International Journal of Nanoelectronics and Materials
IJNeaM
ISSN 1985-5761 | E-ISSN 2232-1535

Application of the extracted microcrystalline cellulose from pineapple leaf fibers for wound dressing materials

Dania Khoirun Nisa [a], Ariadne Lakshmidevi Juwono [b], Ismail Budiman [c], and Yessie Widya Sari [a] *

[a]*Faculty of Mathematics and Natural Sciences, IPB University, Indonesia*
[b]*Faculty of Mathematics and Natural Science, Universitas Indonesia, Indonesia*
[c]*Biomass and Bioproducts Research Center, BRIN, Indonesia*
** Corresponding author. E-mail: Yessie.sari@apps.ipb.ac.id*

Received 17 December 2024, Revised 20 March 2025, Accepted 09 April 2025

ABSTRACT

The wound healing process involves replacing damaged tissue through a series of cellular events. Microtechnology has the potential to provide the specific physicochemical properties and biological responses necessary to facilitate and support the wound healing process. Many natural biopolymers have been developed in the design of wound dressings. Hydroxyapatite and collagen exhibit antibacterial effects, anti-inflammatory and hydrophilic properties that can support wound treatment and prevent infection. Meanwhile, cellulose has been developed a lot regarding the utilization of its mechanical properties. This research aims to analyze the effect of variations in microcrystalline cellulose (MCC) composition on the physical and mechanical properties of HA/collagen composite scaffolds. The samples obtained were characterized by XRD, SEM, EDS, and UTM. The cellulose sample showed a diffraction peak at 21.7°, which increased significantly in the MCC spectrum. The intensity value was higher after going through the hydrolysis process, indicating that the hydrolysis process was able to increase the MCC intensity with a crystallinity index reaching 70.2%. The addition of MCC in the HA/collagen composite scaffold had an impact on increasing the strain value by 63.77% and relatively reducing the fiber diameter and membrane porosity.

Keywords: *Collagen, Composite, Hydroxyapatite, Microcrystalline cellulose, Wound dressing*

1. INTRODUCTION

The need for wound dressing materials increases every year along with increasing cases of tissue damage. The development of material technology is an effort to restore the function of damaged tissue. Biomaterials are materials in medical applications that have been engineered to interact with biological systems. Biomaterials interact with living tissues or organisms with the aim of repairing, replicating, or replacing the function of damaged or lost tissues/organs [1]. Nowadays, the development of material technology is expected to accelerate, including wound covering membranes (scaffolds). Apart from functioning to protect new tissue, scaffolds are also expected to speed up the wound healing process [2].

Scaffolds can be made from various types of materials, including ceramics, polymers, or a combination of both (composites). Scaffolds made from composite materials show better properties and performance compared to scaffolds made from organic materials alone. Scaffolds made from composites have better mechanical strength, cell proliferation, bioactivity, and hydrophilicity [3]. Hydroxyapatite (HA) is an active biomaterial that has now been widely developed. HA can be obtained from various sources, one of which is eggshells. Egg shells are a source of Calcium (Ca) in the form of Calcium carbonate ($CaCO_3$) [4]. Chicken eggshells contain around 94% $CaCO_3$, the high $CaCO_3$ content in eggshells has the potential to be used as a matrix in the synthesis of biomaterials. The results of research that have been carried out show that the use of $CaCO_3$ can improve the mechanical properties of composites with a polymer matrix [5].

Hydroxyapatite in wound coating membrane applications can be increased in effectiveness by adding reinforcing polymers [6]. Pineapple leaves are the most waste produced from pineapple farming, which is around 70% at each harvest [7]. Pineapple leaf fiber has a fairly high cellulose content, namely between 69.5% and 71.5%. Pineapple leaf cellulose can be extracted using several methods, including alkaline hydrolysis, acid hydrolysis, alkaline methods, and enzymatic methods. The alkaline method is an effective and efficient method used in cellulose extraction. In this research, NaOH was used in the cellulose delignification process.

Cellulose is a polymer material that can be used as reinforcement and improves the mechanical properties of composite scaffolds [8, 9]. Cellulose has biodegradable, antimicrobial, and antioxidant properties, which can help prevent infection and speed up the wound healing process [2, 10]. Previous research included biocomposites with the addition of pineapple leaf fiber as the filler [11]. The test

results indicated that the highest tensile strength value was 17.11 MPa at the addition of 25% w/w of cellulose, while the lowest tensile strength value was 1.99 MPa at the addition of 10% w/w of cellulose. This indicates that the tensile strength will increase along with the weight percentage of cellulose [11].

Microcrystalline cellulose (MCC) is a microparticle crystal isolated from cellulose and has potential as a biomaterial reinforcement. Smaller sizes have the potential to provide greater surface area. Microcrystalline cellulose has a small size, large surface area, and a relatively high level of crystallinity and modulus young. Isolation of microcrystalline cellulose can be carried out by carrying out acid hydrolysis of the amorphous part of the cellulose fiber, leaving micro-sized crystalline domains [12].

Collagen is a material that is biocompatible, biodegradable, easily absorbed by the body, non-toxic, and relatively stable [13]. The results showed that the collagen-HA composite increased the percentage of live cells in the cytotoxicity test. This proves that using collagen and hydroxyapatite together is beneficial in terms of cell growth. HA-collagen composites when implanted in the human body show better osteoconductive properties compared to monolithic HA [14]. Based on the results of mechanical properties tests with the hydroxyapatite/chitosan/collagen composite composition, it was obtained that the best sample with a composition of 7:1.5:1.5 had the highest compressive strength, namely 219.77 kPa. A sample with a composition of 7:2:1 only has a compressive strength of 155.31 kPa [15].

The electrospinning technique is a method for making composite scaffolds that can produce fibers in nano to micro sizes [16]. The advantage of electrospinning lies in the very small fiber size, resembling the structure of natural collagen fibers in skin tissue so that it can increase contact with the skin surface, as well as facilitate cell proliferation and tissue healing [17]. The fiber structure in the resulting scaffold can support the cell growth process due to the formation of an extracellular matrix structure.

