV/PS 3:5 has the best coating characteristics where hydrophobic properties have a contact angle of 150.23° and anti-pathogenic properties have an inhibition area diameter of 20.18 mm.

2. EXPERIMENTAL SECTION

2.1 Materials

The materials used in this study were *Aloe vera* taken from the surrounding area (Tunggul Hitam, Padang, Indonesia) and polystyrene (Merck Indonesia, Jakarta, Indonesia). Other ingredients used are aquadest (CV. Novalindo, Padang, Indonesia), tween 80, maltodextrin and toluene (CV. Nitrakimia, Bantul, Indonesia).

2.2 Methods

Starting with washing the *Aloe vera* leaves using aquadest, then separate the gel from the skin. The separated gel was prepared using the foam-mat drying method by adding 0.3% tween 80 and 15% maltodextrin. The dried gel was ground using a mortar and filtered using a 125 μm mesh. Finally, the fine powder was refined again using the High Energy Milling E3D (HEM-E3D) tool to obtain nanoparticle sizes. Next, AV/PS solution was made using the sol-gel method. First, weigh *Aloe vera* and polystyrene with a composition of 1:5, 2:5, and 3:5, measuring 2 mL of toluene. Second, stir the *Aloe vera* with 1 mL of toluene for 4 hours using a magnetic stirrer at a speed of 1000 rpm; in a separate glass stir polystyrene by adding 1 mL of toluene using a magnetic stirrer at a temperature of 60°C and a speed of 1000 rpm. Third, drip the *Aloe vera* solution into the polystyrene solution while stirring at 250 rpm for 1 hour. Finally, the AV/PS solution was allowed to stand for 24 hours to obtain a homogeneous solution. Then, the homogeneous AV/PS solution was coated on a 2×2 cm cloth using the dip coating method, and the thin layer that had been formed was calcined using an oven with a temperature of 50°C for 30 minutes. The calcined fabrics are ready to be characterized using FTIR, SEM, and sessile drop to determine the contact angle. Last, the prepared AV/PS solution was tested for the activity of microorganisms using gram-negative Klebsiella pneumonia. The bacteria are smeared on the nutrient agar plate, then 3 holes are punched in order with a diameter of 8 mm and then 20 μL of the solution is put into the well. After that, the plate was left at 10°C for 30 minutes; then, the plate was incubated at 37°C for 36 hours. Then the bacterial inhibition zone was measured and documented.

3. RESULTS AND DISCUSSION

3.1 X-Ray Diffraction Analysis for *Aloe vera* Powder

The phase of the *Aloe vera* powder was analyzed, as shown in Figure 1.

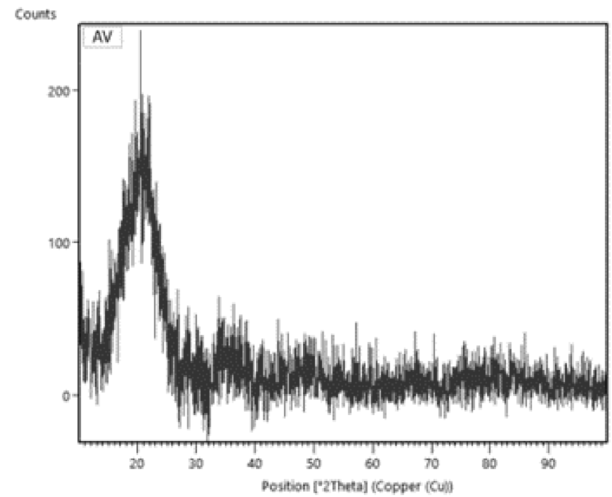


Figure 1. XRD Analysis of *Aloe vera* Powder

Figure 1 shows the results of the phase analysis of *Aloe vera* nanopowder with a milling time of 5 hours in the form of a spectrum or graph between the scattering angle (2θ) taken at a scattering angle of 10°-90° and the intensity (I). From the spectrogram presented in Figure 1 it can be seen that the *Aloe vera* nano powder formed is amorphous (no structure), this is indicated by the absence of diffraction peaks that appear at 2θ 20°. These results are consistent with the research Subramani et al. (2018) which prepared *Aloe vera* gel into *Aloe vera* nano powder where the XRD pattern of the prepared herbal nanoparticles showed no crystal diffraction peaks at a value of 2θ in the range of 20°-30°, as shown in Figure 1 below. Thus, the XRD pattern confirms the amorphous nature of the nanoparticles prepared from *Aloe vera* gel. This result was also studied by Gontijo et al. (2013) where the XRD pattern of *Aloe vera* flour showed an amorphous profile, *Aloe vera* is an organic compound.

