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PRODUCING BACTERIAL CELLULOSE FROM OIL PALM SAP AS A WOUND DRESSING MATERIAL

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Abstract

This study aims to utilize oil palm sap into bacterial cellulose (*nata*) with the assistance of *Acetobacter xylinum* bacteria which is then applied as a material for making wound dressings. Bacterial cellulose has a high absorption capacity and is hydrophilic so it supports optimal wound healing. This study used a non-factorial Complete Randomized Design (CRD) method consisting of three treatment stages, namely treatment P1 with a fermentation time of 8 days, treatment P2 with a fermentation time of 10 days, and treatment P3 with a fermentation time of 12 days. The results showed that fermentation time affected the thickness and absorption capacity of the produced bacterial cellulose. Treatment P3 produced bacterial cellulose with an average thickness of 15.8 mm and a better absorption capacity of 3.68 g. All the test results that have been carried out (membrane thickness, absorption capacity, tensile strength and wound dressing quality test) in the P3 treatment concluded that the cellulose produced by the *Acetobacter xylium* bacteria is in the form of pure cellulose which has high mechanical strength and absorption capacity so that it is suitable for use as a wound dressing material.

Keywords: *Acetobacter xylinum,* oil palm sap, wound dressing, bacterial cellulose.

INTRODUCTION

A wound is a condition in which damage occurs to body tissue caused by a blow from a sharp or blunt object, chemical substances, temperature changes, explosions, hot air, or animal bites (Amalia & Purwaningtyas, 2024). The shape of the wound can vary depending on the causative factor, for example, a vulnus scissum or a cut caused by a sharp object. To prevent infection in the wound, antibacterial products are needed to inhibit bacterial growth and accelerate wound healing (Indriyani et al., 2023). According to Sari et al. (2024), wound care can be performed using a wound dressing to cover the wound and prevent bacterial contamination.

A wound dressing is a material used to cover and protect wounds, keeping them clean, preventing infection, and accelerating healing (Suryati et al., 2021). Currently, conventional wound dressings still use materials that can cause wound damage, such as bandages, cotton, and gauze. Mechanically, these materials can adhere to the wound surface, damaging new epithelial tissue and stimulating the growth of external bacteria. Furthermore, these materials can also make the dressing change process uncomfortable. The ideal wound dressing should maintain the necessary moisture conditions for healing and act as a barrier to microorganisms (Liau et al., 2023). According to Naomi et al. (2020), bacterial cellulose can be used for wound healing, including in the manufacture of wound dressings.

Bacterial cellulose is a nanocellulose produced by the bacteria *Acetobacter xylinum* (Potivara & Phisalaphong, 2019). Bacterial cellulose is formed from the degradation of carbohydrates by *Acetobacter xylinum* bacteria. Increasing the amount of carbohydrates in the medium increases the number of bacterial celluloses produced, as does the weight and total cellulose fibers (Melindasari et al., 2025). Cellulose from *Acetobacter xylinum* has high purity and absorption capacity, is hydrophilic, has good permeability, and has unique mechanical properties. Based on these properties, bacterial cellulose can be considered an ideal wound dressing because it creates a moist environment that supports optimal wound healing at every stage. A moist environment can facilitate cell movement and bonding, which will help close the wound (Liau et al., 2023).

Two factors influencing the formation of bacterial cellulose are the carbohydrate source and fermentation time. The carbohydrate source can come from the raw materials used or from the addition of other carbohydrates, such as sucrose, glucose, and fructose (Fernandes et al., 2020). Raw materials frequently used as growth media for *Acetobacter xylinum* include *nata de coco* (nata from coconut water), *nata de pina* (nata from pineapple juice or

pineapple peel), *nata de mango* (nata from mango juice), *nata de soya* (nata from tofu liquid waste), *nata de cacao* (nata from cocoa waste), and others (Santosa et al., 2022). Furthermore, according to Raja et al. (2023) *nata* can also be made from oil palm sap by utilizing old oil palm trunks. This is because oil palm sap contains relatively high levels of sugar, which is the main carbohydrate source for *Acetobacter xylinum* during fermentation. Palm sap is a sweet liquid obtained from unproductive and felled oil palm (*Elaeis guineensis*) plants (Purwandani et al., 2020). According to Agustira et al. (2019), oil palm sap produced in oil palm rejuvenation areas has great potential to serve as a source of income to finance rejuvenation investments and the living costs of smallholder farmers during the rejuvenation process. One felled oil palm tree can produce 10 liters of sap per day for one month.