Pineapple leaf fiber cellulose as a reinforcing polymer in composite scaffolds has not been widely used in previous research. The proposed research will investigate the characteristics of cellulose produced from pineapple leaf fibers and the effect of adding cellulose as reinforcement and HA/collagen as a matrix in composite scaffolds using the electrospinning method. Sample characterization includes the Scanning Electron Microscope (SEM) test to determine diameter and morphology, X-Ray Diffraction (XRD) test to determine the fiber phase structure, the optical microscope test, the proximate test, and the tensile test.

2. METHODOLOGY

2.1. Materials

The materials used are 60 mesh dried pineapple leaves, sodium hydroxide (NaOH), hydrogen peroxide (H_2O_2), sulfuric acid (H_2SO_4), acetic acid (CH_3COOH), polyvinyl alcohol (PVA), hydroxyapatite, collagen, and deionized water.

2.2. Preparation of MCC from Pineapple Leaf Fibers

Pineapple leaf fibers were cleaned using running water to separate dirt or other substances mixed with the fibers. Next, the pineapple leaf fibers are dried using an oven. The process of grinding pineapple leaf fiber was conducted using a grinding machine or fiber cutter. Pineapple leaf fibers were crushed and sieved until they reached a size of 60 mesh [18]. At this stage, the sample in powder form was subjected to a proximate test to determine its chemical content.

Cellulose isolation was carried out by modifying the alkaline method [8]. Dry pineapple leaf powder measuring 60 mesh was mixed with 10% w/v NaOH in a ratio of 1:8. The delignification process uses a 10% w/v NaOH solution and was heated at 112°C for 60 minutes. Next, the delignification product was filtered using a filter cloth and washed with distilled water. Next, the delignification product was dried in an oven at 80°C with a power of 400 W until a constant weight was obtained. The bleaching process was carried out by reacting the delignification product with 10% v/v H_2O_2, then the sample was heated at 60°C for 90 minutes, then filtered, and the precipitate was washed with distilled water until the pH was neutral. The bleached samples were dried in an oven at 80°C until a constant weight was obtained.

Microcrystalline cellulose was made by modifying the acid hydraulic method [8]. Acid hydrolysis was carried out based on the ratio of cellulose to sulfuric acid (H_2SO_4) 1:20. Cellulose was dissolved in a 60% v/v H_2SO_4 solution, then put into a beaker and stirred and heated using a magnetic stirrer at a temperature of 50°C for 5 hours. Next, the solution was cooled to room temperature, and 2 times the amount of acid was added with distilled water. Centrifugation was carried out at 5000 rpm for 10 minutes to remove any remaining acid. After that, an ultrasonication process was carried out for 10 minutes to break down the particle sizes into smaller ones. Next, the sample was centrifuged again to separate the liquid from the sediment. Wet microcrystalline cellulose was dried in an oven at 60°C until a constant weight was obtained. At this stage, the sample in powder form is subjected to an XRD test.

2.3. Fabrication of MCC/HA/Collagen Composite Scaffold

The composite scaffold manufacturing process was carried out repeatedly with the MCC components varied (0, 0.5, 1.0, 1.5, and 2.0)w/v. HA and collagen solutions were mixed with a ratio of 1:1 and a PVA concentration of 10% w/v. Table 1 is a modification of the composition variations of MCC/HA/collagen in making the composite.

Scaffolds MCC/HA/collagen composite was produced using the electrospinning method. The homogenized MCC/HA/collagen solution was inserted into an injection equipped with a syringe. The needle was connected to a positive voltage source while the low voltage source

Table 1. Variations in MCC/HA/collagen compositions

Sample code	PVA (%w/v)	MCC (%w/v)	HA (%w/v)	Collagen (%w/v)
A	10	-	2.00	2.00
B		0.5	1.75	1.75
C		1.0	1.50	1.50
D		1.5	1.25	1.25
E		2.0	1.00	1.00

(ground) was connected to a collecting plate as a container for the fibers produced from electrospinning. The electrospinning voltage was 12 kV with a distance from the tip of the syringe to the collector wire of 8 cm. The MCC/HA/collagen solution was passed through a spinneret (jet) hole with a diameter of 0.1 mm and a spray speed on the pump of 1.08 μL/hour. The solution that has been induced by an electric charge under the influence of the electric field will jump towards the electrode with the opposite charge and be accompanied by the process of evaporation of the polymer solvent so that only the polymer fiber remains on the collecting plate.

2.4. Physical Characteristics and Morphology

2.4.1. Crystalline Analysis

The crystalline analysis of the MCC samples was examined using X-ray Diffraction (XRD, PANalytical AERIS). Characterization using XRD was carried out to identify the phases, lattice parameters, and degree of crystallinity contained in the samples. The tested sample is exposed to X-rays at an angle of 2θ from 5° to 90°. Samples in powder form are compacted and leveled on the available holder. The XRD source is Cu K-alpha with a wavelength of 1.54060 Å. The crystal size can be calculated using the Debye-Scherrer equation as in Equation (1):

$$L = \frac{K\lambda}{\beta \cos\theta} \times 100\% \quad (1)$$

where L is crystallite size (nm), λ is the X-ray wavelength, β is the full width half maximum (FWHM, in rad), and θ is the corresponding Bragg angle.

2.4.2. Morphology Structures

Composite scaffold morphology testing was carried out using an optical microscope. The test was carried out by cutting the membrane measuring 1 cm × 1 cm, then placing the sample on the preparation, after which it was observed using a microscope so that the surface structure of the membrane could be seen.

2.4.3. Scanning Electron Microscopy and Energy Dispersive X-Ray Spectroscopy

The surface morphology and the elemental composition of the cellulose and MCC samples was examined by using Scanning Electron Microscopy (SEM, Quattro S). The sample specimens were coated with gold (30 μm thickness) in an automated sputter coater. The composite scaffold sample to be tested is placed on an aluminium plate and then observed using SEM at 5000x magnification. The acceleration potential is 20 kV. Characterization using SEM was carried out to determine the surface morphology and diameter of the MCC/HA/collagen scaffold composite.

