The Impact of the Mechanical Modification of Bacterial Cellulose Films on Selected Quality Parameters

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Abstract: This study investigated the effect of the homogenization of bacterial cellulose particles and their reintegration into a membrane on the mechanical and physical parameters of the films produced from them in relation to films made of native cellulose (not subjected to the homogenization process). Bacterial cellulose was obtained from a culture of microorganisms forming a conglomerate of bacteria and yeast, called SCOBY. The research has shown that the mechanical modification of bacterial cellulose contributes to an increase in the elongation of the material. Modified polymer films were characterized by a higher Young’s modulus and a much higher breaking force value compared to native cellulose. The mechanical modification of cellulose contributed to an increase in hygroscopicity and changes in water vapor permeability. The obtained results may provide significant information on the methods of modifying bacterial cellulose, depending on its various applications.

Keywords: bacterial cellulose; quality parameters; tensile strength; water vapor permeability; contact angle

1. Introduction

Bacterial cellulose is an exopolymer composed of β 1,4-D glucopyranose units, produced by acetogenic fermentation microorganisms that mainly belong to Komagataebacter, Aerobacter, Achromobacter, and Agrobacterium [1,2]. In contrast to its plant counterpart, it is characterized by better mechanical properties, a greater degree of polymerization, greater crystallinity, and smaller fiber diameters. Bacterial cellulose, unlike its plant counterpart, is devoid of naturally accompanying substances, such as hemicellulose or lignin, whose separation from cellulose requires methods that often affect the structure and properties of the cellulose itself. Since its discovery, bacterial cellulose (BC) has shown tremendous potential as an effective biopolymer in various fields. Many interesting studies indicate the multidisciplinary possibilities of using bacterial cellulose, ranging from medicine, cosmetology, biotechnology, and environmental protection to the paper, clothing, and wood industries [1–6]. Much interesting research concerns the method of modifying bacterial cellulose in order to impart new functions, including surface functions [7–9]. Frone et al. [7] produced a surface treatment of cellulose to increase the barrier properties in regard to moisture penetration, and an interesting method of mechanically modifying the cellulose structure is described by Gao et al. [10]. The authors of the study subjected the polymer to cyclic biaxial stresses to elucidate the mechanisms causing the deformation and cracking of fibrous cellulose. Additionally, bacterial cellulose as a completely natural, ecological, nontoxic, and biodegradable polymer, with very good physical and mechanical parameters, can be an alternative to many products based on synthetic petroleum-derived polymers.
Essential for the potential application of biocellulose are its physical and mechanical properties, which can be modified during its synthesis and after its production. Scientific research shows that the strength properties of cellulose depend on the nutrients of the culture medium [11]. Cellulose obtained from a substrate rich in nitrogen has a multilayer and greater surface ripple, in contrast to a polymer obtained from a substrate poor in nutrients [12]. Vigentini et al. [13] found that an important factor influencing the quality of the synthesized cellulose is the addition of a suitable nitrogen source. Chen et al. [14] analyzed the effect of various saccharide additives on the quality of cellulose synthesized by *Komagataeibacter xylinus*, ATCC 23770 strain. It was found that in the presence of glucose and maltose the highest yield of cellulose synthesis was obtained; however, cellulose produced in the presence of mannose, xylose, and galactose had a larger diameter of fibers and higher crystallinity than cellulose synthesized in a medium with glucose or maltose. Another important factor determining the properties of bacterial cellulose is the drying method and the drying temperature. Stanisławska et al. [15] found that the tensile strength of cellulose dried at 25 °C was 15 times higher than that of cellulose dried at 105 °C. Similar results were obtained by Domskiene et al. [16]. Cazon et al. [17] showed that water contained in cellulose acts as a plasticizer, contributing to changes in strength parameters and changes in water vapor permeability.

Comparing the strength properties of bacterial cellulose, they are much higher than for other commonly used polymers when in the form of a film. Stanisławska et al. [18] provide, as a comparative example, the tensile strength values for cellophane (20–100 MPa) and polypropylene (30–40 MPa), which are definitely lower than the tensile strength values for bacterial cellulose (200–300 MPa).

Undoubtedly, chemical modification has a significant impact on the strength properties, water vapor permeability, and hygroscopicity of bacterial cellulose. By adding various substances to the polymer, desired features and properties can be obtained. However, each time synthetic substances are added to the biopolymer, they may change its toxicity or bioutilization capacity. The effect of chemical modification of bacterial cellulose on its hydrophobic properties, flexibility, and strength has been the subject of numerous scientific papers [19–21]. Indriyati et al. [19] confirmed a significant increase in the hydrophobicity and elasticity of bacterial cellulose by introducing beeswax into the polymer, as well as noticing that the addition of a hydrophobic substance significantly decreased the tensile strength of such a modified polymer. Another example of modifying the properties of bacterial cellulose is the study by Sommer et al. [22]. The authors of that research modified the cellulose-based material used in food packaging by adding montmorillonite, which increased the water vapor permeability of the polymer but at the same time reduced its tensile strength.