3.2 Scanning Electron Microscopy analysis (SEM)

Analysis of characterization results using SEM for 9 variations of *Aloe vera*/PS coating samples. Differences in the morphological form in each variation of composition and temperature can be seen in Figure 2.

Based on Figure 2, it can be seen the differences in the morphology of the *Aloe vera*/PS nanocomposite with variations in composition and calcination temperature. From Figure 2 with 1:5/10°C shows the morphology with a granular but not dense surface shape, Figure 2b-2c with a temperature of 10°C shows the morphology with the surface shape where there are large round piles and visible small round particles scattered randomly, Figure 2d-2f shows the surface morphology at 50°C

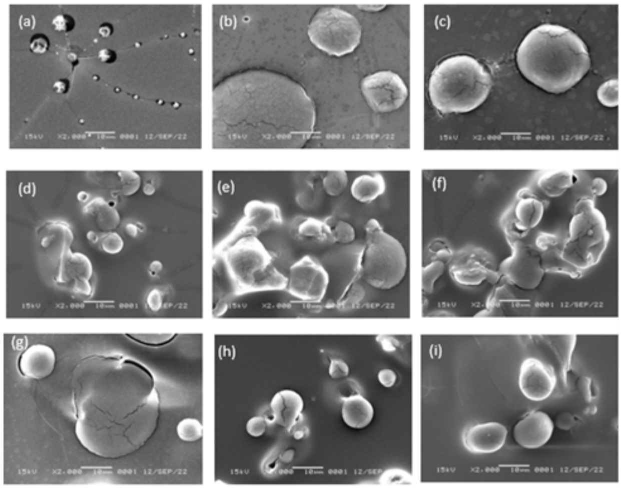


Figure 2. SEM Characterization Results of Nanocomposite *Aloe vera*/PS Surface with Variations in Composition and Temperature (a). 1:5/10°C (b). 2:5/10°C (c). 3:5/10°C (d). 1:5/50°C (e). 2:5/50°C (f). 3:5/50°C (g). 1:5/90°C (h). 2:5/90°C (i). 3:5/90°C

showing the shape of the surface which is formed by many small globular lumps, tightly arranged and there are small particles that are scattered randomly and tightly. And in Figure 2g-2i showing the morphology at 90°C it appears that the surface shape is composed of large clumps that are arranged sparsely and there are small particles that are tightly arranged. To distinguish the particle size of the 9 morphological variations used Image-J Software and get Particle size results as Table 1.

Table 1. Particle Size of *Aloe vera*/PS Thin Film

Composition *Aloe vera*/PS	Calcination temperature (°C)	Particle size (nm)
1:5	10	95.133
	50	51.455
	90	93.166
2:5	10	71.380
	50	50.496
	90	60.656
3:5	10	69.471
	50	21.243
	90	49.885

Based on Table 1, the smallest particle size is obtained at 50°C with variations in the composition of *Aloe vera*: PS a) 1:5 b) 2:5 and c) 3:5 having particle sizes of 51.455 nm, 50.496 nm, and 21.243 nm respectively. The more *Aloe vera* composition used the smaller the particle size obtained, this is because the more *Aloe vera* composition given the rougher the surface. The surface of *Aloe vera*/PS becomes harder because there is more agglomeration in the thin layer. According to Ratnawulan et al. (2020) to get good morphological results, there must be a lot of reinforcing deposits in the composite and this causes the grain size of the precipitate to increase and the surface morphology to become rough. The rough surface morphology will increase the hydrophobicity of the film. In the Cassie-Baxter model, air is trapped in the roughness when the air is suspended at nanometer surface dimensions. Water cannot enter into the gap, reducing the interaction between water and the surface. According to the Cassie-Baxter model, the smaller the surface area of the particles that interact with the air, the greater the contact angle that is formed (Rahayu and Zainuri, 2016). Therefore the 3:5 composition is the best composition variation to be used on the surface of the *Aloe vera*/PS nanocomposite and the temperature of 50°C is the best temperature for drying because the temperature of 50°C produces a better surface because it is a more porous and the movement of molecules in the solution is faster and the thickness of the solution decreases thereby increasing the stability and dispersion of the particles in the solution making the grain size finer and making the surface rougher.

3.3 Fourier Transmission Infrared analysis (FTIR)

Analysis of the characterization results using FTIR for 9 variations of *Aloe vera*/PS coating samples. The difference in the shape of the graph for each composition and temperature variation can be seen in Figure 3.