2.5. Mechanical Properties of MCC/HA/Collagen Composite Scaffold

2.5.1. Tensile Test

Determination of the mechanical properties of MCC/HA/collagen composite scaffolds was carried out using a Universal Testing Machine (UTM, Shimadzu 10 kN). This test aims to determine mechanical properties such as strength, elasticity, stiffness, and plasticity. The tool speed is 2 mm/minute [19]. The results obtained from the tensile test are the stress value (σ), elongation at break (ε), so that the tensile strength and elongation values are obtained.

3. RESULTS AND DISCUSSION

3.1. Proximate Test of Pineapple Leaf Fibers

Proximate testing was carried out to analyze the contents that make up pineapple leaf fiber before cellulose extraction. The chemical composition value is needed to identify the properties possessed by the material. Pineapple leaf fiber, which has been analyzed proximately contains ash, crude protein, crude fiber and crude fat. Table 2 shows the chemical composition of 60 mesh pineapple leaf fiber.

3.2. Morphology and Physical Properties of Cellulose and Microcrystalline Cellulose

3.2.1. Morphology Structures

Optical microscopy (OM) analysis aims to observe the surface appearance of cellulose, microcrystalline cellulose (MCC), and the resulting composite scaffold. Observations carried out using an optical microscope with 1000x magnification are shown in Figure 1. Changes during the hydrolysis process can be identified by observing the morphology before and after the hydrolysis process is carried out.

Figure 1 shows the surface appearance of cellulose and microcrystalline cellulose (MCC). Observation results in Figure 1 (left) of cellulose obtained through the bleaching process show that not all of the cellulose fibers have been defibrillated into microcellulose fibrils; some of the

Table 2. Chemical composition of pineapple leaf fibers

Chemical composition	%Composition
Ash	9.03±0.01
Crude protein	9.58±0.03
Crude fiber	23.83±0.01
Crude fat	1.58±0.02

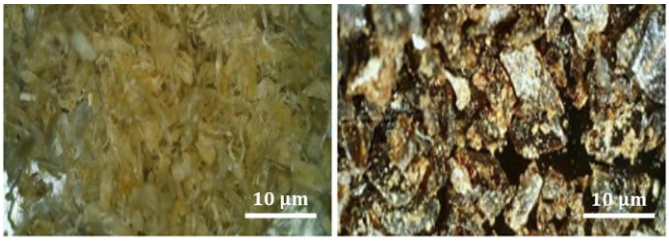

Figure 1. Surface morphology of cellulose (left) and microcrystalline cellulose (right) using optical microscope 1000x magnification

cellulose fibers are still united to form bundles. Further mechanical processes need to be carried out to obtain fibers in microcellulose size. Figure 1 (right) displays microcrystalline cellulose, which appears to have formed crystals, where under controlled conditions hydrolysis can remove the amorphous areas of the cellulose fibers and leave the crystalline areas.

3.2.2. Crystalline Structures

The XRD diffractogram obtained for a cellulose sample from pineapple leaf fiber is shown in Figure 2. The cellulose sample shows a diffraction peak at 12.0°, 16.4°, and 21.7°. The diffraction peak at 21.7° increased significantly in the MCC spectrum, indicating a larger cellulose crystalline area in the sample. Similar results were obtained in other studies, the cellulose diffraction pattern peaked at a 2θ angle of about 11.7°; 17.3°; and 21.0° [20]. The intensity value of cellulose is higher after going through the hydrolysis process, so it can be concluded that the hydrolysis process is able to increase the intensity value of microcrystalline cellulose (MCC). XRD characterization at this stage is used to determine initial conclusions about cellulose which will be characterized further.

The crystal structure of cellulose has a lattice parameter ac, with a = 7.87; b = 10.31; and c = 10.13 [21]. The X-ray diffraction results from MCC show agreement with JCPDS 03-0226 data, at an angle of 2θ = 20.6° with the diffraction plane (0 0 2). The value of the degree of crystallinity of MCC was obtained at 70.2% using Origin. These results are not much different from similar research; the extraction of

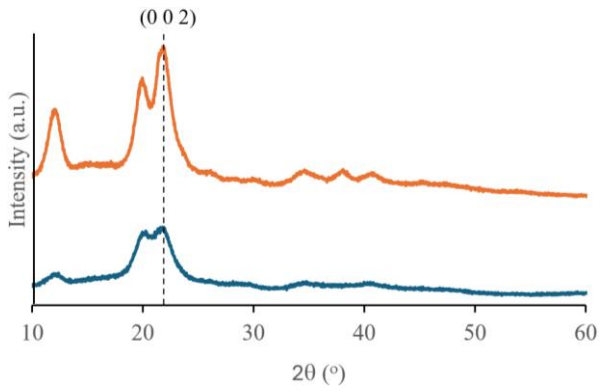

Figure 2. XRD patterns of cellulose and microcrystalline cellulose (MCC)

microcrystalline cellulose obtained a crystallinity index of 72.53% [22]. The increase in the crystallinity value of the MCC sample after going through the hydrolysis process was caused by the amorphous area containing the crystalline part having dissolved during acid hydrolysis and releasing more individual crystals [23].

The crystal size was calculated using the Debye-Scherrer equation, the FWHM (β) value was obtained using the Match application. The average crystal size of all cellulose peaks was 113.55±0.06 nm and the MCC was 86.68±0.03 nm. The average crystallite size of cellulose samples is larger than that of microcrystalline cellulose (MCC). The decrease in crystallite size can be attributed to the acid hydrolysis reaction process which breaks down the cellulose layer and hydrolyzes the cellulose crystals into smaller ones. Removal of amorphous regions increases crystallinity and decreases crystallite size [23].

