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Short communication

Facile synthesis of spherical cellulose nanoparticles

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Abstract

A practical procedure for synthesizing cellulose nanospheres with sizes ranging from 60 to over 570 nm was developed. This methodology provides a near linear relationship between cellulose nanoparticle size and treatment time. The hydrolyzed nanocelluloses are predominantly cellulose II polymorphic crystalline structure and relatively uniform in particle size.

Keywords: Acid sonication; Cellulose II; Cellulose electron microscopy; Cellulose AFM; Crystallinity index; Nanosphere

1. Introduction

Cellulose is the most ubiquitous and abundant biopolymer in nature and its biosynthesis, chemistry, and ultra structure remains an active field of study (Klemm, Schmauder, & Heinze, 2002; Klem, Heublein, Fink, & Bohn, 2005). Over the past decades, interest in sustainability and green chemistry has lead to a renewed interest in novel cellulosic materials (Ragauskas et al., 2006) and composites (Gradwell et al., 2004) derived from a variety of cellulosics including cellulose whiskers (Samir, Alloin, & Dufresne, 2005). These latter structures typically have fiber lengths ranging 100 to several hundred nanometer and diameters of 3–20 nm and are usually prepared by acid hydrolysis of cellulosic fibers. The exact size, dimensions, and crystallinity of these whiskers is dependent upon the acid hydrolysis conditions and source of cellulose employed. Typically, the use of tunicate and bacterial cellulose yield larger nanocrystalline cellulose whiskers, while cellulose from wood yields smaller structures (Beck-Candanedo, Roman, & Gray, 2005; Bondeson, Mathew, & Oksman, 2006). Recently, Li, Ding, and Li (2001) reported the potential of preparing nanospherical cellulose structures with diameters of several hundred nanometers from short-staple cotton by pre-swelling the fibers prior to acid hydrolysis. This note provides a high-yielding, size-predictable methodology for the synthesis of cellulose nanoparticles.

2. Experimental

2.1. Pretreatment

Cellulose fibers (30.00 g, Buckeye cellulose, 100% cellulose) were processed through a Wiley mill (Arthur H. Thomas Co.) equipped with a 20 mesh screen. The obtained cellulose was transferred into 5.00 M NaOH solution (250.00 ml) warmed to 80 °C for 3 h. The slurry was then filtered and thoroughly washed with DI water until the wash water was neutral pH. The resulting cellulosic fibers were air-dried, and then added to DMSO (250 ml) in a 80 °C water bath for 3 h. The fibers were then filtered and washed with DI water (3 × 250 ml).

2.2. Synthesis and purification

The pretreated fibers were transferred into an acidic aqueous solution consisting of 1000.00 ml mixed acid

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(made of 600.00 ml DI water, 100.00 ml 12.1 N HCl and 300.00 ml 36.0 N H_2SO_4 H_2SO_4) and this suspension was sonicated in a VWR 150 HT ultrasonicator with an internal heater at 80 °C for eight hours, during which the mixture was stirred vigorously with a mechanical stirrer.

After the hydrolysis process, the fiber slurry turned into milky colloid suspension. It contained different sizes of nanocrystalline cellulose particles. The mixture was then transferred into centrifuge bottles and centrifuged. The fractions were continuously washed by addition of DI water and centrifuged at 2000 RCF. After washing, the products were neutralized with 2.00 N NaOH to pH 7.00. The neutralized products were further washed another 3 times (3×150 ml). Afterward, the fractions were dialyzed (Spectra/Por dialysis membrane MWCO: 1000) against water. The thoroughly washed products were freeze dried and stored at 5 °C for further testing. Ultrasonication of the nanoscaled cellulose in dilute H_2SO_4–HCl solution (Ultrasonication 2 and 3) yielded smaller sized nanocellulose following a similar preparation procedure.

