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Wound Dressing Based on Banana Peels Waste and Chitosan by Strengthening Lignin as Wound Healing Medicine

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Abstract. Recent studies in new applications of wound dressing offer and promote the process of wound healing. The purpose of this study was to develop lignin-based wound dressing on banana peel and then put it into chitosan film for wound dressing applications. The use of banana peels helps the formation of skin compounds so that they can be used to treat better wounds. Banana peel added to chitosan as matrix filler with concentration (0, 1, 3, 5, 7, 10% wt), then the dressing membrane swelling in 48 hours is at 0% wt 120.5931%, at 1% wt 99. 9981%, 3% wt 79.2916%, 5% wt is 68.1819%, 7% wt is 61.9173% and 10% wt is 45.3981%. Fourier Transform Infrared (FTIR) test, Scanning Electron Microscopy (SEM), swelling properties were carried out to characterize the prepared film. Antimicrobial tests were carried out by disc diffusion method and film growth inhibitory effects including different amounts studied in Escherichia coli and Staphylococcus aureus. FTIR at a concentration of 7% wt showed that there was an interaction between banana peel and chitosan on the absorption band 3185,728 cm^{-1}. In SEM the best sample morphology structure at 5% wt shows a good interface interface. Addition of banana peel as lignin decreases the level of swelling of water in wound dressing. In addition, Staphylococcus aureus is the most sensitive strain recorded in wound dressing. Banana skin which is included in chitosan film seems to be a potential and new biomaterial for wound healing applications.

Keyword : *chitosan, banana peels powder, fourier transform infrared scanning electron microscope*

1. Introduction

Wound infections are serious infections that have occurred in all parts of the world [1]. Many solutions have been studied and found to be able to cure wound infections. Wound healing requires a complex method and certainly does not restore the skin as usual. Severe wounds can even die from dark scars due to damage to skin tissue [2,3].

At present there are many multifunctional biomaterials for cell scaffolding, one of them is chitosan which is highly recommended in the engineering of skin tissue because it has structural characteristics similar to glucosamine from the natural extracellular matrix [4]. Chitosan has good biocompatibility, low toxicity, anti-infection activity and biodegradability [5]. Judging from the physical-chemical properties of chitosan, it can be used for various cell forms such as wound dressing, membranes, coatings, fibers, and sponges. Several studies have shown that higher molecular weight chitosan has better film-forming properties in skin tissue engineering [6]. This is the reason for the use of chitosan as a cell scaffold, specifically in making wound dressing.

Up until now, there have been many studies and development of wound dressing as an approach to the field of tissue engineering to deal with the effects of injuries in a faster period of time [7].

Several studies have shown that chitosan has a weakness if it is used as a wound dressing, this is due to the low mechanical strength of chitosan. In [8], trying to add lignin to chitosan, the results show that the tensile strength, storage modulus, thermal degradation temperature and chitosan can be increased by adding lignin.

Lignin is considered a raw material with high recovery potential, widely available in nature, cheap and environmentally friendly [9]. Lignin is almost present in all types of wood fiber plants [10], such as bananas, sugar cane, sengon and so on.

Especially bananas, containing about 10% lignin [11]. For making wound dressing, the banana part used is the skin, this is because besides lignin, the banana skin also helps the formation of skin compounds so that it can be used to treat bruises, burns, and other wound infections [12]. In addition, the use of banana peel as a wound dressing is supported by the availability of banana skin as waste [13]. The reuse of banana peel waste into a product can increase its economic value.

The development of chitosan-based wound dressing and banana peel waste in Indonesia tends to be lacking, due to limited information and research conducted. This is a good opportunity, especially in the medical field in Indonesia, which generally relies on healing methods using chemicals. The incorporation of natural polymers namely chitosan with banana peel is expected to be able to create wound dressing products with characteristics that are appropriate and provide benefits, both in terms of science, health, and in terms of economics. The purpose of this study was to make chitosan-based wound dressing and banana peel waste which were characterized using Fourier Transform Infrared (FTIR), Scanning Electron Microscopy (SEM), Swelling Properties and antibacterial susceptibility to Escherichia coli and Staphylococcus aureus.