The XRD results show that the MCC crystallinity is more than 50%, namely 70.2%, and the average crystallite size of all MCC peaks is less than 100 nm, namely 86.6±0.03 nm. The crystallinity index and measurements obtained show that hemicellulose and lignin in the amorphous part of cellulose have been successfully separated from the crystalline part of cellulose through alkaline treatment, bleaching and acid hydrolysis. High cellulose crystallinity is more effective in producing materials of good quality and high strength.

3.2.3. Surface Morphology

Scanning electron microscopy (SEM) analysis was used to identify the surface morphology of cellulose and microcrystalline cellulose (MCC). The electron source is fired at the sample, then the sample scatters electrons based on its electrical properties. Coating with gold metal needs to be done because the sample is not conductive. The image formed shows the structure of the sample being tested. SEM characterization of cellulose using 5000x magnification. Figure 3(A) shows an SEM image of cellulose from pineapple leaf fiber. Based on the SEM image results after chemical treatment, it appears that most of the pineapple leaf fiber cellulose is still united to form bundles, not yet completely defibrillated into small cellulose fibrils. The hydrolysis process needs to be carried out to obtain fibers in microcellulose size. In this research, the diameter of the cellulose fibers produced varied, ranging from 2 µm to 11 µm.

SEM characterization of microcrystalline cellulose (MCC) using 5000x magnification. Figure 3(B) shows the MCC morphology of pineapple leaf fibers. SEM image analysis after chemical treatment, it is known that the MCC of pineapple leaf fibers has an irregular surface. These results are consistent with previous studies. SEM imaging revealed that the analyzed material exhibited an irregular shape and a polydisperse particle size distribution. These results are consistent with previous studies. SEM imaging revealed that the analyzed material exhibited an irregular shape and a polydisperse particle size distribution [24]. In this study,

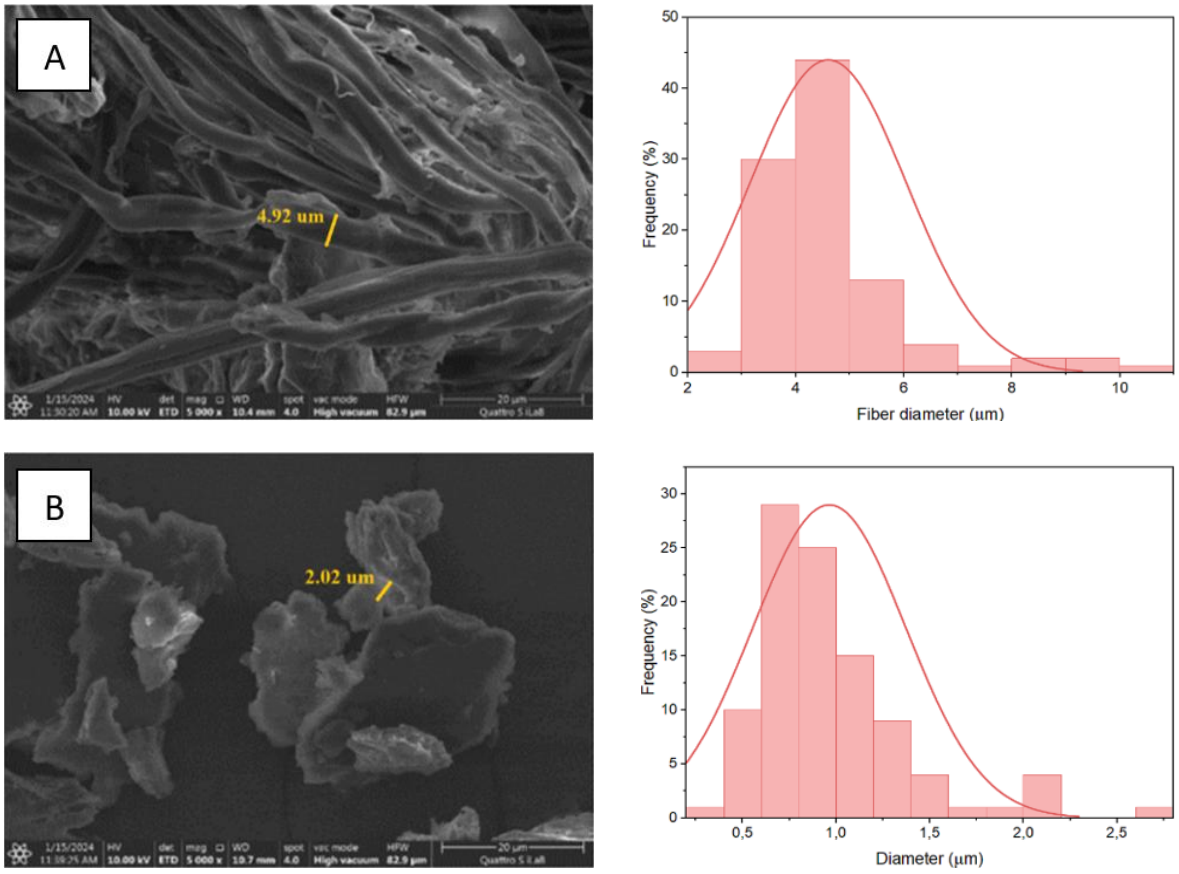

Figure 3. SEM image of (A) cellulose and (B) microcrystalline cellulose (MCC) of pineapple leaf fiber

the size of the MCC particles produced varied, ranging from 0.1 µm to 3 µm. Based on the MCC particle size, it can be concluded that the cellulose produced in this study includes microcellulose crystals.

3.3. Characterizations of Morphology and Mechanical Properties of MCC/HA/Collagen Composite Scaffold

3.3.1. Optical Microscopy

Figure 4 shows the surface morphology of the HA/collagen scaffold composite obtained from optical microscope. The figure confirmed that fibers have been formed. The intertwined fiber pattern, indicating that the fiber-based material has been formed. This structure can indicate that the fabrication process of electrospinning has been successful. Figure 4 is the composite scaffold (HA/collagen) under 1000x microscope magnification, without a wire collector (left) and with a wire collector (right).