The mechanical modification of the polymer, e.g., by grinding and reprocessing, may also cause changes in the quality of its characteristics. Luddee et al. [23], by adding various sizes of ground cellulose fractions to polylactic acid (PLA), showed that with an increase in the size of cellulose particles added to PLA the tensile strength and elongation of the obtained biocomposite decreased. Moreover, the size of cellulose particles in the biocomposite significantly affects the water vapor permeability of the material [18]. Another interesting example of mechanical modification is the fragmentation of bacterial cellulose presented by Yousefi et al. [24]. According to these authors, this process influenced the strength properties of nanopaper. They indicated that the strong fragmentation of bacterial cellulose increases its tensile strength repeatedly and increases the Young’s modulus. Evaluation of the rheological properties of milled bacterial cellulose was carried out by Balquinta et al. [25]. Researchers have shown that the ground polymer had strong gelling properties with a large decrease in viscoelastic modules.

Obtaining bacterial cellulose with appropriate quality parameters is important from the point of view of its applications and technological processes. Beneficial strength properties of bacterial cellulose will favorably affect the mechanical properties of the final product made on the basis of this polymer. Gao et al. [26] showed that the addition of
bacterial cellulose to the paper pulp influenced the paper stiffness and increased the tensile strength of the paper. In another example of research, conducted by Yamada et al. [27], it was proven that the mechanical strength of cellulose varied depending on the method of polymer modification. On the other hand, in the patent application No. P.433630, the authors state that the tensile strength of chipboards for internal layers containing 15 ± 2.5% by weight of bacterial cellulose was not worse than that of a standard chipboard [28]. Unfortunately, in the literature on the subject, there are still few examples of research on the mechanical modification of bacterial cellulose and its impact on the behavior of quality parameters, such as breaking force, elongation, sorption and transport of water vapor, or surface properties.

The aim of this study was to investigate the effect of the mechanical modification of bacterial cellulose through its fragmentation and then reintegration of particles on selected properties of this polymer regarding various applications, including wood-based composites and modified wood surfaces. Verification was performed on bacterial cellulose in the form of a film, based on the analysis of properties such as tensile strength, elongation, Young’s modulus, water sorption, water permeability, and surface properties. The obtained measurement results were compared to the results obtained for native, non-modified cellulose.

2. Materials and Methods

2.1. Samples

Bacterial cellulose was produced by a consortium of bacteria and yeast microorganisms called SCOPY, grown on a substrate containing 10% food sucrose (Krajowa Spółka Cukrowa SA, Toruń, Poland), 0.03% peptone (Biomaxima SA, Lublin, Poland), and 0.05% yeast extract (Biomaxima SA, Lublin, Poland). SCOPY microorganisms were obtained from the organic farm Wolanin (Wolanin, Szczawnik, Poland). The cultivation of the microorganisms was carried out for 14 days in a heat incubator (J.P. Selecta Laboratory Equipment Manufacturer, Barcelona, Spain). The incubation temperature of the culture was 26 ± 2 °C, and the humidity was 66 ± 2%. After the assumed cultivation time, the produced cellulose was removed from the substrate, which was then purified according to the method described by Betlej et al. [11]. For this purpose, the obtained polymer was thoroughly rinsed in a detergent containing 5–15% anionic surfactants and <5% non-ionic surfactants, then rinsed twice with distilled water, then rinsed with 0.1% NaOH (POCH, Gliwice, Poland), and finally rinsed twice with distilled water. Then, the cellulose was washed with 0.1% citric acid (POCH, Gliwice, Poland) and again rinsed twice with distilled water. The washing process was carried out on unmodified cellulose. After the rinsing process was completed, the polymer was weighed and divided into two (identical in weight) parts. One part of the polymer was modified through the fragmentation and then reintegration of particles. For this purpose, one part of the polymer was ground in a shredder, model MMBM401W (Bosch, Gerlingen, Germany), to the form of a paste, as a precursor for film forming. The size of cellulose particles was in the range of 18–30 µm. The cellulose particle size was measured using a Delta Optilac Evolution 100 light microscope (Delta Optical sp. z o. o., Warsaw, Poland), using a 60× and 100× magnification, equipped with a Levenhuk M1000Plus camera (Levenhuk Poland sp. z o.o., Warsaw, Poland). The particle size was measured in at least 2 opposite directions, using the DLTCamViewer program. The produced paste was degassed for 1 h in a vacuum dryer (model 1425, ShelLab, Oregon, USA) with a vacuum of 100 hPa. The degassed mass of cellulose was poured into silicone molds with dimensions of 250 mm × 350 mm in order to obtain a polymer in the form of a thin film. The polymer was spread over the surface of the silicone mold by means of silicone rollers, which simultaneously smoothed the upper surface of the polymer. The second batch of the polymer was left intact. The cellulose, prepared in this way, was dried at a temperature of 24 ± 2 °C in a laboratory dryer (J.P. Selecta Laboratory Equipment Manufacturer, Barcelona, Spain) until a constant mass of the polymer (about 120 h) in the form of a thin film was obtained.
For such prepared cellulose films, their thickness and apparent density were determined. The thickness was measured with an ultrasonic thickness gauge, model Ultrameter AB400 (Metrison Sp. z o.o., Mościska, Poland), with an accuracy of 0.001 mm (±3%). The measurement was performed in ten repetitions. To determine the apparent density, cellulose samples with dimensions of 50 mm × 50 mm were cut, and then their volume and weight were determined. The weight of the cut pieces was determined using a precision balance model 1000.X2 (Radwag, Radom, Poland).