2. Materials and Methods

2.1 Materials

Chitosan banana peel waste, glycerol, acetic acid, Escherichia coli bacteria and Staphylococcus aureus.

2.2 Processing of Banana Peels Powder

Banana Peels that have been collected, washed with water to remove dirt, and cut into small pieces and then dried in an oven at 120°C for 24 hours. After drying, the banana peel is ground with micro milling. Then the skin is crushed and sifted so that the particle size remains stable and stored at room temperature in a plastic container until it is used. Furthermore, banana peel powder was carried out by morphological testing using Scanning Electron Microscope (SEM) and functional group testing using the Fourier Transform Infrared (FTIR).

2.3 Synthesis of Membrane Wound Dressing Chitosan and Banana Peels Powder

Prepared as much as 1 gram of chitosan powder, dissolved in 2% acetic acid solution and stirred for 4 hours at room temperature. Prepared 1 ml of glycerol and added as a crosslinker and plasticizer. Chitosan solution was put into Teflon mold and mixed with banana peel powder with different concentrations (0, 1, 3, 5, 7 and 10% wt), dried at 40°C for 24 hours to evaporate the solvent and form a membrane. The prepared membrane is gently peeled, and further dried by keeping the oven 40°C for 4 hours. To give a better sight, the membrane wound dressing sample were depicted in Figure 1.

Figure 1. Membrane Wound dressing sample

2.4 Measuring Techniques

2.4.1 Fourier Transform Infrared (FTIR)
The testing of synthesized chitosan functional groups was characterized using Fourier Transform Infrared (FTIR) on wave absorption of 4000-500 cm^{-1}. For the wound dressing sample a thin and clear sized layer is formed. After that the sample is inserted into the FTIR device tube to get the functional group contained in the sample. The spectrum will be seen in the range 4000-500 cm^{-1}.

2.4.2 Scanning Electron Microscopy (SEM)
The homogeneity of the morphological structure of banana skin powder and wound dressing membrane as distribution of chitosan matrix fillers was seen by Scanning Electron Microscope (SEM).

2.4.3 Swelling Properties
Weight specimens were immersed in distilled water at 37°C. After 24 hours of immersion, the specimen is removed from the water, dried with a paper filter and weighed. The sample was soaked again for another 24 hours and then removed from the water, dried with filter paper and weighed.

$$\% \text{ Swelling} = \frac{(Mw - Md)}{Md \times 100} \tag{1}$$

where, *Mw* is the wet weight and *Md* is the dry weight of the sample.

2.4.4 Antimicrobial Susceptibilty
The antibacterial activity of membrane chitosan-banana powder wound dressing was investigated using the disc diffusion method on a petri dish [14]. Bacterial cultures that grow in the mid-logarithmic phase are placed in agar media. Escherichia coli and Staphylococcus aureus are injected into agar media. After compaction to coat the agar, the chitosan-banana powder (15 mm diameter) membrane with different concentrations (0, 1, 3, 5, 7 and 10% wt) was placed on the agar surface. The layers were incubated at 37°C for 24 hours and at 28°C for the next 72 hours.

3. Result and Discussion
3.1 Fourier Transform Infrared (FTIR)
The spectra of FTIR membrane chitosan wound dressing-waste banana peel with different concentrations (0, 1, 3, 5, 7 and 10% wt) has been recorded in wave spectrum of 4000-500 cm^{-1}. The chitosan spectrum and banana peel powder are shown in Figures 2 and 3, respectively.