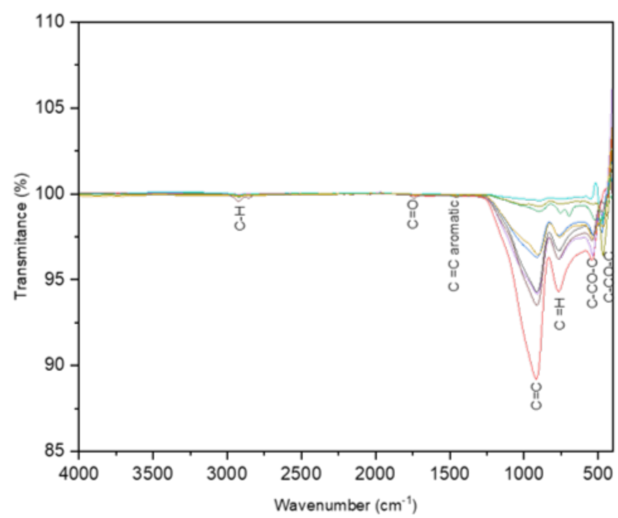


Figure 3. Graph of *Aloe vera*/PS Surface Analysis with Variations in Composition and Temperature

Figure 3 shows the relationship between wave number and surface transmittance of *Aloe vera*/PS with a wave number range of 4000 cm^{-1}-500 cm^{-1}. The graph consists of 9 samples distinguished from the color chart as follows: black indicates a variation of 1:5/10°C, red indicates a variation of 2:5/10°C, dark blue indicates a variation of 3:5/10°C, dark green indicates a variation of 1:5/50°C, purple indicates a variation of

2:5/50°C, orange indicates a variation of 3:5/50°C, light blue indicates a variation of 1:5/90°C, brown indicates 2:5/90°C and light green indicates 3:5/90°C. Based on the Figure 3, the spectrum has a C-H stretching vibration band at 2926.99 cm^{-1}, a C-O stretching vibration band at 1740 cm^{-1} shows the carbonyl group compound in *Aloe vera*, a C=C aromatic stretching vibration band at 1500 cm^{-1} shows the presence PS, vibration band C=C stretching at 980 cm^{-1}, vibration band C=H stretching at 700 cm^{-1} and vibration band C-CO-C stretching at 400 cm^{-1} and 500 cm^{-1} as fingerprints of aloin compounds from *Aloe vera*. The stretching experienced by the spectrum indicates a covalent bond, the atoms are not bound by a rigid relationship. Two atoms are related to each other because the two atomic nuclei are bonded to the same pair of electrons. The two atomic nuclei vibrate back and forth or sideways over one another. Composition and temperature have no significant effect on the dominant functional groups of *Aloe vera*/PS.

The FTIR results of this study are in accordance with Seifunnisha et al. (2021) and Chauhan and Kumar (2020) where the FTIR spectra of *Aloe vera* thin films have also been studied by Seifunnisha et al. (2021) where the dominant functional group was analyzed in the range 4000 cm^{-1}–400 cm^{-1}, there is a peak at 3026 cm^{-1} and 752 cm^{-1} correspond to C-H stretching. The symmetric and asymmetric vibrations of -CH2 are associated with the vibration bands at 2850 cm^{-1} and 2926 cm^{-1}. The vibrational band at 1805 cm^{-1} is caused by the C=O characteristic which indicates the presence of a carbonyl group in the *Aloe vera* sample. The C=C vibrations of the PS aromatic ring appear at 1598 cm^{-1} (Seifunnisha et al., 2021). According to Chauhan and Kumar (2020) the FTIR spectra of *Aloe vera* extract have a C=C vibration band at 1500 cm^{-1} and a C=O vibration band at 1700 cm^{-1} indicating the presence of aromatic rings and flavonoids in *Aloe vera*.

3.4 Hydrophobic Thin Film Properties

The contact angle was measured using ImageJ software and the measurement results were obtained directly. Contact angle testing was carried out with variations in the composition of nanocomposite *Aloe vera*/PS surface. This measurement was carried out with the aim of knowing at what temperature and composition variations the layers were more hydrophobic. Surfaces that are hydrophobic have a contact angle greater than 90°. The amount of contact angle produced at variations can be seen in Figure 4.

Documentation of the comparison of water droplets before being coated and after being coated with nanocomposite *Aloe vera*/PS surface on cloth can be seen in Table 2.