2.2. Tensile Strength Test

Tensile strength tests for bacterial cellulose film were performed on an Instron 5544 testing machine (Instron, High Wycombe, UK). The test samples were prepared in accordance with the guidelines of ISO 527-3, while the tensile tests were carried out in accordance with the guidelines of ISO 527-1. The bacterial cellulose was cut into rectangles with dimensions of 80 mm × 25 mm. At equal distances from the narrower edges, a measuring distance of 50 mm was marked. The tensile tests were carried out at a head speed of 100 mm/min. The breaking force (N) and the absolute elongation (mm) at the moment of rupture were measured. The tensile strength value (MPa) and Young’s modulus (MPa) were calculated. The mechanical properties were conducted in at least 5 repetitions.

2.3. Water Vapor Transport and Hygroscopicity of Bacterial Cellulose

Water vapor permeability was determined by the cell method with the use of a Max 50 moisture analyzer (Radwag, Warsaw, Poland). The samples of bacterial cellulose film were cut into the shape of discs with a diameter of 54 ± 2 mm. Samples were placed on the surface of a perforated dish with distilled water located in a moisture analyzer, which maintains a temperature of 30 ± 2 °C. After 1 h of acclimatization of the samples in these conditions, the initial mass of the water vessel, m₁ (the acclimatization was applied to eliminate the bound water from the measurements of water transport, which cellulose absorbs in the first stage), was measured. Then, the sample continued to acclimatize under the same conditions for another 1 h, and the weight of the vessel with water was measured again, to determine the m₂. The value of water vapor permeability, expressed in g/m²·24 h, was calculated according to Formula (1):

\[
\text{WVP} = \frac{m_1 - m_2}{S} \times \frac{24}{t} \times 10^4 \left[ \frac{\text{g}}{\text{m}^2 \cdot 24\text{h}} \right]
\]

where: m₁—mass of water in the moisture analyzer after 1 h (g), m₂—mass of water in the moisture analyzer after 2 h (g), S—surface area of the test sample (19.625 cm²), t—determination time (1 h), and 10⁴—value resulting from the conversion of cm² into m².

The hygroscopicity was determined by the gravimetric method as the difference between the mass of the samples acclimatized for 2 h at 30 °C and RH-100% in relation to the dry mass of the sample dried at 105 °C. Both measurements were conducted with 5 repetitions.

2.4. Determination of the Contact Angle and Surface Free Energy (SFE)

The contact angle measurement was performed with a Haas Phoenix 300 goniometer (Surface Electro Optics, Suwon City, Korea) based on the sessile drop method. Using an image analysis system (Image XP, Surface Electro Optics, version 5.8, Suwon City, Korea), the angle between the tangent to the drop contour and the straight line passing through its base was determined, which was recorded with a video camera. The drop was dispensed from a height of 3 mm. A computer-controlled, precise stepper motor dispensed drops with a volume of 1 µL. Using computer software (Image XP, Surface Electro Optics, version 5.8, Suwon City, South Korea), the image of the applied drop was analyzed and the contact angle was automatically measured on the basis of the preset liquid volume. Distilled water and diiodomethane (Acros Organics, Geel, Belgium) were used to calculate the surface free energy. The measurement of wetting for a water droplet was performed after 5, 20, 40, and
60 s from the moment of depositing the water drop on the cellulose surface (measurements were made in air with 50% humidity and a temperature of 21 ± 2 °C). The measurement of surface free energy (SFE) was performed according to the Owens–Wendt method, which is one of the most common methods for calculating the SFE of polymeric materials, water and diiodomethane being used the most frequently as measuring liquids [29,30]. The surface free energy \( \gamma_{\text{tot}} \) is the sum of two components: dispersion \( \gamma_D \) and polar \( \gamma_P \). The liquid drops were dispensed onto samples with dimensions of 50 mm × 50 mm. Twenty measurements were made on samples from each variant.

2.5. Scanning Electron Microscopy (SEM)

The microstructure of bacterial cellulose was examined using a Hitachi scanning electron microscope, model TM-3000 (Hitachi Ltd., Tokyo, Japan), with a digital image record. Prior to the analysis of the bacterial cellulose structure, the material was covered with gold and the cross-section was then observed. The photos of the polymers at accelerating voltages equal to 5 kV were taken with 500-1000 magnification and the record was saved using SEM software (TM3000, Hitachi Ltd., Tokyo, Japan).

2.6. Test Standards

Statistical analysis of the test results was carried out using Statistica v. 10 software (TIBCO Software Inc., CA, USA). Data were analyzed and provided as the mean ± standard deviation. To compare and determine the significance of difference between data, a one-way analysis of variance (ANOVA) was used at a 0.05 confidence level.