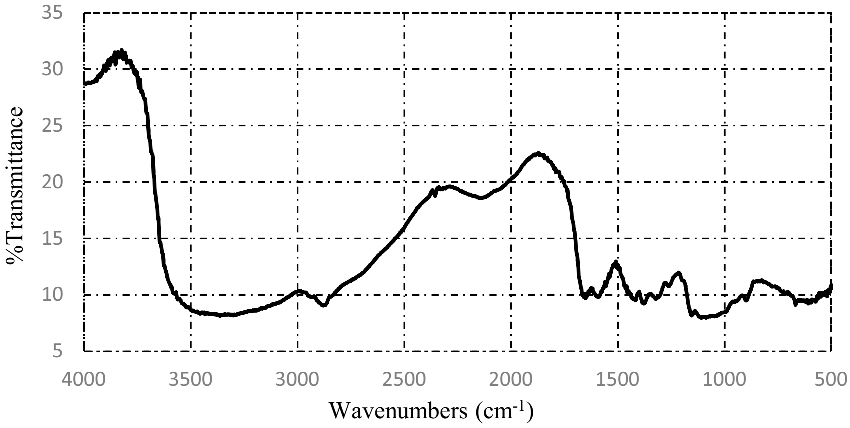


Figure 2. FTIR Spectrum of Chitosan

From Figure 2 it is shown that chitosan contains many functional groups (hydroxyl, carbonyl, carboxyl, amine, and amide). Please note that adding glycerol to chitosan does not provide a new band in the chitosan spectrum. Previous studies on banana peel powder revealed that banana peel powder contains polymers (pectin, hemicellulose and lignin) and they are rich in organic functional groups such as -COOH, $-NH_2$ and O-H [15]. Characteristics of absorption of chitosan and banana peel bands are similar to those previously studied [16]. FT-IR on banana skin chitosan (10% by weight), as shown in the picture there is a clear composite difference between the ingredients compared to the pure chitosan spectrum. Its peak at 3520 cm^{-1} corresponds to the expanding O-H stretch wave and shifts to 3628 cm^{-1}. Increasing the intensity of the band 2898 cm^{-1} which corresponds to the C-H wave. The new band appears at 1760 cm^{-1} which corresponds to the carboxyl group.

FTIR results also show the role of acid in chitosan. With FTIR profiles that show absorbance similar to each other, this shows the role of acids in the reaction of chitosan nanoparticles preparation. Acids are not intra- and intermolecular-bound with di- and tri- carboxylic acids [17]. The FTIR spectrum also does not show the formation of new polymers. This shows that chitosan does not polymerize with acid. It can be concluded that acids only act as proton donors which can dissolve chitosan [18].