2.3. Characterization

Electrophoretic mobilities of the particles in pure water were measured with a Malvern 3000 Zetasizer. A nanocrystalline cellulose suspension (0.03%) was prepared, and analyzed with a Malvern 3000 Zetasizer, with the low size limit set to 5 nm, and the high level set to 1500 nm. For smaller sized fractions, the low limit was set at 2 nm, while the high level was 500 nm. Scanning electron microscopy (SEM) was performed on using a LEO 1530 thermally-assisted field emission (TFE) microscope at 3 kV; the samples were freeze dried and coated. High-resolution transmission electron microscopy (TEM) was carried out with a JEOL 100 CX II microscope at 100 kV; the sample was free-dried before TEM. The ^{13}C CP/MAS NMR spectra were recorded at room temperature on a Bruker Advance/DMX-400 operating at 100.06 MHz using a MAS WB CP BB VTN-BL 4 mm probe and ZrO_2 rotors. The MAS spin rate was 5 kHz. Acquisition was performed with a CP pulse sequence using 4.5 μs pulse, 2.0 ms contact pulse and 3.0 s delay between repetitions. Five thousand scans were accumulated for each sample. X-ray diffraction (XRD) of the cellulose samples were recorded on a Phillips PW 1800 diffractometer with Cu Kα. Atomic force microscopy was done using a Digital Instruments Nanoscope IIIa scanning probe microscope with Dimension 3100 controller. Images were collected using a tapping mode etched silicon tip.

3. Results and discussion

Synthesis of cellulose nanosphere structures involved an initial swelling of cellulose cotton with 5.00 N NaOH solutions, at 80 °C for 3 h followed by an analogous treatment with DMSO. The pretreated cellulose was then acid hydrolyzed with a mixed HCl–H_2SO_4 solution at 80 °C in a sonicator for 6 h. The resulting hydrolyzed products were purified by ultra-centrifugation, followed by dialysis with Spectra/Por membranes. Particle size analysis of the hydrolyzed cellulose suspension with a Malvern 3000 Zetasizer indicated that the product mixture consisted of two different particle sizes averaging approximately 500 nm and 70–200 nm.

Additional studies demonstrated that product yield, particle dimension, and distribution could be improved by mechanically refining the starting cellulose fibers to pass through a 20 mesh Wiley mill before pretreatment. Nonetheless, this approach failed to eliminate the bimodal distribution of cellulose nanoparticles. A detailed investigation of the hydrolysis process indicated that the bimodal particle size distribution occurred during ultrasonication (Table 1), possibly due to the heterogeneous distribution of the suspension during the treatment. Mechanical stirring during ultrasonic treatment addressed this issue as summarized in Table 2. Key to a uniform, high-yielding process was the initial mechanical refining of cellulose fibers followed by ultrasonic acid hydrolysis with mechanical stirring.

The AFM image shown in Fig. 1 is that of cellulose nanoparticles obtained from acid hydrolysis of cellulosic fibers with an average starting length of 0.237 mm. The particle shapes of these larger cellulosic particles were irregular but overall spherical in shape.

In order to synthesize cellulose nanoparticles of smaller sizes, particles with size distribution of 470 ± 100 nm were further sonicated at pH 2.5 in a HCl–H_2SO_4 solution (3:1) at 80 °C, and changes in particle length were monitored as a function of treatment time as summarized in Fig. 2 (this is called ultrasonication 2). Acidic ultrasonic treatments

Table 1
Relationship between initial cellulose size and hydrolyzed particle after ultrasonic, acid catalyzed hydrolysis[a] of cellulose particles at 80 °C for 8 h without stirring

Initial cellulose length (mm)	Yield (%)	Average cellulose particle diameter (nm) with yield (%)
0.465	62	505 ± 100 (29%) and 190 ± 100 (33%)
0.452	65	495 ± 100 (29%) and 185 ± 100 (36%)
0.347	71	485 ± 100 (31%) and 175 ± 100 (40%)
0.237	74	460 ± 100 (31%) and 170 ± 100 (43%)

[a] 12.1 N HCl – 36.0 N H_2SO_4, 2.5% cellulose by weight solution.

Table 2
Relationship between initial cellulose size and hydrolyzed particle after ultrasonic, acid catalyzed hydrolysis[a] of cellulose particles at 80 °C for 8 h with stirring (ultrasonication process 1)

Initial cellulose length (mm)	Yield (%)	Average cellulose particle diameter (nm)
0.465	66	560 ± 100
0.435	69	510 ± 100
0.347	72	490 ± 100
0.237	76	470 ± 100

[a] 12.1 N HCl – 36.0 N H_2SO_4, 2.5% cellulose by weight solution.