3.3.2. Surface Morphology

SEM characterization of composite scaffolds using 10000x magnification. Figure 5(A) shows the morphology of the composite scaffold (HA/collagen). SEM observation results show that after fabrication it appears that the resulting composite scaffold has formed fibers. However, some parts of the composite scaffold still appear to have beads, this could be caused by the polymer being thrown off during the electrospinning process. In this study, the fiber diameter of the sample A scaffold (HA/collagen) produced varied, ranging from 0.05 µm to 0.4 µm.

Figures 5(B), (C), (D), and (E) show the morphology and fiber diameter size distribution of composite scaffolds (MCC/HA/collagen) with MCC concentrations (0.5; 1.0; 1.5; 2.0)% w/v. The varying fiber diameters of samples B, C, D, and E show that there is inhomogeneity in the morphology of the scaffold in some parts, this can be caused by the addition of cellulose with micro- and nano-sized particles. Image processing and MCC particle size measurements show that micro-sized cellulose is in the range of 0.11 to 3 µm, while nano-sized is in the range of 0.05 to 0.1 µm.

The results of SEM visualization show that all composite scaffold samples have formed fibers with quite high porosity. The interconnected porous structure in the composite scaffolds provides space for tissue growth and also facilitates cell motility [25].

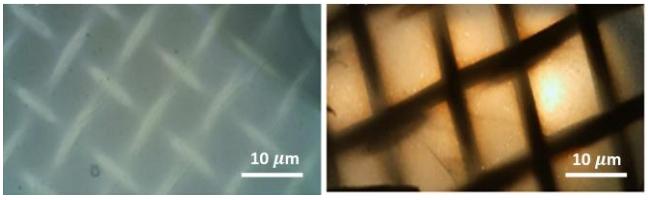

Figure 4. Composite scaffolds (HA/collagen) without wire collector (left) and with wire collector (right) under optical microscope 1000x magnification

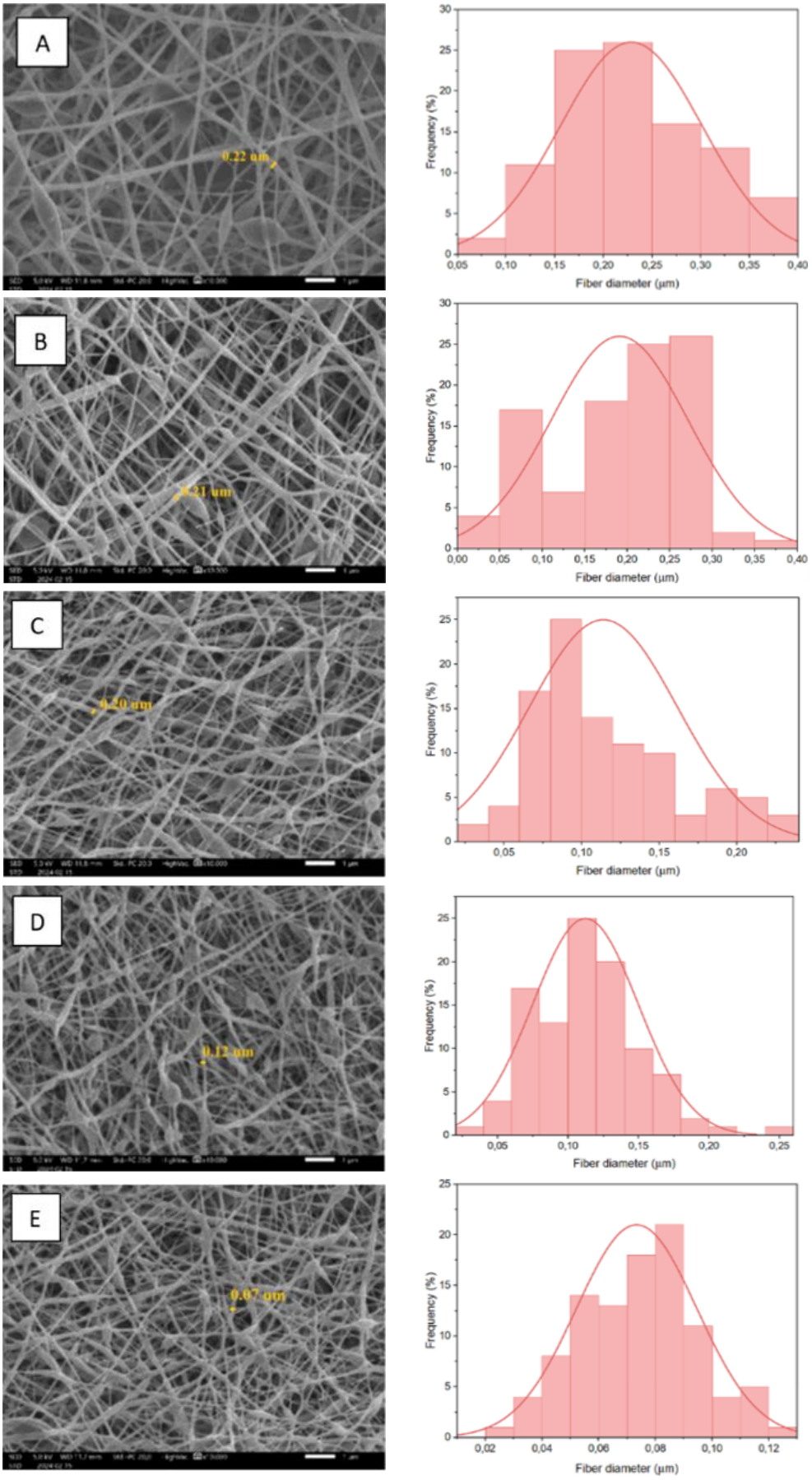

Figure 5. SEM image of scaffold nanofiber (A) HA/collagen, (B) MCC(0.5%)/HA/collagen, (C) MCC(1%)/HA/collagen (D) MCC(0.5%)/HA/collagen, and (E) MCC(0.5%)/HA/collagen

3.3.3. Elemental Analysis

Table 3 showed the EDS results that consist of carbon (C), oxygen (O), calcium (Ca), and phosphorous (P). The mass % content of element C is 61.2%, element O is 35.8%, element Ca is 2.1%, and element P is 0.6%.

