Synthesis of cellulose nanocrystals (CNCs) from cotton using ultrasound-assisted acid hydrolysis

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ABSTRACT

This present work reports the synthesis of Cellulose nanocrystals (CNCs) from cotton using an ultrasound-assisted acid hydrolysis. Further, the synthesized CNCs was comprehensively characterized using Fourier Transform Infrared Spectroscopy (FTIR) to analyze surface functional groups and X-ray diffraction (XRD) in studying structural characteristics. Differential Thermal Analysis (DTA) and Thermogravimetric Analysis (TGA) have been used to study the thermal properties of CNCs. Morphology of CNCs was studied using a Transmission Electron Microscope (TEM) and Scanning Electron Microscope (SEM). The crystallite size was found to be 10–50 nm using XRD data and the average particle size to be 221 nm using PSD analysis.

1. Introduction

Cellulose is abundantly available in India and worldwide that could be obtained from cotton and wood resources. Due to excellent surface and structural properties, cellulose is being researched extensively for various applications such as food, chemicals, textile, biomaterials, electronics, electrochemical devices, and pharmaceuticals. [1–2]. Cellulose has been further classified into Cellulose microcrystals (CMCs), Cellulose nanofibers (CNFs), and Cellulose nanocrystals (CNCs) according to its morphology and structural characteristics. CNCs exhibit spindle-like morphology with a length of 50–350 nm, a width of 5–20 nm, and aspect ratios of 5–30 nm [3]. CNFs have fibrillar morphology with flexible structure and their typical length is more than 1 μm, width is 20–100 nm, and aspect ratio is 10–100. The cellulose-based products could be recycled or degraded wherein sonochemical degradation and enzymatic degradation are attractive approaches [4–5]. Various cellulose-rich materials have been used as raw materials in the process of CNCs synthesis. Among them ‘Cotton’ it is a promising resource, because its cellulose content is more than 90% and it is abundantly available in the Indian subcontinent. Moreover, waste cotton and cotton products could also be used for the synthesis of CNCs. The production of CNCs from waste cotton or raw cotton is a promising alternative for its valorization. The CNCs obtained using cotton as a raw material could have competitive physicochemical properties and cheaper cost. Their further use in applications such as; nanocomposites, cosmetics, tissue engineering, polymer filler, and food processing could significantly reduce the production cost [6–10] in this 21st century.

In the past, various approaches have been used for the preparation of CNCs. These methods determine the size, shape, morphology, and structural properties of the cellulose nanomaterials. Acid hydrolysis is a widely used method in the process of separation of CNCs from amorphous cellulose. However, the use of acid causes potential degradation of cellulose and increases the susceptibility to corrosion due to residual acid [11–12]. The ultrasound-assisted method is found to be a promising technique to extract CNCs from a whole cellulose matrix. The cavitation phenomenon in ultrasound provides energy in the extraction of CNSs through the breakdown of amorphous cellulosic bonds. The chemical effects during cavitation are derived from hotspots generated due to the collapse of bubbles. The prior studies have shown that the ultrasound degrades polysaccharides linkages. It has also shown that the cavitation effect due to ultrasound excessive access and reactivity of cellulosic bonds [13]. High-intensity ultrasonication could also isolate/separate micro- and nano-scale fibrils from cellulose resources. Owing to the various advantages of the ultrasound-assisted method, the comparison study of CNCs and its characteristics obtained from various sources and extraction methods as shown in Table 1.

In this work, the acid hydrolysis has been performed on cotton in the presence of ultrasound cavitation to isolate CNCs. Two different concentrations of acids have been used to compare the effect of acid treatment in the presence of ultrasound. Further, the CNCs was characterized to study the surface functional groups, structural conformation, thermal properties, and morphology of the obtained product. The results indicated that the use of ultrasound along with acid hydrolysis is a promising technique to extract CNCs from the cotton source.

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2. Experimental details

2.1. Materials

The cotton raw material was purchased from the Warangal local market, India. Sulfuric acid (H$_2$SO$_4$, 98%) was purchased from Zen Chemicals, India. Sodium hydro chlorate (NaOCl) and Sodium hydroxide (NaOH) were acquired from Sisco Research Laboratories, India. Deionized (DI) water was used in all experiments.

2.2. Method

2.2.1. Synthesis and characterizations of cellulose nanocrystals (CNCs)

15 g of cotton was cleaned with DI water followed by drying and bleaching at 60 °C for 4 h using by 1% of 500 mL NaOCl. The bleached cotton was again cleaned with DI water and desiccated at 60 °C for 4 h. 10 g of bleached cotton was further hydrolyzed using 100 mL of H$_2$SO$_4$ (30% or 50%) for 4 h. The reaction was terminated by adding 100 mL of water and cooling the reaction mixture with an ice bath. Further, pH was adjusted to 6–7 by the addition of NaOH in the presence of an ultrasound probe sonicator for 45 min. Later, the resultant solution was sonicated for 60 min (2 s ON time and 1 s OFF time, 20 kHz Frequency, tip diameter 10 mm, 120 W, M/s Dakshin Ultrasonic probe sonicator, India). The experimental setup is shown in Fig. 1. Later, the product was collected through filtration and subsequent washing with DI water and ethanol followed by drying at 60 °C in a vacuum oven. Fig. 2 shows the step-by-step process of Cellulose nanocrystals (CNCs) extraction from the cotton. As indicated, initially the cellulose fibers were bleached using NaOCl, then in stage two acid hydrolysis is carried out in the presence of ultrasound.