This sap contains the main sugars sucrose (11.6%), glucose (2.32%), and fructose (1.47%) and has a water content that varies depending on the length and time of tapping. The quality of palm sap is influenced by several factors, such as tapping time, tree age, tapping method, and climate conditions (Widyaningsih et al., 2022). According to Litana et al. (2018), the length of withering and the distance of sap collection can affect the quality of the sap produced. The length of oil palm withering can affect different pH and total acid levels. At the same time, the distance of sap collection can also affect pH, water content, total acid, total dissolved solids, and alcohol content. The processing and total sap production last 5 days in the treatment of oil palm tree trunks withering. The time interval during sap collection affects changes in water quality over time, with the best results obtained on the 15th and 20th days (Rahmaini et al., 2023). This study aims to evaluate the potential of palm sap as an alternative material for the production of bacterial cellulose, a material for wound dressings. The relatively high sugar content of oil palm sap can serve as the primary carbohydrate source for *Acetobacter xylinum* in the fermentation process. This research is expected to provide a solution for utilizing palm oil trunk waste to produce palm sap, which can then be used as an effective and affordable alternative material for wound dressings.

METHOD

Research Design

This research was conducted for one year, from May 2025 to May 2026. This study used a non-factorial completely randomized design (CRD) with 3 treatment levels and 3 repetitions per sample, yielding 9. The experiment consisted of three treatment levels: P1 (fermentation time 8 days), P2 (fermentation time 10 days), and P3 (fermentation time 12 days). The dependent variables observed were the thickness of bacterial cellulose, absorption capacity, tensile strength, and the quality of the wound dressing produced.

Materials and Equipment

Oil palm sap was collected from a local palm oil plantation in Dolok Masihul District, North Sumatra Province. The chemicals used included sodium metabisulfite, 25% acetic acid, *Acetobacter xylinum* starter culture, sucrose, 4% NaOH, potassium bromide, and distilled water. The equipment used included a fermentation tray, a measuring cylinder, a stirrer, a dropper, a beaker, an oven, and bandages.

Procedure

Oil Palm Sap Tapping

Oil palm sap tapping was begun with felling the oil palm tree, followed by clearing the tip of the trunk using an axe or knife until the white, soft-textured palm fronds are exposed. The trunk was left to wither for 4-7 days, followed by tapping it at approximately 0.5 cm per day with a sharp knife to extract the sap. The sap was collected in a bucket. The extracted sap was treated with 0.01% sodium metabisulfite preservative in the bucket, then covered with plastic and palm fronds. During the withering and tapping process, the oil palm trunk was covered with plastic and fronds to protect it from rain, hot sun, insects, and livestock, which can affect sap quality (Agustira et al., 2019).

Bacterial Cellulose Production

One liter of filtered oil palm sap was heated in a saucepan, and 40 g of sucrose, 15 ml of acetic acid, and 10 g of ZA fertilizer were added. The mixture was stirred and heated to a boil. After boiling, the sap was removed, placed in a fermentation tray, and covered with newspaper. After the sap solution had cooled to room temperature, 50 ml of *Acetobacter xylinum* starter solution was added. The fermentation tray was covered again with newspaper and stored at room temperature. The fermentation process was carried out for 8, 10, and 12 days, as specified in the research design. After fermentation was complete, the bacterial cellulose was harvested and rinsed with clean water, then soaked in boiling water for 10 minutes to kill the bacteria. The bacterial cellulose was neutralized in 4% NaOH

for 24 hours, followed by another wash with clean water. After washing, the bacterial cellulose was soaked in boiling water and left in a closed container for one day. This process was repeated until the soaking water became clear, indicating that the bacterial cellulose was ready for analysis. Prior to analysis, the bacterial cellulose was soaked in distilled water for 24 hours.