Table 3. EDS characterization results of HA/collagen composite scaffolds

Element	Weight%
Carbon (C)	61.2
Oxygen (O)	35.8
Calcium (Ca)	2.1
Phosphor (P)	0.6

3.3.4. Tensile Test

The tensile test is used to obtain tensile strength and max strain values for composite scaffolds with various variations in MCC concentration. Figures 6 and 7 show that the addition of MCC to composite scaffolds has a tendency to result in a decrease in tensile strength values and an increase in breaking strain (max strain). Sample A (HA/collagen) is a scaffold without the addition of MCC and has a tensile strength of 0.79 MPa. This value is higher than composite scaffolds with the addition of MCC.

Scaffolds composed of MCC/HA/collagen composition have a tensile strength value of less than 0.7 MPa. This could be because the addition of PVA polymer with a concentration of 10% w/v causes hydrogen bonds between molecules which contribute to increasing the tensile strength of the scaffold. The tensile strength and breaking strain values for samples A, B, C, and D are significantly different. The cellulose structure provides high strength and stiffness to the scaffold, but the addition of 0.5% and 1% MCC results in decreasing tensile strength values, as seen in Figure 6. This could be because the addition of MCC has exceeded the optimal limit so that the resulting scaffold is brittle (brittle, brittle). This resulted in a decrease in the tensile strength value of 11.91% and the addition of a higher MCC variation was no longer optimal [26].

MCC increases the flexibility of the composite scaffold as shown by the increase in strain at break in Figure 7. Sample D is a composite scaffold with the highest MCC concentration added, namely 1.5% and resulting in an increase in strain value of 63.77%. The addition of cellulose to the membrane can increase the maximum value of strain that can be tolerated before failure occurs. This is because cellulose is a natural fiber polymer that is flexible and pliable, so it can increase the elasticity of the scaffold, allowing it to stretch further before reaching the breaking point. Apart from that, cellulose also functions as a reinforcement in the material matrix, increasing the strength and resistance of the membrane to stretching [9]. Cellulose acts as a structural component of the membrane, allowing greater elongation before failure (reaching the breaking point).

The stress-strain curve obtained from tensile strength measurements is shown in Figure 8. Sample A shows an initial linear elastic region that is quite steep compared to the other three samples, indicating a fairly high modulus of elasticity (Young's modulus). The entire sample also has a non-linear region that represents plastic deformation, where the sample shows a lower slope of the stress-strain curve until the breaking stress is reached. The energy that the membrane can absorb until it breaks can be determined based on the modulus of resilience value. The calculation is carried out by integrating the area under the stress-strain curve. Figure 9 visualizes that the addition of MCC tends to increase the modulus of resilience value of the resulting composite scaffold. Sample D with the addition of the highest MCC concentration (1.5%), shows a higher modulus of resilience value compared to samples A, B, and C. Other research results show that the polymer-hydroxyapatite (pHA) composite has a higher strength value than pure hydroxyapatite (HA) [25].

