Preparation and characterization of chitosan-starch Janeng membranes cross-linking with citric acid

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Abstract. Neat chitosan membranes have shortcomings in their application owing to weak mechanical properties, thus, requiring modification. In this study, a chitosan membrane was modified utilizing starch and cross-linking agent (citric acid) addition. The characterization of chitosan-starch membranes using Universal Testing Machine, Scanning Electron Microscope (SEM), Fourier Transform Infra-Red (FTIR), Differential Scanning Calorimetry (DSC), and Thermogravimetric Analysis (TGA) had proven the cross-linking formation. The cross-linked membrane had better tensile strength (11.07 kgf/mm²) than the uncross-linked membrane (66.07kgf/mm²). The DSC thermogram of uncross-linked and cross-linked membranes showed endothermic and exothermic peaks at different temperatures. In addition to the membranes’ thermal characteristics, the DTA thermogram showed the decomposition process on the uncross-linked membrane uncross-linking occurred at a temperature range of 277.74-363.27 °C, while the decomposition of the cross-linked membrane was observed at a range of 287.83-356.38 °C. The FT-IR spectra confirmed a decrease in the intensity of the absorption peak associated with cross-linking formation within the membrane.

1. Introduction
Chitosan-based membranes have been reported to have wide applicability, including in the desalination of seawater and brackish [1-4]. However, the application of neat chitosan membranes still has several weaknesses, such as brittleness, stiffness, and weak resistance against the acidic solution. The weaknesses can reduce the performance of the chitosan membrane in water purification. One alternative route to overcome the weaknesses is through physical and chemical modifications [5-7]. In this study, we explore the modification by means of polymer blending and cross-linking.

Chemical modification of the membrane surface through cross-linking is an effective technique to overcome the aforementioned weaknesses [8, 9]. Cross-linking reactions are used to link polymer chains, one to another, by forming permanent bonds through covalent/ionic bonds. It also provides a more flexible bond since it breaks the intramolecular bonds that contribute to the membrane stiffness. One of the polymers that can be used to blend with chitosan is starch [10]. Starch is a natural carbohydrate polymer that can be obtained from various bio-resources. Dioscorea hispida (the local name: janeng) tubers had been reported to be an alternative resource for starch due to their low price and high availability [11]. The use of starch in membrane manufacturing can increase the economic
potential of janeng and environmentally friendliness. Starch possesses active sites, contributed by the presence of hydroxyl groups in their chemical structure that can form bonds with polymers or other compounds. The addition of a cross-linking agent assists the formation of the chitosan-starch cross-linking. The addition of a cross-linking agent in the polymer mixture can increase the stability (especially in acidic condition) and mechanical durability [10, 12]. One of the cross-linking agents that can be used is citric acid. Citric acid is chosen as a cross-linking agent stem from its low toxicity and non-carcinogenicity, where the product can be safe to use.

In this study, we explore the modification using polymer blending and cross-linking. A technique of blending chitosan and janeng starch was carried out to improve the mechanical properties, assisted by cross-linking formation by citric acid. The success of this process was investigated by means of mechanical strength, thermal behavior, functional group and morphology analysis.

2. Materials and methods

2.1. Materials

Janeng was collected from Aceh Besar Regency (Aceh, Indonesia) and used for starch production. Medical grade chitosan with an acetylation degree above 85% was used in a membrane preparation. Glacial acetic acid was as a polymer-solvent, citric acid was used as a cross-linking agent, and sodium metabisulfite and Argentum nitrates were used in starch extraction. Glacial acetic acid, citric acid, sodium sulfite, and Argentum nitrates were purchased and supplied from Merck KgaA (Germany).

2.2. Starch isolation from janeng

The janeng-based starch (JS) was isolated using the method proposed by Saiful et al. [11]. Janeng tubers, as much as 5 kg, were firstly cleaned by soaking in distilled water. The procedure was followed by peeling and chopping the cleaned janeng tubers, then rinsed with distilled water. Next, sodium metabisulfite solution (1.12 g / l) was added to the janeng tuber pieces and processed using a crusher. The water and the pulp of the janeng are separated by filtration. The filtrate obtained is left to stand for 24 hours until the precipitate is produced. The precipitate obtained is JS, which is then washed repeatedly to remove the remaining HCN in the JS. The loss of HCN was indicated by a negative test result using 1 M AgNO₃ solution. Then, the JS was dried for 24 hours at 70 °C. After drying, the JS is mashed and sifted (100 mesh) to give JS powder.