Measuring Bacterial Cellulose Thickness

The bacterial cellulose, which had been soaked in distilled water for 24 hours, was surface-dried using a tissue. The thickness of the cellulose acetate was measured with a digital calliper to 0.05 mm precision. The measurement results are the average of three data collection sessions (Liau et al., 2023).

Absorption Capacity Test

Absorption capacity testing was conducted using the gravimetric method. Oven-dried bacterial cellulose was cut into 3 × 3 cm pieces and then soaked in distilled water for two hours. Next, the bacterial cellulose was removed, and the surface was gently dried with tissue (Liau et al., 2023). The absorption capacity (g/g) was calculated using equation (1), where w_o and w_t are the masses of bacterial cellulose before and after soaking.

$$\text{Absorption capacity} = \frac{w_t - w_o}{w_o} \quad (1)$$

Tensile Strength Test

Before analysis, bacterial cellulose was dried in an oven for 2-4 hours at 60-65°C. Afterward, the bacterial cellulose was cut into 1.5 × 6 cm pieces and mounted on a tensile tester. The stress value was determined as the maximum force during pulling, while the difference in length after pulling was measured as the elongation of the bacterial cellulose (Liau et al., 2023).

Effectiveness Testing as a Wound Dressing

Oven-dried bacterial cellulose was cut into 3 × 3 cm pieces, then gently dried with tissue (Liau et al., 2023). Afterward, the bacterial cellulose was applied to the hamster's skin wound using a bandage. The skin area was then observed at intervals (3-6 days) to determine the effectiveness of the bacterial cellulose in wound absorption.

RESULTS AND DISCUSSION

Thickness of Bacterial Cellulose

The thickness of bacterial cellulose at various fermentation times is shown in Figure 1. In sequence, the average thickness of bacterial cellulose in treatments P1, P2, and P3 was 9.63 mm, 11.3 mm, and 15.8 mm, respectively. The length of fermentation time can affect the thickness of the bacterial cellulose produced. The longer the fermentation time, the thicker the bacterial cellulose becomes, as Acetobacter xylinum can utilize nutrients in the substrate optimally, thereby increasing cellulose production. This is also in line with the research of Putri et al. (2021), which found that the optimal thickness of bacterial cellulose was achieved at a fermentation time of 14 days.

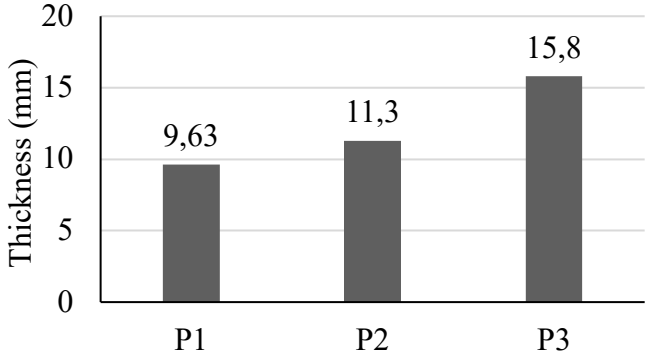

Figure 1. Thickness of bacterial cellulose

Table 1 displays the results of the ANOVA test for bacterial cellulose thickness. The significance value of 0.000 is less than 0.05 ($p < 0.05$), so it can be concluded that the treatments are significantly different (have a significant effect) on bacterial cellulose thickness. Next, a Duncan test was conducted to determine which treatment group had a significant effect on bacterial cellulose thickness, as summarized in Table 2. Based on the results of the Duncan test, it was found that treatment P1 was significantly different from treatments P2 and P3, treatment P2 was

significantly different from treatments P1 and P3, and treatment P3 was significantly different from treatments P1 and P2. In other words, all three treatments had a significant effect on bacterial cellulose thickness.