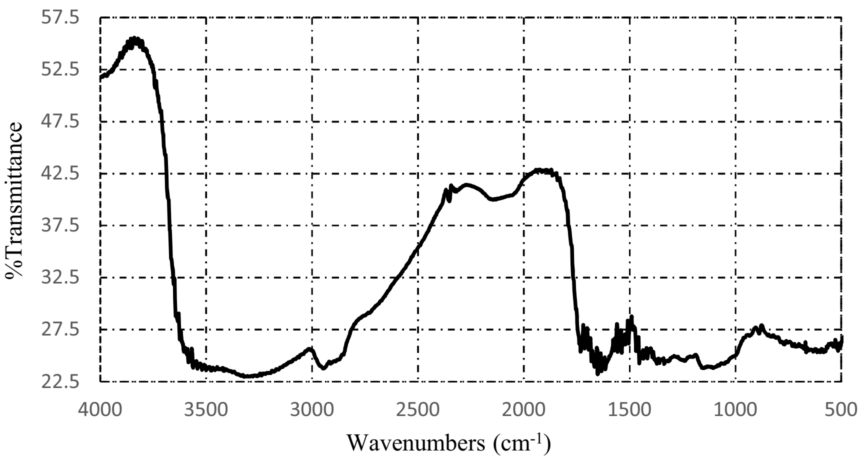


Figure 3. FTIR Spectrum of Banana Peels Powder

Figure 3 shows the membrane membrane of wound dressing containing banana peel powder with different concentrations (0, 1, 3, 5, 7 and 10% wt). From the picture it was noted that there was an increase in amide intensity which reached its peak at 1638 cm^{-1} with the addition of fillers. Bands on the absorption band of 2890 cm^{-1} and 2,900 cm^{-1} have increased as indicated by the characteristics for C-H due to the addition of fillers. This band appears on the absorption band 1760 cm^{-1} which corresponds to the carboxyl group in addition to increasing the intensity of the absorption band 1440 cm^{-1} which occurs an interaction between the positive charge of the amine chitosan group and the negatively charged carboxyl group on banana skin which makes good interactions filler and matrix.

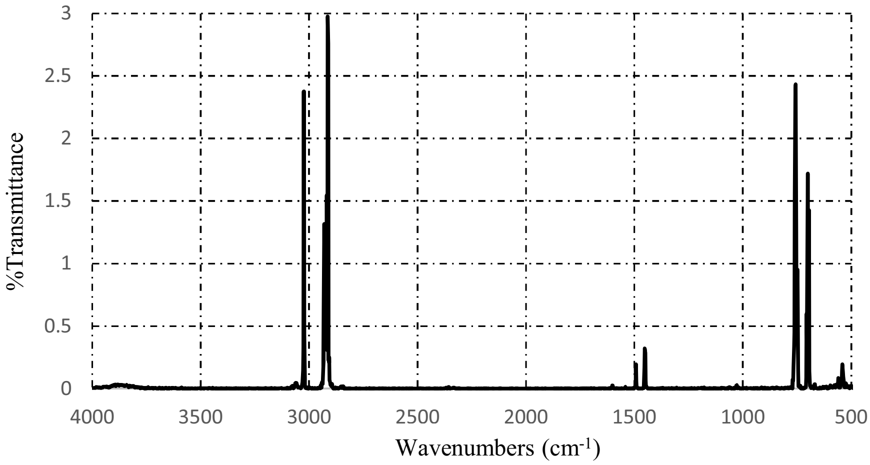


Figure 4. The FTIR of the membrane wound dressing of 7% wt

3.2 Scanning Electron Microscope (SEM)

In Figure 5 shows the SEM morphological structure of banana skin powder particles. The particle size is in the range of 20-80 µm in length and 20-30 µm in width.

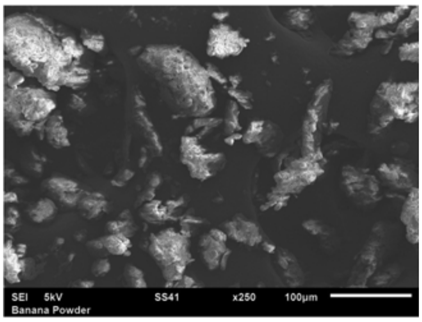
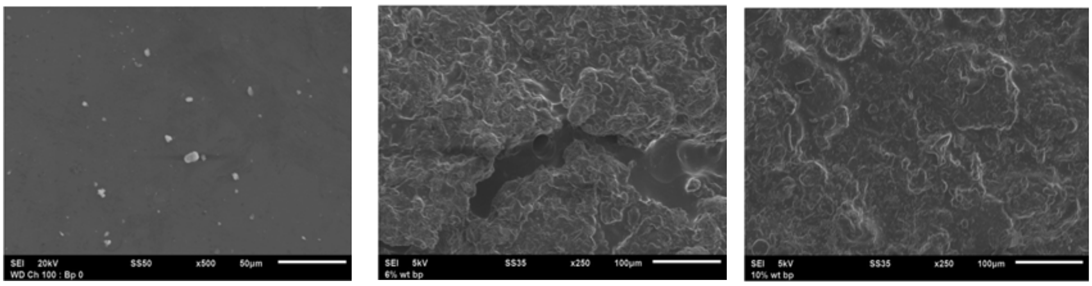


Figure 5. Morphological structure of banana peels powder

The morphological structure of SEM with concentrations (0, 1, 3, 5, 7 and 10% wt) of chitosan and banana peel powder is shown in Figure 5. Morphological analysis of SEM showed that uniform wound dressing membrane fillers were distributed in a 5% wt concentration chitosan matrix, with the increase in filler concentration in irregular distribution with several filler aggregates.