The nanocomposite surface mimics the wax coating on hydrophobic leaves. If water is dropped on a leaf surface that contains oil/wax, polar water cannot unite with the leaf surface because the attractive force between water molecules is stronger than the attractive force between water and the wax surface. The hydrogen bonding force between water molecules in the form of positive hydrogen atoms will attract oxygen atoms of other water molecules, hydrogen bonds cause water molecules to tend to stick together (cohesion). The strong cohesive force between water molecules at the boundary between water and air seems to form a skin that is strong enough to be called surface tension (Ratnawulan et al., 2020), therefore water will form an arc on a waxy surface such as the surface of the nanocomposite *Aloe vera*/PS.

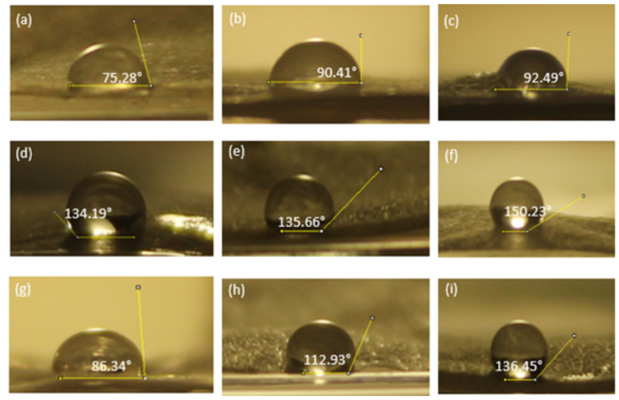


Figure 4. The Contact Angle of Water Droplets in Detail on 9 Surface Variations of a Thin Layer of *Aloe vera*/PS that has been Coated on a Cloth (a). 1:5/10°C (b). 2:5/10°C (c). 3:5/10°C (d).1:5/50°C (e). 2:5/50°C (f). 3:5/50°C (g). 1:5/90°C (h). 2:5/90°C (i). 3:5/90°C

The change in the size of the box angle obtained is related to the polarity or nonpolarity of a substrate surface. According to Pereira et al. (2015) on a more polar surface the wettability will be higher (lower contact angle), conversely on a more non-polar surface the wettability will be lower (larger contact angle). This low wettability is also due to the cohesion forces that occur between water molecules that are greater than the adhesion forces that occur between the water and the surface of the nanocomposite layer, which results in the formation of water droplets on the surface of the nanocomposite layer. Water is unable to wet hydrophobic surfaces because the cohesion force is greater than the adhesion force. So that in this study initially the substrate was not yet hydrophobic, but with increasing the calcination temperature to the optimum temperature, the surface properties which were not previously hydrophobic (polar) became hydrophobic (non-polar) (Pereira et al., 2015).

3.5 Antibacterial Performance

Nanocomposite *Aloe vera*/PS solution with composition 1:5, 2:5, dan 3:5 was analyzed for the activity of antimicrobial pathogens using the agar well diffusion method and gave results as shown in Figure 5.

Based on Figure 5 the research of pathogenic antimicrobials using the agar well-diffusion method was carried out on 3 variations of the composition of nanocomposite *Aloe vera*/PS namely 1:5, 2:5 dan 3:5 to observe the antimicrobial activity of nanocomposite *Aloe vera*/PS against Klebsiella pneumonia bacteria which will be used as a coating against pathogenic mi-

Table 2. The Difference in Fabric Properties Before and After Being Coated with *Aloe Vera*/PS Solution

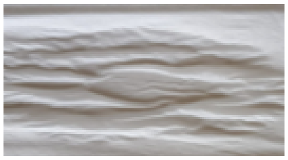
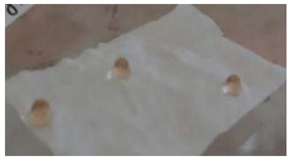
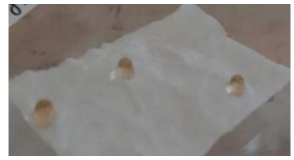
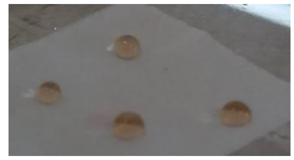
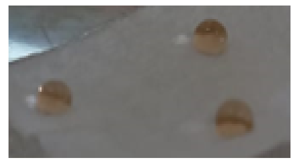
Before coating		After coating		
	°C	1:5	2:5	3:5
	10			
	50			
	90			

Table 3. Zone of Inhibition of Nanocomposite *Aloe vera*/PS Pathogenic Antimicrobial Activity