Table 1. ANOVA test results for thickness of bacterial cellulose

ANOVA					
	Sum of Square	DF	Average Sum	F_{count}	Sig.
Between groups	62.487	2	31.243	57.977	.000
Within groups	3.233	6	.539		
Total	65.720	8			

Table 2. Duncan test results for thickness of bacterial cellulose

Duncan[a]				
Treatment	N	Homogeneous Subsets = 0,05		
		1	2	3
P1	3	9.6333		
P2	3		11.3000	
P3	3			15.8667
Sig.		1.000	1.000	1.000
Means in the same subset are not significantly different.				
a. Uses the harmonic mean sample size = 3.000				

Absorption Capacity of Bacterial Cellulose

Absorption capacity is an important parameter that must be evaluated in wound dressings—the nature of the absorption capacity functions as an absorber of excess exudate in watery wounds. The results of the analysis of bacterial cellulose absorption capacity across three treatment groups are shown in Figure 2. Sequentially, the absorption capacity of bacterial cellulose in treatments P1, P2, and P3 was 2.84 g/g, 3.23 g/g, and 3.68 g/g, respectively. The fermentation time can affect absorption capacity because cellulose produced by the static method forms dense sheets (Sarkono et al., 2015). According to Liau et al. (2023), longer fermentation time results in more cellulose microfibrils forming. The gaps between the microfibrils function to absorb water (exudate) in the wound.

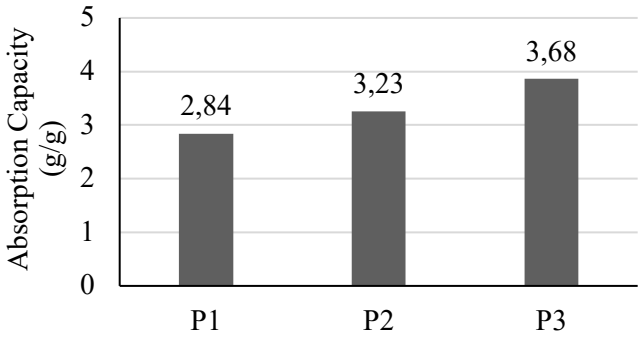

Figure 2. Absorption capacity of bacterial cellulose at various treatments

Table 3 displays the results of the ANOVA test for bacterial cellulose absorption capacity. The significance value was 0.001, which is less than 0.05 ($p < 0.05$). Therefore, it can be concluded that the treatments were significantly different (had a significant effect) on the bacterial cellulose absorption capacity. Next, a Duncan test was conducted (Table 4) to identify significant treatment groups. The Duncan test results showed that treatment P1 was significantly different from treatments P2 and P3, treatment P2 was significantly different from treatments P1 and P3, and treatment P3 was significantly different from treatments P1 and P2. In other words, it can be concluded that all three treatments had a significant effect on the bacterial cellulose absorption capacity.

Table 3. ANOVA test results for absorption capacity of bacterial cellulose

ANOVA					
	Sum of Square	DF	Average Sum	F_{count}	Sig.
Between groups	1.059	2	.530	24.588	.001
Within groups	.129	6	.022		
Total	1.189	8			

Table 4. Duncan test results for absorption capacity of bacterial cellulose

Duncan[a]				
Treatment	N	Homogeneous Subsets = 0,05		
		1	2	3
P1	3	2.8400		
P2	3		3.2367	
P3	3			3.6800
Sig.		1.000	1.000	1.000
Means in the same subset are not significantly different.				
a. Uses the harmonic mean sample size = 3.000				

Tensile Strength of Bacterial Cellulose

Tensile strength analysis is performed to determine the strength and mechanical properties of a film. The quality of the resulting film depends on the material's high tensile strength (Yanti et al., 2021). In this study, tensile strength analysis was only performed on the bacterial cellulose sample with the best thickness and absorption capacity, namely the P3 treatment. Based on the test results, the P3 sample had a maximum force of 97.286 N, a maximum stress of 16.919 MPa, and an elongation of 4.010 mm. These values indicate that bacterial cellulose from oil palm sap has sufficient mechanical properties for biomaterial applications, particularly as a wound dressing.