(a) (b) (c)

Figure 6. Membrane morphological structure of wound dressing at concentrations (a) 0% wt; (b) 5% wt; and (c) 10% wt

3.3 Swelling Properties

It is important to determine the swelling properties of water for biomaterials if they are used as wound cover material. This is a measure of the film's capacity to absorb wound exudates. The ideal environment for good wound healing is to keep the wound moist [19].

Table 1. Swelling Properties of Wound Dressing

Sample	*Swelling* (%)		
	0 h	24 h	48 h
0% wt	57.94	93.14	120.59
1% wt	33.06	60.17	100
3% wt	19.22	50.94	79.29
5% wt	10.18	40.94	68.18
7% wt	59.72	33.73	61.92
10% wt	29.92	24.18	45.4

Water absorption from chitosan is related to the hydrophilic group (hydroxyl and amino groups) of the polysaccharide. Membrane swelling of chitosan-banana powder wound dressing after 24 hours and 48 hours of immersion in distilled water were shown in Figure 7.

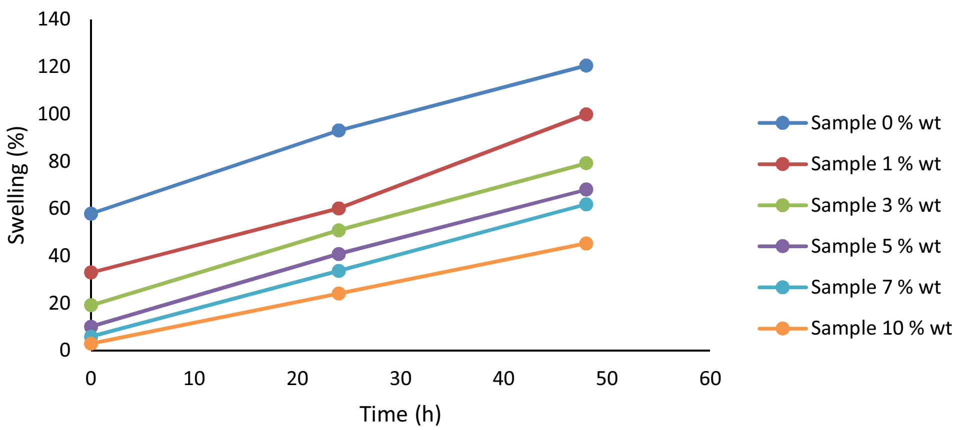


Figure 7. Swelling properties on membranes wound dressing chitosan-banana peels powder

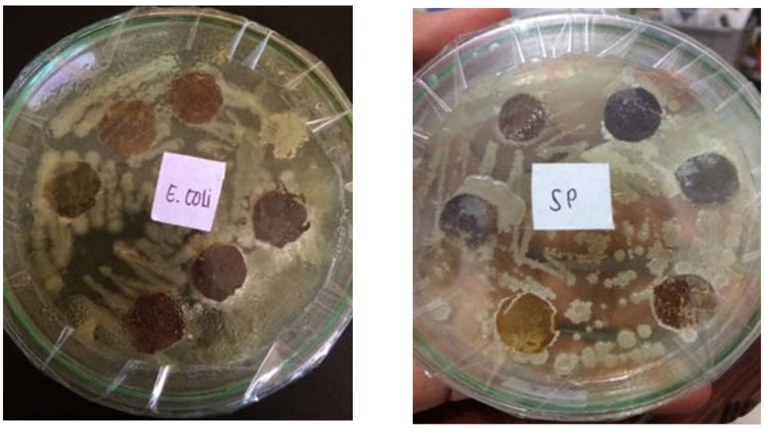
It is clear from the figure that the 0% wt sample shows a higher swelling rate compared to the added filler, adding banana skin powder to chitosan results in a decrease in swelling and hence increases water resistance This behavior is caused by a kind of interaction between negatively charged compounds in banana peel powder such as carboxylic acids and positively charged groups on the chitosan backbone chain. This interaction limits the mobility of the chitosan chain which reduces water uptake. To better understand the effect of banana peel powder on chitosan swelling behavior, the plot of water uptake versus time is illustrated in Figure 7.