2.3. Preparation of chitosan-starch membrane

The process of making the chitosan-starch cross-linking membrane using the citric acid cross-linking agent referred to the method developed by Lusiana et al. [14]. A 1.5% (w/v) pure chitosan solution was prepared by dissolving 2.5 gram chitosan in 200 ml of 1% (v/v) acetic acid solution. The starch used was priorly gelatinized, prepared by mixing 3 grams of starch in 200 ml of distilled water. The gelatinization process was carried out by heating the mixture at 70-80 °C for 5 minutes. Furthermore, the gelatinized starch was mixed in the chitosan solution, which was stirred using a magnetic stirrer. The chitosan-starch mixture was added with 0.01 mol citric acid (1.78 g) and the mixture was stirred at 70-80 °C for 24 hours to produce a casting solution. The thin membrane layer was printed on a granite plate with a thickness of 45 μm and oven-dried at 35 °C. The formed membrane was then washed with 1% NaOH to remove the solvent and followed by washing with distilled water repeatedly until the pH was neutral. Then, the membrane was allowed to dry at room temperature. These membranes were used for further characterization.

2.4. Characterization of chitosan-starch membrane

The citric acid uncross-linked and cross-linked chitosan-starch membranes were characterized for mechanical strength, Differential Scanning Calorimetry (DSC), Thermal Gravimetry Analysis (TGA), Scanning Electron Microscope (SEM) analysis, and Fourier Transform-Infrared (FT-IR) analysis.
2.5. Tensile and elongation test

The membrane tensile test referred to the ASTM D-368 method. This test was conducted at the Physics and Materials Laboratory of the Department of Physics, Faculty of Mathematics and Natural Sciences, Universitas Syiah Kuala. The membrane tensile test specimens are presented in Figure 1.

![Diagram of tensile strength test specimen](image)

**Figure 1.** The shape of the tensile strength test specimen. Source: ASTM D-368.

In testing, the two ends of the specimen (the tip of the specimen) were clamped using a piece of mechanical test equipment; observations were made on the tensile strength (kgf/mm^2) and the elongation percentage.

2.6. FT-IR analysis

Membrane testing using FT-IR (Shimadzu FT-IR—Prestige 21 Series, Shimadzu, Japan) was carried out by cutting the membrane with a size of 5x5 cm and then analyzed at a wavenumber of 4000-500 cm^-1 in transmittance mode. This test was carried out at the Instrumentation Laboratory, Department of Chemistry, Faculty of Mathematics and Natural Sciences, Universitas Syiah Kuala.

2.7. Thermal analysis

The membrane with a size of 1x1 cm was tested for thermal resistance using the Shimadzu DSC-60 differential scanning calorimeter and Shimadzu DTG-60 thermal gravimetric analyzer (Shimadzu, Kyoto, Japan) instruments. This test was carried out in the Department of Chemical Engineering, Lhokseumawe State Polytechnic.

2.8. SEM analysis

The membrane structure was analyzed using SEM JEOL JSM 6510 LA to see the membrane surface with several times the magnification. This test was carried out at the FMIPA Laboratory of the Bandung Institute of Technology. The membrane was cut to a size of 0.5 x 0.5 cm and tested using a magnification of 1500x.

3. Results and discussion

3.1. Janeng-based starch powder isolation

JS powder has been isolated from the janeng tubers appeared to be white in visual color. The JS powder was free from cyanide toxins. Free cyanide was proven based on the negative results from the testing with 1 M AgNO3 reagent solution. The yield of was obtained to be 7% (w/w). These characteristics and yield percentage are consistent with the previous studies [11]. The starch obtained in this study was used as a base material for membrane preparation in this study. The SEM analysis on the JS powder in Figure 2 shows that the particles produced have a relatively uniform structure and shape with an average size of 2.5 µm and have a porous structure.
Figure 2. SEM images of isolated starch particles from janeng tubers with magnification of (a) 5,000x and (b) 50,000x.