2.3. Characterization

X-ray Diffraction (XRD) patterns were obtained using Bruker D8 Advanced X-ray diffractometer. A Fourier Transform Infrared Spectroscopy (FTIR) spectrum was acquired using PerkinElmer Spectrum 100 FTIR spectrophotometer. Scanning electron microscope (SEM) and Transmission electron microscope (TEM) analysis were obtained by Philips CM 200. Particle size distribution was obtained using a Malvern nanosizer (Nano S90 version 7.02). Thermogravimetric analysis (TGA) curves were obtained using Perkin Elmer at a heating rate of 10 °C/min over the temperature range of 30 °C to 600 °C.

3. Results and discussion

Fig. 3 shows the cellulose cotton fibers are well dispersed and treated using the NaOCl. In the cotton fibers, there is a stack of fibers containing crystalline and amorphous phases. Due to the addition of NaOCl the bleaching takes place in the first part. Later with the addition of H$_2$SO$_4$, the amorphous phase will be removed from the crystalline phase. Sonication will have major impact on the diffusion of acid inside the cotton fibers. Due to sonication the acid diffusion takes place faster when compared to the conventional process. While, the separation of the crystalline region from the amorphous region takes place due to sonication effect (mechanical shearing effect).
3.1. XRD analysis

The structural information of CNCs was obtained using XRD data (Fig. 4). The observed peaks at 2θ = 14.65°, 16.74°, 22.58°, and 34.55° that were assigned to (111), (110), (200), and (004) planes of the CNCs, respectively [14]. For the comparison purpose, it showed that the XRD patterns of cotton and CNCs prepared with 30% and 50% acid concentrations. The characteristic peaks of CNCs confirm the isolation of CNCs from cotton using an ultrasound-assisted method [15]. The XRD pattern of cotton showed a broad and diffused spectrum indicating mainly amorphous material. As the acid concentration was increased from 30 to 50%, certain peaks disappeared due to the complete isolation of the amorphous part of the cotton. The synergetic effect of ultrasound and acid hydrolysis enhanced the oxidation of cellulose and the dissolution of the amorphous structure. The comparison of XRD pattern has shown clearly the transformation from amorphous to crystalline that was attributed to the integration of ultrasound with acid treatment. At a molecular level, ultrasound exerted shear, which facilitated the access of acid to glycosidic bonds of cellulose [16]. Further CNCs has been calculated the FWHM of all peaks, d-spacing, and crystallite size and reported in Table 2. The crystallite size of CNCs was calculated using the Scherrer formula [17].

\[
D = \frac{K\lambda}{\beta\cos \theta}
\]  

(1)
where, $D$ is crystallite size, $K$ is Bragg’s constant, $\beta$ is Full width at half maximum, $\theta$ is Bragg angle, and $\lambda$ is X-ray wavelength.

Several researchers calculated the crystallite size based on the dominant (200) peak that exhibited the crystallite size of 44.81 nm in this work. The d-spacing of the crystals (based on (200) peak) was 3.93 Å. Later, briefly compared the crystallinity of CNCs obtained in this work and the previous literature [18]. The crystallinity of synthesized CNCs was obtained 81.23% whereas previous reports [32] could obtain 72.22% crystallinity. This comparison confirmed that the use of ultrasound in the acid hydrolysis treatment increased the % crystallinity of CNCs. The synthesized CNCs morphology, size, shape, and crystallinity have been studied and compared with previous literature and reported in Table 3.

3.2. FTIR analysis

The FTIR spectra were used to analyze the surface functional groups of CNCs and raw cotton. Fig. 5 compares the FTIR spectra of cotton and the CNCs prepared using 30 and 50% acid concentrations. The absorption peaks at 3417 and 2902 cm$^{-1}$ were assigned to O–H stretching of hydrogen bonds and C–H stretching vibration, respectively. The peaks at 1644, 1317, 1162, and 817 cm$^{-1}$ were assigned to C-O stretching vibration, C-O of the aromatic ring, C-O-C pyranose ring skeleton, and C-O-C stretching, respectively [18]. The combined use of acid hydrolysis and ultrasound produced CNCs that was confirmed with FTIR spectra. Cotton showed peaks at 3417, 2902, 2131, 1644, 1317, 1162, and 893 cm$^{-1}$ that were its characteristic features [19]. The band at 3417 cm$^{-1}$ was attributed to the O–H stretching in cellulose. The characteristic peak at 2902 cm$^{-1}$ was assigned to C–H stretching asymmetric vibrations. Overall, the results indicated that the use of ultrasound during conventional acid hydrolysis produced CNCs that exhibited its characteristic functional groups as observed in the FTIR spectra.