The mechanical properties of bacterial cellulose are highly dependent on processing conditions and water content. According to de Amorim et al. (2022), bacterial cellulose in a hydrated state has a relatively low tensile strength, 0.3-1 MPa, because its nanofibrillar structure absorbs large amounts of water, reducing the material's stiffness. Medical applications, such as wound dressings, do not require a material with extremely high mechanical strength like structural materials, but they must maintain a balance between strength and flexibility. This is because wound dressings must maintain structural integrity without inhibiting skin tissue mobility and maintain a moist wound environment (Elangwe et al., 2022).

In another study, Masoud et al. (2025) reported that bacterial cellulose-based hydrogels have tensile strengths of 10–50 MPa and elongations of 5–300%, which are considered ideal for medical applications because they maintain structural integrity without inhibiting tissue movement. Compared with the results of that study, the tensile strength of bacterial cellulose in this study falls within the range of medical biomaterial hydrogels (10–50 MPa), making it suitable for non-load-bearing wound dressing applications. Although lower than dry bacterial cellulose, which can reach >100 MPa, this is irrelevant because medical applications require the material to be in a moist (hydrated) state.

Furthermore, the maximum force value of 97.286 N is also consistent with the report by Potivara & Phisalaphong (2019), which states that bacterial cellulose has a maximum tensile strength of 50–120 N, depending on the density of the fibril network. An elongation of 4,010 mm or approximately 6.68% indicates that the material still has sufficient flexibility to follow skin deformation. Ideal wound dressing materials have a minimum elongation of 5-300% (Masoud et al., 2025), so the elongation value in this study is still within the acceptable lower limit.

Quality Test of Bacterial Cellulose as Wound Dressing

Wound dressing quality testing was conducted to assess the effectiveness of bacterial cellulose in direct wound closure. Measurements showed that the width of the wound in the hamster after being bandaged with oil palm sap bacterial cellulose was 0.52 mm on the first day, 0.25 mm on the third day, and the wound was completely closed on the sixth day. Visually, the wound appearance after being bandaged with palm sap cellulose is shown in Figure 3.

(a) (b) (c)

Figure 3. Condition of wounds in hamsters after administration of bacterial cellulose: (a) first day; (b) third day; (c) sixth day

On the first day, the wound was still open with an average wound width of 0.52 mm. This condition indicates the early inflammatory phase, in which bacterial cellulose acts as a wound barrier, maintaining moisture in the wound area and assisting in the absorption of exudate. According to Portela et al. (2019) bacterial cellulose can create a moist wound environment, supporting accelerated healing in the early phase. On the third day, the average wound size decreased to 0.25 mm, a 51.9% reduction from the first day. This decrease indicates the onset of the proliferation phase, which involves wound contraction and new tissue formation. The porous structure and hydrophilic properties of bacterial cellulose facilitate the absorption of wound fluids and oxygen exchange, supporting tissue regeneration (Atykyan et al., 2020). On the sixth day, the wound was completely closed in all replicate observations (100% wound closure), indicating that the epithelialization and early maturation processes were well underway. The relatively rapid wound closure indicates that bacterial cellulose has the potential to be an effective wound dressing material. These results are in line with Tabri's research (Tabri, 2018), which reported that bacterial cellulose-based wound dressings accelerated wound healing compared with conventional dressings. Overall, the test results indicate that bacterial cellulose supports a gradual reduction in wound size to complete closure by the sixth day, suggesting its potential as a wound-dressing biomaterial.

CONCLUSION

This study showed that the optimal fermentation time was 12 days, with bacterial cellulose produced at this time having the highest thickness and absorption capacity compared to the 8-day and 10-day fermentation treatments. Cellulose produced by *Acetobacter xylium* bacteria has high mechanical strength and absorption capacity, making it suitable as a wound dressing material. Experiment as a bandage on hamster skin wounds showed that bacterial cellulose can absorb exudate and protect wounds from bacterial contamination.

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