3. Results and Discussion

3.1. Assessment of Mechanical Properties

Table 1 presents the results of the mechanical and physical properties of film samples obtained from native and modified cellulose. One-way ANOVA showed significant differences for tested parameters of modified and native cellulose. No statistically significant differences were only found in the case of tensile strength.

| Table 1. Statistical evaluation of mechanical and physical properties of native and modified cellulose. |
|---------------------------------------------------------------|
| **Parameter** | **Native Cellulose** | **Modified Cellulose** | **Fisher's F-Test** | **Significance Level** |
|---------|-----------------|-----------------|-----------------|-----------------|
| SS      | F               | p               | SS              | F               | p               |
| Breaking load (N) | 269.90 (56.51) | 461.38 (78.88) | 125868          | 28.6218         | 0.000132 ** |
| Elongation (mm)   | 2.413 (1.110)  | 4.234 (1.430)  | 10.5571         | 6.5903          | 0.023420 ** |
| Young’s modulus (MPa) | 70.12 (5.16)  | 89.51 (8.17)   | 952.97          | 21.959          | 0.001569 ** |
| Tensile strength (MPa) | 90.14 (11.69) | 103.70 (21.29) | 626.3           | 2.2067          | 0.163207 NS  |
| Thickness (mm) | 0.117 (0.015)  | 0.175 (0.011)  | 0.009779        | 56.229          | 0.000007 ** |
| Density (g/mm³) | 0.0040 (0.0005) | 0.0026 (0.0002) | 0.000005      | 29.4344         | 0.000154 ** |

*—means and standard deviations in parentheses, **—significant at a 0.05 confidence level, and NS—not significant.

The results of the research indicate that the modification of cellulose produced on the microbiological substrate, by grinding it to the form of paste and reintegrating it, affects its physical and mechanical properties. It was found that the polymer subjected to initial homogenization after reintegration revealed a much higher breaking load, greater elongation expressed in elongation at break, and a higher value of Young’s modulus compared to native cellulose (Table 1). The results of our research are therefore in line with those presented by Yousefi et al. [24], mentioned in the Introduction. The maximum breaking load at which the sample breaks in the case of modified cellulose was 461.38 N, which was nearly two times higher (170%) compared to the breaking load determined for native cellulose.

The higher values of the breaking load for the modified samples may be related not only to their processing, but probably to their thickness as well, which was approximately 67% higher compared to the film of unmodified cellulose. This is confirmed by the tensile
strength values, which in the case of modified cellulose films are not significantly higher compared to those of native cellulose films, and as shown by the statistical analysis, these differences are not significant. However, considering the scale of these differences, it can be presumed that the applied cellulose modification process influences changes in mechanical properties. These assumptions are confirmed by the elongation values at the maximum breaking strength for modified cellulose, which were higher compared to native cellulose (approximately 76%) and were statistically significant. It should be added that despite a large number of measurements, a relatively high standard deviation of elongation results was observed; however, for the microbiological origin material these values of the obtained variability can be considered acceptable [31].

The Young’s modulus values for native and modified cellulose also differed and were at the level of 70 and 89 MPa, respectively. That indicates that the value of this parameter for modified cellulose is also about 27% higher. Although compared to the literature data, the Young’s modulus values are relatively low, as for cellulose films, it should be noted that the mechanical properties of the polymer are influenced by many factors, such as the cultivation method or drying conditions and parameters [15]. The removal of water from bacterial cellulose changes its structure, which results in its mechanical properties [32].

According to Swingler et al. [6] the mechanical strength of cellulose changes with its moisture content. They noticed that the tensile strength values of dry cellulose are ca. 240 MPa and are two times lower than that of wet cellulose. Similar observations were made by Domskiene et al. [16].

Analyzing the effect of cellulose film thickness on the measured parameters, a very high correlation was observed between the thickness of the native bacterial cellulose samples and their elongation and breaking load, while a moderate correlation between these parameters was observed in the case of the modified cellulose (Figure 1). Moreover, the correlations between thickness and mechanical properties are characterized by the opposite direction of dependence for native and modified polymers. In the case of native cellulose, this is a positive correlation, consisting of the fact that with increasing sample thickness, their elongation and breaking load increases. This phenomenon can be explained by a greater degree of ordering of the native cellulose structure and its homogeneous structure with a layer, which is confirmed by photos of microscopic images (Figure 2a). This results in a greater density of the polymer, so as the thickness increases, the mechanical parameters also increase. The opposite effect, observed for modified cellulose, can be explained by the greater irregularity of the structure of this polymer, associated with an increase in porosity causing a decrease in the apparent density of the material. In this case, the difference in thickness between the samples is related to an increase in air spaces between the polymer layers, which does not translate into an increase, but on the contrary, a decrease in mechanical parameters with increasing film thickness (Figure 1b). It should be emphasized, however, that this correlation is average and results from a relatively high variability of the measurement results. However, it shows that the applied mechanical treatment changes the mechanical properties of bacterial cellulose.