3.3. Morphological investigations

Fig. 6 shows the SEM and TEM images of CNCs synthesized using different acid concentrations and ultrasound. Images revealed that at the acid concentration of 30% the CNCs had elliptical shape. Further increase in acid concentration (50%) resulted in a spherical shape. The overall particle size of CNCs was in the range of 20 to 100 nm. Sonication time during both levels of acid concentration (30 and 50%) was one hour. This process indicated that a combination of ultrasound and acid hydrolysis could isolate more CNCs from cotton. The images also indicated that acid concentration had a significant effect on the shape of CNCs. The acid concentration of 30% was insufficient to remove the complete amorphous phase from cotton leading to higher cluster formation. This resulted in the size of CNCs in the range of 500 to 800 nm. Similar morphology studies have reported in the literature, which showed the effect of acid hydrolysis [20-21].

3.4. Particle size distribution (PSD) of the Cellulose nanocrystals (CNCs)

The PSD of CNCs obtained using different acid concentrations and ultrasound treatment is shown in Fig. 7. PSD measurement was carried out using a dynamic light scattering method. The PSD analysis indicated that the majority of the particles ranged from 100 to 800 nm

| Source            | Morphology | Size (nm) | Shape            | Crystallinity | Reference     |
|-------------------|------------|-----------|------------------|---------------|---------------|
| waste cellulose   | spherical  | 50        | rod-like shape   | 81.23%        | Present study |
| native cotton fibers | spherical | 94.26     | rod-like shape   | 72.22%        | [32]          |

Table 2

Structural parameters of the Cellulose nanocrystals (CNCs) was obtained from XRD.

Table 3

Comparison morphology, size, shape, and crystallinity with reference work.

Fig. 4. XRD pattern of (a). Raw cotton material, (b). Cellulose nanocrystals (CNCs) with 30% of acid concentration, and (c). Cellulose nanocrystals (CNCs) with 50% of acid concentration.

Fig. 5. FT-IR spectra of (a). Raw cotton material, (b). Cellulose nanocrystals (CNCs) with 30% of acid concentration, and (c). Cellulose nanocrystals (CNCs) with 50% of acid concentration.
size. The smaller particles were in the range of 20 to 100 nm size. The comparison of PSD histograms indicated that acid concentration of 50% was effective when compared to 30%. The 50% acid concentration could narrow the particle size distribution than 30%, involving the same sonication time. The PSD analysis showed CNCs had an average size of 221 nm. Meyabadi et al. [22] obtained the particle size of CNCs in the range of 100 to 300 nm by varying the acid concentration and sonication time.

3.5. Thermal properties

Thermogravimetric analysis (TGA) and Differential thermal analysis (DTA) of the CNCs are shown in Fig. 8. TGA was carried out to understand the thermal stability/weight loss at increasing from 30 to 600 °C at the rate of 10 °C/ min. Initial weight loss was 20% below 150 °C due to the loss of volatile components present in the CNCs and moisture. Major weight loss of 30% occurred when the sample is heated from 300 to 400 °C. Both of the losses were due to the presence of low molecular weight components and amorphous components present in the CNCs. All the CNCs samples showed relatively similar thermal characteristics following previously reported literature [23–24]. These results showed that the thermal stability of CNCs extracted using ultrasound-assisted acid hydrolysis was better than the raw materials, based on the comparison with literature. Vanderflee et al. [25] reported

![Fig. 6. SEM images of as-synthesized Cellulose nanocrystals (CNCs) at (a) 50 μm, (b) 1 μm, and (c) 2 μm scale. TEM images at (d) 20 nm, (e) 10 nm, and (f) 5 nm scale (at 50% acid concentration).](image1)

![Fig. 7. Particle size distribution of Cellulose nanocrystals (CNCs) prepared with (a) 30% acid concentration and (b) 50% acid concentration.](image2)
that the CNCs with higher charge density degrades at a lower temperature. Further charge density will have a large impact on the thermal stability of the CNCs.

4. Conclusion

Cellulose nanocrystals (CNCs) were successfully synthesized using combined acid hydrolysis and ultrasound treatment. The results showed that the ultrasound-assisted acid hydrolysis treatment was an efficient method for the preparation of CNCs from the cotton. The functional groups, structural properties, and morphology of the CNCs were comprehensively studied. Crystallite size of the CNCs was calculated from XRD and it was 10–50 nm. The crystallinity of CNCs has been increased due to the combined effect of ultrasound and acid hydrolysis treatment. The TEM images indicated that concentration of the acid had a significant effect on the shape of CNCs. The average particles size of CNCs found around 221 nm. The thermal stability of nano CNCs has showed improved results than the cotton raw materials using an ultrasound-assisted acid hydrolysis. The synthesized CNCs had low particle size, high crystallinity, and stable thermal properties. The synthesized CNCs can potentially be used for various applications such as electronics, sensors and nano composites etc.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ultsonch.2020.105353.

Fig. 8. Thermal analysis of Cellulose nanocrystals (CNCs), TGA and DTG curves.

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