3.2. Membrane chitosan-starch

The citric acid uncross-linked and cross-linked chitosan-starch membranes were prepared by mixing a 1.5\% (w/v) chitosan solution in 1\% (v/v) acetic acid and a 1.5\% (w/v) starch mixture in distilled water. The JS mixed into the casting solution had been priorly gelatinized. Gelatinization was needed to break the starch structure so that it could mix/absorb water by forming a gel through the formation of hydrogen bonds with amylose and amylopectin. Mixing the 1.5\% (w/v) chitosan solution with gelatinized starch resulted in a homogeneous mixture. The visual images of citric acid uncross-linked and cross-linked chitosan-starch membranes can be seen in Figure 3a and b, respectively. The visual observation shows that the physical appearance of the membrane looks the same. However, based on direct observations on the manufacturing process, the uncross-linked chitosan-starch (CS0) membrane has a good physical appearance, elastic, shrink during the drying process, and easily removed from the mold. However, better physical appearance, elasticity, ease of being removed from the mold, and brittleness were observed on the citric acid cross-linked chitosan-starch (CSC) membrane.

Figure 3. A visual depiction of chitosan-starch membranes; (a). CS0 membrane; (b) CSC membrane.
3.3. Mechanical strength of the membrane

The mechanical properties of the membrane were analyzed to determine the tensile strength and elongation of the membrane. The membrane's mechanical properties are shown in Figure 4.

![Tensile Strength (kgf/mm²) vs. Elongation %]

**Figure 4.** Tensile strength and elongation for CS0 and CSC membrane.

Figure 4 shows that the CSC membrane produced a much higher tensile strength than the CS0 membrane. The tensile strength value of the CS0 membrane was 10.07 Kgf/mm² and experiencing an increase after cross-linked up to 556% (66.07 Kgf/mm²). Meanwhile, the elongation values for CS0 and CSC membranes were 11.27% and 12.7%, respectively. It reveals that the CSC membranes had higher elongation compared to CS0. The formation of cross-linking significantly affects the membrane tensile strength value. The cross-linking via citric acid addition had caused the membrane's structure to become denser, which eventually led to more robust mechanical properties. Lusiana et al. [13] reported that the cross-linking reaction between the amine groups in chitosan and the carboxyl groups in citric acid was assigned to increase membrane elasticity. Gohil et al. [14] also found that a cross-linking agent's addition caused the membrane structure to become denser.

3.4. DSC and TGA analysis the membrane

DSC thermal analysis is performed to measure the difference in heat or energy in a sample as a function of time or temperature. TGA thermal analysis is used to measure the change in weight of a compound as a temperature or time function. DSC and TGA thermal analysis were performed on CS0 and CSC membranes. The results of the DSC and TGA testing are in the form of a thermogram, which can be seen in Figure 5.

Figure 5a exhibits the thermogram of CS0 and CSC membranes, where the endothermic and exothermic peaks were observed at different temperatures. Endothermic peaks on the CS0 and CSC membranes were detected at 128.53 °C and 67.48 °C, respectively. The emergence of endothermic peaks is assigned to the evaporation of water in both membranes. Exothermic peaks on CS0 and CSC membranes were respectively detected at 318, 93 °C and 321.24 °C. The appearance of an exothermic peak represents a decomposition process. The TGA analysis supports this. The TGA thermogram results of the two membranes can be seen in Figure 4b, showing a single decomposition stage. The decomposition of the CS0 membrane was detected at temperatures ranged from 277.74 to 363.27 °C with a weight loss of 66.3%. In contrast, the CSC membrane decomposed at a temperature range of 287.83 – 356.38 °C, with a weight loss of 65.4%.
Figure 5. Thermogram results of (a) DSC and (b) TGA analysis for CS0 and CSC membranes.

3.5. FT-IR analysis

FT-IR analysis was performed to determine the functional group in the membrane formed between chitosan, starch, and citric acid. FT-IR testing was performed on the CS0 and CSC membranes, where the spectra can be seen in Figure 6.

Figure 6. FTIR spectra of powder chitosan, powder starch, CS0 and CSC membrane.