As mentioned above, this interesting phenomenon is related to a change in the spatial arrangement of cellulose fibers, but above all their density. Although the modified polymer is thicker than the native cellulose its density is 35% lower (Table 1). In the case of modified cellulose, an increase in film thickness is clearly visible, despite the fact that the amount of the wet weight of cellulose used to prepare the modified film corresponded to the weight of the wet weight of the native bacterial cellulose film. The increase in the thickness of the cellulose film and the lower density suggest that in the integration process after homogenization, spatial reorganization processes of cellulose fibers take place, which occur in a multidirectional manner. Based on the photos from the scanning microscope, it can be observed that the structure of the native polymer is multilayered; moreover, the layers are homogeneous and coherent (Figure 2a). A completely different picture of the organization of fibers is visible in the photo of modified cellulose. This photo shows the irregularity and multidirectional arrangement of cellulose fibers (Figure 2b).
Figure 1. Correlation relationships between the mechanical properties of cellulose and the thickness of the polymer: (a) elongation and (b) breaking load.

Figure 2. Cross-section structure of bacterial cellulose (500× and 1000× magnification): (a) native cellulose and (b) modified cellulose.
The increase in the breaking load of modified cellulose, despite the lower density of the material, is an interesting phenomenon, which certainly requires additional structural tests. Nevertheless, the descriptions of modified materials are characterized by lower density and higher mechanical strength compared to the starting materials [33] or materials where the reduction in density had no effect on strength changes [34]. Perhaps the higher strength results in the change in the degree of crystallinity of the polymer or in the interactions between the molecules. According to Stanisławska et al. [15] during the drying of native and modified cellulose a decrease in the breaking strength of native cellulose was observed. The authors of the research see the reason for these differences in the phenomenon of greater reinforcement of fibers in native cellulose, which occurs during the stretching process.

3.2. Water Vapor Permeability

The course of the water vapor permeability by bacterial cellulose samples was illustrated with the example of modified cellulose (Figure 3), where uniform transport of water vapor through the tested material was observed after about 40 min from the start of the measurement. Until then, the weight of the vessel did not change, which was related to the absorption of water by the tested samples till reaching the maximum. Next, weight loss was observed, which was related to the transport of water to the outside.

![Figure 3. The course of mass measurement in the process of water vapor permeability determination for the examples of native and modified cellulose.](image)

The results of measurements of water vapor permeability through native and modified cellulose show that the modification of cellulose by grinding and reassembling the fibers affects the changes in water vapor permeability. Statistically significant differences were found between the water vapor permeability of the native and modified polymers. Additionally, it was noticed that the hygroscopicity of modified cellulose samples is much higher than that of native cellulose (Table 2). This is probably related to the change in the density and spatial structure of modified cellulose. As the research of Tome et al. [35] has shown, the water vapor permeability of bacterial cellulose increases with increasing moisture content.

3.3. Contact Angle and Surface Free Energy (SFE)

The contact angle measurements confirmed that both types of cellulose exhibit hydrophilic properties (the contact angle is below 90°).
Table 2. Water vapor permeability (WVP) and hygroscopicity (Hg) of native and modified cellulose.

| Parameter       | Native Cellulose * | Modified Cellulose * | Sum of Squares | Fisher's F-Test | Significance Level |
|-----------------|--------------------|----------------------|----------------|-----------------|-------------------|
| WVP (g/m²·24 h) | 1084.3 (36.0)      | 1174.0 (59.3)        | 21933          | 9.612           | 0.012717 **       |
| Hg (%)          | 18.38 (0.4)        | 29.89 (2.23)         | 302.124        | 1077.42         | 0.000000 **       |

*—means and standard deviations in parentheses, **—significant at a 0.05 confidence level.

It was found that there are no statistically significant differences in water wettability between native and modified cellulose. We obtained statistically significant differences in the diiodomethane contact angle of the tested cellulose samples (Table 3). Because the water has a higher surface tension (0.072 N·m⁻¹) than diiodomethane (0.0508 N·m⁻¹) [36], it is possible that the surface quality did not affect the value of the water contact angle as much as it did the value of the diiodomethane contact angle; therefore, it can be assumed that lower SFE values for modified cellulose compared to native cellulose were noticed. General, relatively high values of the water contact angle were recorded compared to the data in the literature [37,38]. Da Silva et al. [39] showed that the value of the water contact angle for bacterial cellulose was equal to 32.22° at the moment of applying a drop of water to the polymer surfaces.