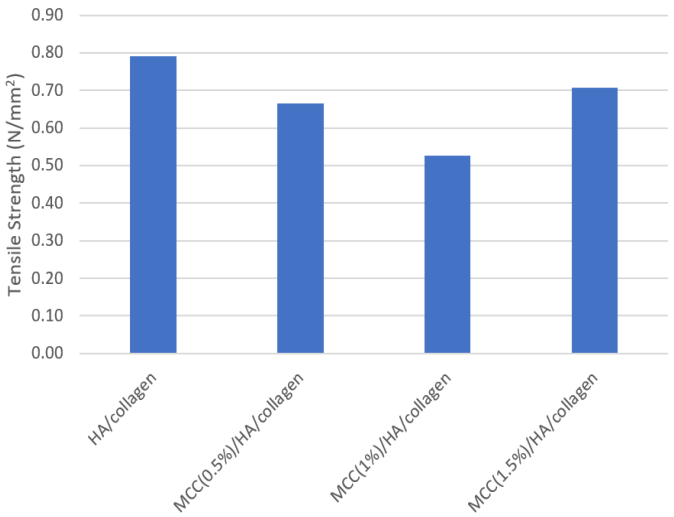

Figure 6. Tensile strength of scaffold nanofiber

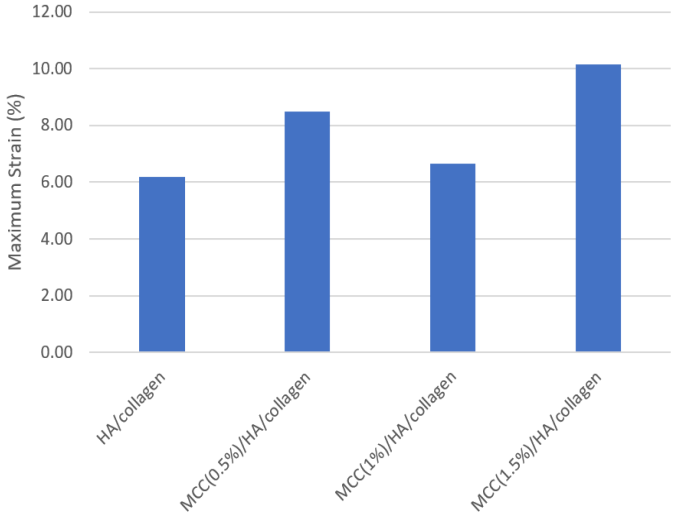

Figure 7. Maximum strain of scaffold nanofiber

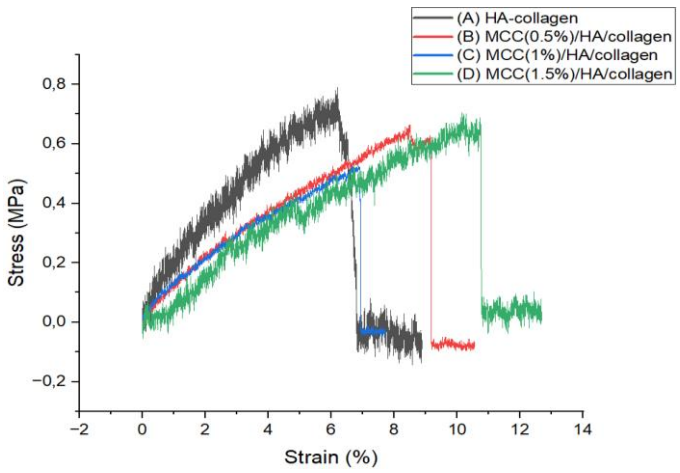

Figure 8. Stress-strain curves of scaffold nanofiber

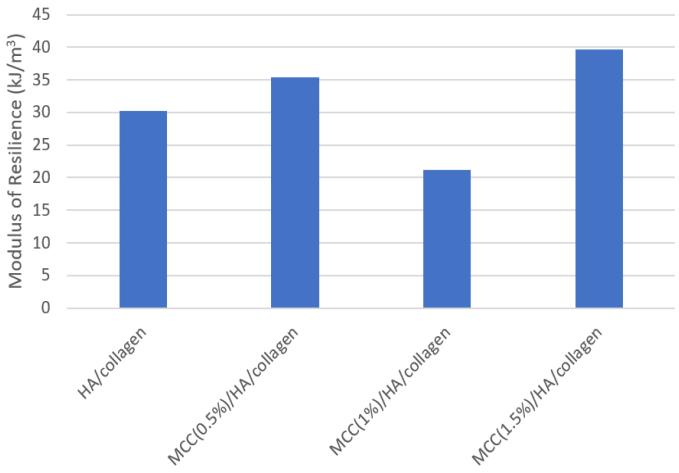

Figure 9. Modulus resillience of scaffold nanofiber

4. CONCLUSION

Isolation of cellulose from pineapple leaf fiber waste and fabrication of MCC/HA/collagen composite scaffolds using the electrospinning method have been successfully carried out. The XRD results show that the degree of MCC crystallinity is 70.2% with an intensity value that increases significantly at 21.7°. MCC as a polymer in composite scaffolds tends to increase the strain value, as well as reduce the fiber diameter and resulting porosity. The addition of MCC has an impact on decreasing the tensile strength value by 11.91% and increasing the strain value by 63.77%. Based on this research, the physical and mechanical properties of MCC/HA/collagen composites with a porous structure have the potential to improve the performance of wound coating membranes in clinical applications.

ACKNOWLEDGMENTS

The authors are thankful to INSPIRASI Indonesia-NTU Singapore Institute of Research for Sustainability and Innovation for partially funding the research [Grant Number: 6636/E3/KL.02.02/2023].

REFERENCES

[1] D. BomBač, M. Brojan, P. Fajfar, F. Kosel, and R. Turk, "Review of materials in medical applications Pregled materialov v medicinskih aplikacijah," *RMZ–Materials and Geoenvironment*, vol. 54, no. 4, pp. 471–499, 2007.

[2] V. Jankauskaitė, A. Balčiūnaitienė, R. Alexandrova, N. Buškuvienė, and K. Žukienė, "Effect of Cellulose Microfiber Silylation Procedures on the Properties and Antibacterial Activity of Polydimethylsiloxane," *Coatings*, vol. 10, no. 6, p. 567, Jun. 2020, doi: 10.3390/coatings10060567.

[3] A. K. Elyaderani, M. del C. de Lama-Odría, L. J. del Valle, and J. Puiggalí, "Multifunctional Scaffolds Based on Emulsion and Coaxial Electrospinning Incorporation of Hydroxyapatite for Bone Tissue Regeneration," *International Journal of Molecular Sciences*, vol. 23, no. 23, p. 15016, Nov. 2022, doi: 10.3390/ijms232315016.

[4] Y. W. Sari, A. Saputra, A. Bahtiar, and N. A. Nuzulia, "Effects of microwave processing parameters on the properties of nanohydroxyapatite: Structural, spectroscopic, hardness, and toxicity studies," *Ceramics International*, vol. 47, no. 21, pp. 30061–30070, Nov. 2021, doi: 10.1016/j.ceramint.2021.07.182.

[5] M. Barczewski *et al.*, "Thermo-mechanical and mechanical behavior of hybrid isotactic polypropylene glass fiber reinforced composites (GFRC) modified with calcium carbonate ($CaCO_3$)," *Polymer Engineering & Science*, vol. 60, no. 7, pp. 1588–1603, Jul. 2020, doi: 10.1002/pen.25404.

[6] M. Oprea and S. I. Voicu, "Recent Advances in Applications of Cellulose Derivatives-Based Composite Membranes with Hydroxyapatite," *Materials*, vol. 13, no. 11, p. 2481, May 2020, doi: 10.3390/ma13112481.

[7] U. Das, "Organic Pineapple Production Technology in Tripura – The lone AEZ for Fruits in North East India," *International Journal of Agriculture Environment and Biotechnology*, vol. 14, no. 2, Jun. 2021, doi: 10.30954/0974-1712.02.2021.4.

[8] F. Fitriani *et al.*, "Isolation and Characterization of Nanocrystalline Cellulose Isolated from Pineapple Crown Leaf Fiber Agricultural Wastes Using Acid Hydrolysis," *Polymers*, vol. 13, no. 23, p. 4188, Nov. 2021, doi: 10.3390/polym13234188.