Figure 6 shows a decrease in the intensity of the absorption peak obtained from the CSC membrane. Li et al. [5] stated that a decrease in peak intensity could be used to indicate the crosslinking formation. The –OH functional groups in CS0 and CSC membranes were detected at wavenumbers around 3250-3500 cm\(^{-1}\) with a strong intensity and broad peaks. The relatively stronger intensity and broader peaks indicated hydrogen bonds forming in the membrane between chitosan and starch. Furthermore, the -NH functional group from chitosan appears in the area 3000-3500 cm\(^{-1}\), overlapping the -OH group spectral band that is widened due to the presence of hydrogen bonds. Stronger intensity is ascribed to the addition of -OH group from citric acid. It is in line with Lusiana et
al. [13], where they reported that the addition of starch and citric acid did not change the type of functional groups in chitosan compounds but only increased the percentage of –OH groups. Absorption peaks at 2922 and 2926 cm\(^{-1}\) indicate the presence of –CH groups on the CS0 and CSC membranes, respectively. The specific difference between the modified chitosan spectra can be observed at the sharp absorption at the wavenumber 1597 cm\(^{-1}\), which indicates a modification of the primary amine group to the secondary amine.

3.6. Scanning Electron Microscope (SEM) analysis
SEM analysis was performed to determine the surface morphology of the upper, lower, and cross-section parts of the membrane. The results of the SEM morphological analysis can be seen in Figure 7.

![Figure 7](image)

**Figure 7.** The SEM images of (a) CS0 and (b) CSC membranes on the top surface, cross-section and bottom surface.

Figure 7 shows that the upper surface of the CS0 membrane is smoother than the CSC membrane, which shows a coarser surface, as well as the lower surface of the membrane which shows the same conditions. The difference in the surface structure is due to the effect of adding citric acid to the membrane as a cross-linking agent. In the cross-section, the CS0 membrane looks coarser than the CSC membrane. Membranes with surfaces with different top and bottom surface structures fall under a category of asymmetric membranes. An asymmetric membrane is a membrane that has a structure and pore size that are not uniform.

4. Conclusion
The chitosan membrane was modified with the addition of starch, followed by a cross-linking reaction using citric acid. The uncross-linked membrane had a tensile strength of 11.07 kgf/mm\(^2\), while the cross-linked membrane had better tensile strength (66.07 kgf/mm\(^2\)). The DSC results of the uncross-linked and cross-linked membrane showed endothermic and exothermic peaks at different temperatures. The membranes’ DTA thermogram showed that the decomposition process on the uncross-linked membrane occurred at 277.74 – 363.27 °C, while in the cross-linked membrane, the decomposition point occurred at 287.83 – 356.38 °C. The FT-IR spectra confirmed a decrease in the intensity of the absorption peak associated with the formation of cross-links on the membrane.
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6. References
[1] Saiful, Riana U, Marlina, Ramli M, Mahmud N 2019 IOP Conf. Ser. Earth Environ. Sci. 273 012047.
[2] Shakeri A, Salehi H, Rastgar M 2017 Carbohydr. Polym. 174 658-68.
[3] Saiful, Riana U, Marlina, Ramli M, Mahmud N 2018 Aceh Int. J. Sci. Technol. 7(8).
[4] Huang Y, Sun J, Wu D, Feng X 2018 Sep. Purif. Technol. 207 142-150.
[5] Li H, Gao X, Wang Y, Zhang X, Tong Z 2013 Int. J. Biol. Macromol. 52 275-279.
[6] Xie Y, Zhang B, Li M N, Chen H Q 2019 Food Chem. 289 187-194.
[7] Chen Y, Qian J, Zhao C, Yang L, Ding J, Guo H 2019 Eur. Polym. J. 118 17-26.
[8] Atangana E, Chiweshe T T, Roberts H 2019 J. Polym. Environ. 27 979-95.
[9] Mukoma P, Jooste B R, Vosloo H C M 2004 J. Power Sources. 136 16-23.
[10] Saiful, Rahmah Z, Ajrina M, Marlina, Rahmi 2020 Rasayan J. Chem. 13 2062-73.
[11] Saiful, Helwati H, Saleha S, Iqbalsyah T M 2019 IOP Conf. Ser. Mater. Sci. Eng. 523 012015.
[12] Prochaska K, Konował E, Sulej-Chojnacka J, Lewandowicz G 2009 Colloids Surf. B Biointerfaces 74 238-243.
[13] Lusiana R A, Siswanta D, Mudasar M 2018 Indonesian J. Chem. 16 144-150.
[14] Gohil J M, Bhattacharya A, Ray P 2006 J. Polym. Res. 13 161-169.