Table 3. Measurement of surface wettability and free surface energy of cellulose samples.

| Parameter              | Time [s] | Native Cellulose * | Modified Cellulose * | Mean Sum of Squares | Fisher’s F-Test | Significance Level |
|------------------------|----------|--------------------|----------------------|---------------------|-----------------|-------------------|
| Contact angle (°)      |          |                    |                      |                     |                 |                   |
| for water              | 5        | 63.3 (7.9)         | 64.4 (5.9)           | 9.8                 | 0.205           | 0.653411 NS       |
|                        | 20       | 60.4 (8.3)         | 61.3 (6.4)           | 5.9                 | 0.110           | 0.742232 NS       |
|                        | 40       | 58.2 (8.5)         | 59.6 (6.8)           | 17.1                | 0.291           | 0.593234 NS       |
|                        | 60       | 57.3 (9.3)         | 58.5 (7.0)           | 12.5                | 0.188           | 0.667767 NS       |
| Contact angle (°)      |          |                    |                      |                     |                 |                   |
| for diiodomethane      | 5        | 30.2 (6.6)         | 44.9 (6.4)           | 2124.9              | 50.645          | 0.000000 **       |
| Free surface energy    |          |                    |                      |                     |                 |                   |
| (SFE) (mJ × m⁻²)       |          | γ<sub>tot</sub> = 53.391 | γ<sub>tot</sub> = 48.073 |                      |                 |                   |
|                       |          | γ<sub>D</sub> = 44.139 | γ<sub>D</sub> = 37.064 |                      |                 |                   |
|                       |          | γ<sub>P</sub> = 9.252 | γ<sub>P</sub> = 11.009 |                      |                 |                   |

*—means and standard deviations in parentheses, **—significant at a 0.05 confidence level, and ***γ<sub>tot</sub>—surface free energy as the sum of two components: dispersion (γ<sub>D</sub>) and polar (γ<sub>P</sub>).

In relation to the presented value of the diiodomethane contact angle of the surfaces of the tested materials, similar observations were obtained by Cunha and Gandini [40]. The obtained differences also concern the values of the surface free energy, including its polar and dispersion components. Therefore, it can be assumed that the method of the applied mechanical modification influences changes in the quality of the cellulose surface.

According to the literature, in the case of contact angle measurements, when a small, single drop of liquid is deposited on the surface, the geometry of the tested material surface has a greater impact on the value of the static contact angle than its chemical composition. This observation was made by Wenzel [41], who supplemented the ‘classic’ theory of surface wettability with issues concerning the influence of its quality on contact angle values [42–45]. These data emphasize the importance of the influence of the tested surface physical structure on the contact angle values. The surfaces of the materials analyzed by us should also be considered as heterogeneous, i.e., surfaces with so-called double roughness, which is created by the air and solids. According to the theory of Cassie and Baxter [46], the rougher the tested surface, the smaller the solid–liquid contact area. It results in a higher contact angle value. The results presented in this paper refer to the above-mentioned assumptions; with regard to bacterial cellulose film, it is justifiable to continue research in this area.
4. Conclusions

The results obtained in this study, dealing with the quality parameters of films made of bacterial cellulose, show that the mechanical modification of bacterial cellulose by the fragmentation and later reintegration of particles influences the strength parameters as well as selected physical and structural properties of this polymer.

The mechanical modification of bacterial cellulose contributes to an increase in the breaking load, Young’s modulus, and elongation of the cellulose film samples. Mechanical damage to cellulose fibers and their reintegration causes a change in the internal structure of the polymer, which is accompanied by a decrease in its apparent density and ability to absorb moisture, as well as its water vapor permeability. The assessment of the surface wettability and the surface free energy of the bacterial cellulose films requires the continuation of research, which will also reflect the comprehensive influence of the surface structure on these parameters.

Undoubtedly, the obtained research results show that the mechanical processing of bacterial cellulose has a significant impact on selected mechanical and physical parameters. This might be of great importance in the case of designing products based on this type of polymer, intended for specific applications.

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References

1. Abba, M.; Abdullahi, M.; Nor, M.H.M.; Chong, C.S.; Ibrahim, Z. Isolation and characterisation of locally isolated Gluconacetobacter xylinus BCZM sp. with nanocellulose producing potentials. IET Nanobiotechnol. 2018, 12, 52–56. [CrossRef]

2. Premjet, S.; Premjet, D.; Ohtani, Y. The effect of ingredients of sugar cane molasses on bacterial cellulose production by Acetobacter xylinum ATCC 10245. Sen-iGakkaishi 2007, 63, 193–199. [CrossRef]

3. Skocaj, M. Bacterial nanocellulose in papermaking. Cellulose 2019, 26, 6477–6488. [CrossRef]

4. Araujo, S.; Moreira da Silva, F.; Gouveia, I.C. The role of technology towards a new bacterial-cellulose-based material for fashion design. J. Ind. Intell. Inf. 2015, 3, 168–172. [CrossRef]

5. Wacikowski, B.; Michałowski, M. The possibility of using bacterial cellulose in particleboard technology. Ann WULS—SGGW Wood Technol. 2020, 109, 16–23. [CrossRef]

6. Swingler, S.; Gupta, A.; Gibson, H.; Kowalczuk, M.; Heaselgrave, W.; Radecka, I. Recent Advances and Applications of Bacterial Cellulose in Biomedicine. Polymers 2021, 13, 412. [CrossRef] [PubMed]

7. Frone, A.N.; Panaitescu, D.M.; Chiulan, I.; Nicolae, C.A.; Casarica, A.; Gabor, A.R.; Trusca, R.; Damian, C.M.; Purcar, V.; Alexandrescu, E.; et al. Surface treatment of bacterial cellulose in mild eco-friendly conditions. Coatings 2018, 8, 221. [CrossRef]