No	Microorganisms	Composition *Aloe vera*/PS	Inhibition zone (mm)	
			Aloe vera/PS	Control (Amoxilin)
1	*Klebsiella pneumonia*	1:5	0 mm	23.09 mm
2	*Klebsiella pneumonia*	2:5	0 mm	23.09 mm
3	*Klebsiella pneumonia*	3:5	20.18 mm	23.09 mm

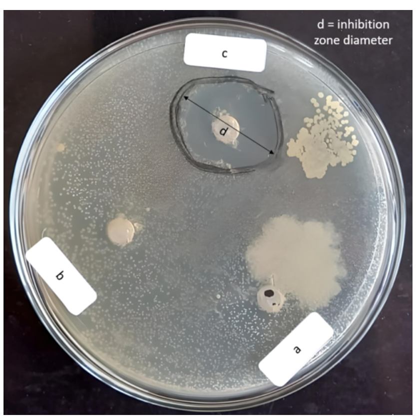


Figure 5. Antibacterial Performance of Nanocomposite *Aloe vera*/PS Solution a. 1:5 b. 2:5 c. 3:5

croorganisms on fabrics. There were 3 wells with the same hole diameter of 0.5 mm, nanocomposite *Aloe vera*/PS solution was put into the hole and incubated for 36 hours, after incubation a bacterial inhibition area was formed in each composition variation which can be seen from the clear area around the wells. The size of the inhibition zone is shown in Table 3.

In the variations of nanocomposite *Aloe vera*/PS 1:5 and 2:5, it can be seen that there are still many bacteria that have gathered and no clear zones have formed around the wellbore, this indicates the inability of 1:5 and 2:5 in holding bacteria. This event is caused by the use of too little *Aloe vera* so that the active compounds in *Aloe vera* are not able to withstand bacteria and are less efficient when used as a base material for anti-microorganism coatings. In the 3:5 variation, a difference was seen where the bacteria were unable to approach the well and there was a clear zone with a diameter of 20.18 mm with a large diameter of control using 23.09 mm amoxicillin. This shows that the variation in the composition of 3:5 can inhibit bacterial growth and is efficient when used as a coating for anti-pathogenic microorganisms. According to Subramani et al. (2018) *Aloe vera* nanoparticle samples had higher work results in AV composition which was largely due to the many phytochemical compounds which included several terpenoids, flavonoids, and tannins, and saponins in *Aloe vera leaves*, which have broad antibacterial activity against pathogenic bacteria. AV also contains Acemannan in the form of a polysaccharide component which has antimicrobial activity through its ability to stimulate phagocytic leukocytes which are commonly used for wound treatment due to the good healing properties of the existing vitamin D (Subramani et al., 2018).

Therefore, the results of the study showed that the fabric nanocomposites coated with Nanocomposite *Aloe vera*/PS with a composition of 3:5 tended to exhibit higher antibacterial properties. resulting in inhibition of DNA synthesis and higher cell death. Along with that, the structure of the nanoparticles also plays a major role in the antibacterial properties because the morphological differences in the nanocomposite samples also add to the higher antibacterial activity. According to Seifunnisha et al. (2021) Measurement of the contact angle can prove that the thin layer exhibits non-wettable properties which are also confirmed by surface roughness analysis, then the Nanocomposite *Aloe vera*/PS surface shows potential antibacterial activity against pathogens, where Nanocomposite *Aloe vera*/PS shows excellent bactericidal activity due to the presence of a large number of phytochemicals such as alkaloids, flavonoids, phenolic compounds and terpenoids.

4. CONCLUSION

The best composition obtained from this study was obtained at nanocomposite *Aloe vera*/PS 3:5 composition with a temperature of 50°C with SEM results of the surface morphology of the thin layer forming a particle size of 21.24 nm, FTIR analysis showed that the dominant functional group is in the vibrational band of 2629 cm^{-1} indicating the presence of a long hydroxyl chain, the vibration band of 1500 cm^{-1} with C-H stretching of the aromatic ring is due to the characteristics of the polystyrene polymer and the wave number is 1700 cm^{-1} with C=O stretching which indicates the characteristics of the carbonyl group in *Aloe vera* samples in the form of flavonoid compounds. The resistance of *Aloe vera*/PS solution to Klebsiella pneumonia resulted in a 20.18 mm diameter barrier showing strong resistance against bacteria and hydrophobic properties with a size of 150.23° and X-Ray diffraction analysis shows amorphous of the *Aloe vera* powder.

5. ACKNOWLEDGMENT

The technical support of Prof. Dr. Ratnawulan, M.Si; Dr. Ramli, M.Si; Dr. Hamdi, M.Si and Irna Humairah S.Si, M. Biomed is gratefully acknowledged.

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