3.4 Antimicrobial Activity

The point of this study shows that the combination of chitosan with banana peel fillers has a synergistic interaction with a broad antimicrobial spectrum against gram negative, gram positive bacteria and even against strains of yeast culture that show the ability of biofilm formation. The results in Figure 8 show that the chitosan-sanitary membrane as a banana peel has a synergistic action with the highest activity of 10% wt. In addition, Staphylococcus aureus is the most sensitive strain recorded for this membrane.

Chitosan-banana peel powder dressing membrane also showed higher activity against Escherichia coli strains (gram negative bacteria) than Staphylococcus aureus strains (gram positive bacteria). On that basis, several studies note that chitosan affects gram negative and positive but rather like argumentative effects, some findings indicate more effectiveness against gram negative compared to gram positive [20].

However, this finding regarding the surface polarity of bacteria, the outer membrane of gram-negative bacteria has a lipopolysaccharide, very negatively charged, which allows attachment to polycational chitosan relative to gram-positive, which consists of peptidoglycan associated with polysaccharides and teicoic acid [21]. which in turn supports glucose [22]. Effects similar to fungal and yeast cells that can interfere with growth [23].



(a) (b)

Figure 8. Effect of antibacterial activity to membrane wound dressing chitosan and banana peel powder on (a) Escherichia coli and (b) Staphylococcus aureus

Scientists support another mechanism, the antimicrobial activity of chitosan which is associated with metal chelation, where chitosan has excellent metal binding ability because the amine group takes cationic metals with chelation [24]. Here, we hypothesize that such a relationship is one of the new approaches to improve the properties of these composites [25] such as increasing biodegradability and antimicrobial activity, moreover banana peels contain polymers such as lignin, hemicellulose and pectin [26] which can help improve composite properties newly formed. Therefore, we can use it for medical purposes as wound dressing. In the food industry, banana flour/chitosan is applied to preserve new vegetables, showing antimicrobial properties against bacteria when used [27].

Although in this study banana peels were used as they are, the results of antibacterial studies were comparable to those studied by P.B. Franco and his co-authors studied the antibacterial activity of chitosan membranes associated with active compounds extracted from banana peels [28].

4. Conclusion

The use of banana skin helps the formation of skin compounds so that it can be used to treat better wounds. Banana skin is added to chitosan as a matrix filler with concentrations (0, 1, 3, 5, 7, 10% wt), then the swelling of the swelling membrane in 48 hours is at 0% wt 120.5931%, at 1% wt 99. 9981 %, 3% with 79.2916%, 5% with 68.1819%, 7% with 61.9173% and 10% with 45.3981%. FTIR at a concentration of 7% wt showed that there was an interaction between banana peel and chitosan on the absorption band 3185,728 cm-1. In SEM the best morphological sample structure at 5% wt shows a good interface. Addition of banana skin because lignin reduces the level of swelling of water in wound dressing. In addition, Staphylococcus aureus is the most sensitive type recorded in wound dressing. Banana peels included in chitosan films seem to be potential and new biomaterials for wound healing applications.

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