[9] H. Amirulhakim, A. L. Juwono, and S. Roseno, "Isolation and characterization of Cellulose Nanofiber from Subang Pineapple Leaf Fiber waste produced using Ultrafine Grinding method," *IOP Conference Series: Materials Science and Engineering*, vol. 1098, no. 6, p. 062067, Mar. 2021, doi: 10.1088/1757-899X/1098/6/062067.

[10] R. Priyadarshi and J.-W. Rhim, "Chitosan-based biodegradable functional films for food packaging applications," *Innovative Food Science & Emerging Technologies*, vol. 62, p. 102346, Jun. 2020, doi: 10.1016/j.ifset.2020.102346.

[11] A. F. A. Hamzah, M. H. Hamzah, H. C. Man, N. S. Jamali, S. I. Siajam, and M. H. Ismail, "Recent Updates on the Conversion of Pineapple Waste (Ananas comosus) to Value-Added Products, Future Perspectives and

Challenges," *Agronomy*, vol. 11, no. 11, p. 2221, Nov. 2021, doi: 10.3390/agronomy11112221.

[12] E. Y. Wardhono, N. Kanani, and A. Alfirano, "A simple process of isolation microcrystalline cellulose using ultrasonic irradiation," *Journal of Dispersion Science and Technology*, vol. 41, no. 8, pp. 1217–1226, Jun. 2020, doi: 10.1080/01932691.2019.1614947.

[13] Y. Zheng, Y. Li, C. Ke, X. Gao, Z. Liu, and J. Chen, "Comparison of Structural and Physicochemical Characteristics of Skin Collagen from Chum Salmon (Cold-Water Fish) and Nile Tilapia (Warm-Water Fish)," *Foods*, vol. 13, no. 8, p. 1213, Apr. 2024, doi: 10.3390/foods13081213.

[14] J. Shi *et al.*, "Frontiers of Hydroxyapatite Composites in Bionic Bone Tissue Engineering," *Materials*, vol. 15, no. 23, p. 8475, Nov. 2022, doi: 10.3390/ ma15238475.

[15] C. Zanca *et al.*, "Behavior of Calcium Phosphate–Chitosan–Collagen Composite Coating on AISI 304 for Orthopedic Applications," *Polymers*, vol. 14, no. 23, p. 5108, Nov. 2022, doi: 10.3390/polym14235108.

[16] S. Bongiovanni Abel, F. Montini Ballarin, and G. A. Abraham, "Combination of electrospinning with other techniques for the fabrication of 3D polymeric and composite nanofibrous scaffolds with improved cellular interactions," *Nanotechnology*, vol. 31, no. 17, p. 172002, Apr. 2020, doi: 10.1088/1361-6528/ab6ab4.

[17] T. J. Sill and H. A. von Recum, "Electrospinning: Applications in drug delivery and tissue engineering," *Biomaterials*, vol. 29, no. 13, pp. 1989–2006, May 2008, doi: 10.1016/j.biomaterials.2008.01.011.

[18] S. Gnanasekaran, Y. Y. Li, J. H. Shariffuddin, and N. I. A. Ahamad Nordin, "Production of cellulose and microcellulose from pineapple leaf fibre by chemical-mechanical treatment," *IOP Conference Series: Materials Science and Engineering*, vol. 991, no. 1, p. 012055, Dec. 2020, doi: 10.1088/1757-899X/991/1/012055.

[19] R. P. Sari, U. Ulyarti, and S. L. Rahmi, "The Effect of Sorbitol Concentration on The Characterictics of Purple Yam Starch (Dioscorea alata) Based Edible Film," *Jurnal Bio-Geo Material Dan Energi*, vol. 2, no. 1, pp. 8–12, Mar. 2022, doi: 10.22437/bigme.v2i1. 31538.

[20] H. Fouad *et al.*, "Preparation and characterization of carboxymethyl microcrystalline cellulose from pineapple leaf fibre," *Scientific Reports*, vol. 14, no. 1, p. 23286, Oct. 2024, doi: 10.1038/s41598-024-73860-4.

[21] S. Zhu, H. Sun, T. Mu, Q. Li, and A. Richel, "Preparation of cellulose nanocrystals from purple sweet potato peels by ultrasound-assisted maleic acid hydrolysis," *Food Chemistry*, vol. 403, p. 134496, Mar. 2023, doi: 10.1016/j.foodchem.2022.134496.

[22] H. Lu, Y. Gui, L. Zheng, and X. Liu, "Morphological, crystalline, thermal and physicochemical properties of cellulose nanocrystals obtained from sweet potato residue," *Food Research International*, vol. 50, no. 1, pp. 121–128, Jan. 2013, doi: 10.1016/j.foodres.2012.10.013.

[23] N. Samsalee, J. Meerasri, and R. Sothornvit, "Rice husk nanocellulose: Extraction by high-pressure homogenization, chemical treatments and characterization," *Carbohydrate Polymer Technologies and Applications*, vol. 6, p. 100353, Dec. 2023, doi: 10.1016/j.carpta.2023.100353.

[24] R. Gaudreault, T. G. M. van de Ven, and M. A. Whitehead, "Mechanisms of flocculation with poly(ethylene oxide) and novel cofactors," *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, vol. 268, no. 1–3, pp. 131–146, Oct. 2005, doi: 10.1016/j.colsurfa.2005.04.044.

[25] C. Sarkar, K. Anuvrat, S. Garai, S. K. Sahu, and J. Chakraborty, "One pot method to synthesize three-dimensional porous hydroxyapatite nanocomposite for bone tissue engineering," *Journal of Porous Materials*, vol. 27, no. 1, pp. 225–235, Feb. 2020, doi: 10.1007/s10934-019-00805-y.

[26] S. M. Noorbakhsh-Soltani, M. M. Zerafat, and S. Sabbaghi, "A comparative study of gelatin and starch-based nano-composite films modified by nano-cellulose and chitosan for food packaging applications," *Carbohydrate Polymers*, vol. 189, pp. 48–55, Jun. 2018, doi: 10.1016/j.carbpol.2018.02. 012.