8. Badshah, M.; Ullah, H.; Khan, A.R.; Khan, S.; Park, J.K.; Khan, T. Surface modification and evaluation bacterial cellulose for drug delivery. Int. J. Biol. Macromol. 2018, 113, 526–533. [CrossRef]

9. Hu, W.; Chen, S.; Yang, Z.; Li, Z.; Wang, H. Functionalized bacterial cellulose derivatives and nanocomposites. Carbohydr. Polym. 2014, 101, 1043–1060. [CrossRef]

10. Gao, X.; Söüzert, E.; Zhi, Z.; Yang, G.; Silberchmidt, V.V. Mechanical modification of bacterial cellulose hydrogel under biaxial cyclic tension. Mech. Mater. 2020, 142, 103272. [CrossRef]
11. Betlej, I.; Salerno-Kochan, R.; Krajewski, K.J.; Zawadzki, J.; Boruszewski, P. The influence of culture medium components on the physical and mechanical properties of cellulose synthesized by kombucha microorganisms. BioResources 2020, 15, 3125–3135. [CrossRef]

12. Betlej, I. Studies on the diversity of substrate composition in the culture medium of Kombucha microorganisms and its influence on the quality of synthesized cellulose. Ann WULS—SGGW Wood Technol. 2019, 108, 21–25. [CrossRef]

13. Vigentini, I.; Fabrizio, V.; Dellaca, F.; Rossi, S.; Azario, I.; Mondi, C.; Benaglia, M.; Foschino, R. Set-up of bacterial cellulose production from the genus Komagataeibacter and its use in a gluten-free bakery product as a case study. Front Microbiol. 2019, 10, 1953. [CrossRef] [PubMed]

14. Chen, G.; Wu, G.; Chen, L.; Wang, W.; Hong, F.F.; Jönsson, L.J. Comparison of productivity and quality of bacterial nanocellulose synthesized using culture media based on seven sugars from biomass. Microb. Biotechnol. 2019, 12, 677–687. [CrossRef] [PubMed]

15. Stanislawksa, A.; Staroszczyk, H.; Szkodo, M. The effect of dehydration/rehydration of bacterial nanocellulose on its tensile strength and physicochemical properties. Carbohydr. Polym. 2020, 236, 116023. [CrossRef]

16. Domskiene, J.; Sedoraviciute, E.; Simonaityte, J. Kombucha bacterial cellulose for sustainable fashion. Int. J. Cloth. Sci. Technol. 2019, 31, 644–652. [CrossRef]

17. Cazón, P.; Velázquez, G.; Vázquez, M. Bacterial cellulose films: Evaluation of the water interaction. Food Packag. Shelf Life 2020, 25, 100529. [CrossRef]

18. Stanislawksa, A. Bacteria nanocellulose as a microbiological derived nanomaterial. Adv. Mater. Sci. 2016, 16, 45–57. [CrossRef]

19. Indriyati, I.; Indrati, L. Incorporation of citrus essential oils into bacterial cellulose-based edible films and assessment of their physical properties. IOP Conference Series: Earth Environ. Sci. 2017, 60, 012080. [CrossRef]

20. Yano, S.; Maeda, H.; Nakajima, M.; Hagiwara, T.; Sawaguchi, T. Preparation and mechanical properties of bacterial cellulose nanocomposites loaded with silica nanoparticles. Cellulose 2008, 15, 111–120. [CrossRef]

21. Cazón, P.; Vázquez, M.; Velázquez, G. Composite films with UV-Barrier properties of bacterial cellulose with glycerol and poly(vinyl alcohol): Puncture properties, solubility, and swelling degree. Biomacromolecules 2019, 20, 3115–3125. [CrossRef] [PubMed]

22. Sommer, A.; Staroszczyk, H.; Sinkiewicz, I.; Brużdziak, P. Preparation and characterization of films based on disintegrated bacterial cellulose and montmorillonite. J. Polym. Environ. 2020, 29, 1526–1541. [CrossRef]

23. Luddee, M.; Pivsa-Art, S.; Sirisansaneeyakul, S.; Pechyen, C. Particle size of ground bacterial cellulose affecting mechanical, thermal, and moisture barrier properties of PLA/BC biocomposites. Energy Procedia 2014, 56, 211–218. [CrossRef]

24. Yousefi, H.; Faezipour, M.; Hedjazi, S.; Mousavi, M.W.; Azusa, Y.; Heidair, A.H. Comparative study of paper and nanocomposite properties prepared from bacterial cellulose nanofibers and fibers/ground cellulose nanofibers of canola straw. Ind. Crops Prod. 2014, 43, 732–737. [CrossRef]

25. Balquinta, M.L.; André, S.C.; Cerrutti, P.; Califano, A.N.; Lorenzo, G. Effect of bacterial nanocellulose post-synthetic processing on powders and rehydrated suspensions characteristics. J Food Eng. 2020, 280, 109994. [CrossRef]

26. Gao, W.H.; Chen, K.F.; Yang, R.D.; Yang, F.; Han, W.J. Properties of bacterial cellulose and its influence on the physical properties of paper. Bioresources 2011, 6, 144–153. [CrossRef]

27. Yamada, S.; Ishihara, M.; Sugiyama, J. Structural modification of bacterial cellulose. Cellulose 2000, 7, 213–225.

28. Boruszewski, P.; Betlej, I. Płyta Wiórowa Modyfikowana Celulozą Bakteryjną. Patent Application No. P.433630, 21 April 2020.

29. Owens, D.K.; Wendt, R.C. Estimation of the surface free energy of polymers. J. Appl. Polym. Sci. 1969, 13, 1741–1747. [CrossRef]

30. Liptakova, E.; and Kudel, J. Analysis of the wood-wetting process. Holzforschung 1994, 48, 139–144. [CrossRef]

31. Stefiukien, E.; Bosacka, K.; Hryniewicz, W. Wadlacja i weryfikacja metod i testów diagnostycznych w laboratorium mikrobiologicznym. Post Microbiol. 2015, 54, 415–424.

32. Ul-Islam, M.; Khattak, W.A.; Kang, M.; Kim, S.M.; Khan, T.; Park, J.K. Effect of post-synthetic processing conditions on structural variations and applications of bacterial cellulose. Cellulose 2013, 20, 253–263. [CrossRef]

33. Yang, S.; Yao, X.; Li, J.; Wang, Z.; Zhang, C.; Wu, S.; Wang, K.; Wang, W. Preparation and properties of ready-to-use low-density foamed concretederived from industrial solid wastes. Constr. Build. Mater. 2021, 287, 122964. [CrossRef]

34. Boruszewski, P.; Borysiuk, P.; Mamiński, M.; Czechowska, J. Mat compression measurements during low-density 2 particleboard manufacturing. Bioresour. Technol. 2016, 11, 6909–6919. [CrossRef]

35. Tome, L.C.; Brandão, L.; Mendes, A.M.; Silvestre, A.J.D.; Neto, C.P.; Gandini, A.; Freire, C.S.R.; Marruco, I.M. Preparation and characterization of bacterial cellulose membranes with tailored surface and barrier properties. Cellulose 2010, 17, 1203–1211. [CrossRef]

36. Mamiński, M.; Mierzejewska, K.; Borysiuk, P.; Parzuchowski, P.; Boruszewski, P. Surface properties of octadecanol—Grafted pine veneers. Int. J. Adhes. Adhes. 2009, 29, 781–784. [CrossRef]

37. Petrie, R.A.N. Estudo in vitro da interação da linhagem de fibroblastos L929 com membranas de cellulose bacteriana para aplicações em engenharia de tecidos. Ph. D. Dissertation, Universidade Federal de Santa Catarina, Florianópolis, Brazil, 2007.

38. Lee, K.Y.; Blaker, J.J.; Bismarck, A. Surface functionalisation of bacterial cellulose as the route to produce green polyacrylate nanocomposites with improved properties. Compos. Sci. Technol. 2009, 69, 2724–2733. [CrossRef]

39. da Silva, C.M.; Bottene, M.K.; de Oliveira Barud, H.G.; da Silva Barud, H.; Ligabue, R.A.; Jahn, V.D. Wettability and morphological characterization of a polymeric bacterial cellulose/corn starch membrane. Mater. Res. 2015, 18, 10–113. [CrossRef]
40. Cunha, A.G.; Gandini, A. Turning polysaccharides into hydrophobic materials: A critical review. Part 1. Cellulose. *Cellulose* 2010, 17, 875–889. [CrossRef]

41. Wenzel, R.N. Resistance of solid surfaces to wetting by water. *Ind. Eng. Chem.* 1936, 28, 988–994. [CrossRef]

42. Kim, J.; Choi, S.O. Superhydrophobicity. In *Waterproof and Water Repellent Textiles and Clothing*; Williams, J., Ed.; Woodhead Publishing: Cambridge, UK, 2018; pp. 267–297.

43. Barnat-Hunek, D. *Swobodna Energia Powierzchniowa Jako Czynnik Kształtujący Skuteczność Hydrofobizacji w Ochronie Konstrukcji Budowlanych*, 1st ed.; Politechnika Lubelska: Lublin, Poland, 2016; pp. 23–38.

44. Celia, E.; Darmanin, T.; de Givenchy, E.T.; Amigoni, S.; Guittard, F. Recent Advances in Designing Superhydrophobic Surfaces. *J. Colloid. Interf. Sci.* 2013, 402, 1–18. [CrossRef]

45. Kowalski, M. Trwałość Właściwości Biofizycznych Wielowarstwowych Materiałów Odzieżowych w Procesach Konserwacji. Ph.D. Thesis, Cracow University of Economics, Cracow, Poland, 2021.

46. Cassie, A.B.D.; Baxter, S. Wettability of Porous Surfaces. *Trans. Faraday Soc.* 1944, 40, 546–551. [CrossRef]