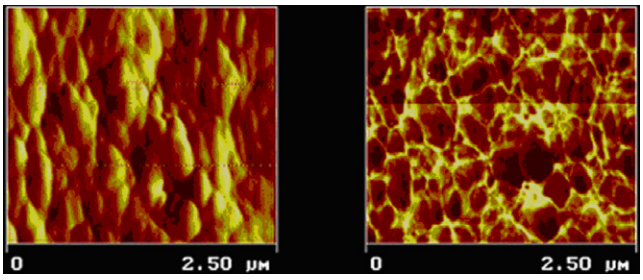

Fig. 1. Both amplitude and phase AFM images of cellulose nanoparticles with an average size of 470 ± 100 nm (2.5 micron scan).

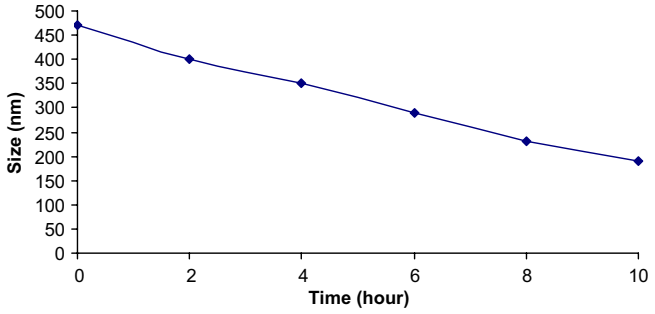

Fig. 2. Change in cellulose particle size (standard deviation of 40 nm) as a function of treatment time upon exposure to a diluted H_2SO_4–HCl at 80 °C, pH 2.5, the concentration of the dispersion is 5 mg/ml (ultrasonication 2).

extending beyond 10 h began to decrease in hydrolysis efficiency and the curve quickly reached plateau.

The modified procedure employed to produce the results in Fig. 2 provided a monodisperse sample size with a typical size dispersion of ±50 nm. To produce cellulosic particles of less than 100 nm in size, the acid hydrolyzed cellulosic particles were treated to an additional acidic sonication treatment. For example, nanocrystalline cellulose particles with a particle diameter of 190 nm were ultrasonicated in an HCl–H_2SO_4 solution (3:1) at pH 2.5, and changes in particle size were monitored as a function of treatment time, as summarized in Fig. 3. As before, the acidic sonication treatment yielded monodisperse cellulose

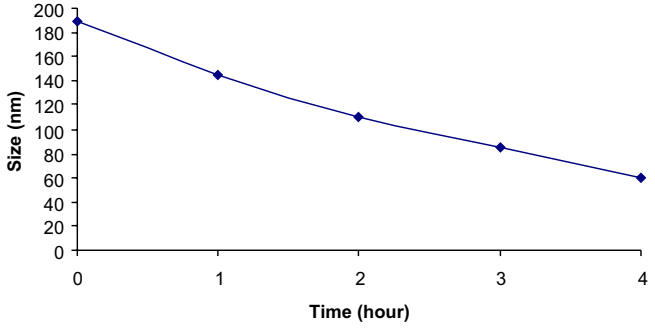

Fig. 3. Change in cellulose particle size (standard deviation of 15 nm) as a function of treatment time upon exposure to diluted H_2SO_4–HCl solution at 80 °C, pH 2.5, the concentration of the dispersion is 5 mg/ml (ultrasonication 3).

nanoparticles with a typical size dispersion of ±20 nm in yields above 70%. (This is called ultrasonication 3). Hence, this facile multi-stage treatment provides a viable and simple means of producing cellulose nanoparticles with diameters of 570–60 nm.

The samples reported in Figs. 2 and 3 were initially characterized with a Malvern 3000 Zetasizer, and then by AFM, SEM, TEM, and ^{13}C CPMAS NMR. High resolution SEM images of the nanocrystalline cellulose acquired after 3 h acidic of sonication of the 190 nm cellulose particles are shown in Fig. 4. These results indicate that the nanocrystalline cellulose particles were spherical in shape with an average diameter of 85 nm. TEM images (Fig. 5) of aggregated cellulose nanoparticles provides supporting information indicating that the particles were generally spherical in shape with average aspect ratios of 0.91–1.10.

The crystal form and degree of crystallinity of the original and treated cellulose was determined by ^{13}C CP/MAS NMR (Dinand, Vignon, Chanzy, & Heux, 2002; Maunu, Liitia, Kauliomaki, Hortling, & Sundquist, 2000). The most informative region in the NMR spectra of cellulose I is a signal cluster with a chemical shift between δ 80 and 92 ppm. The starting pulp was clearly cellulose I with

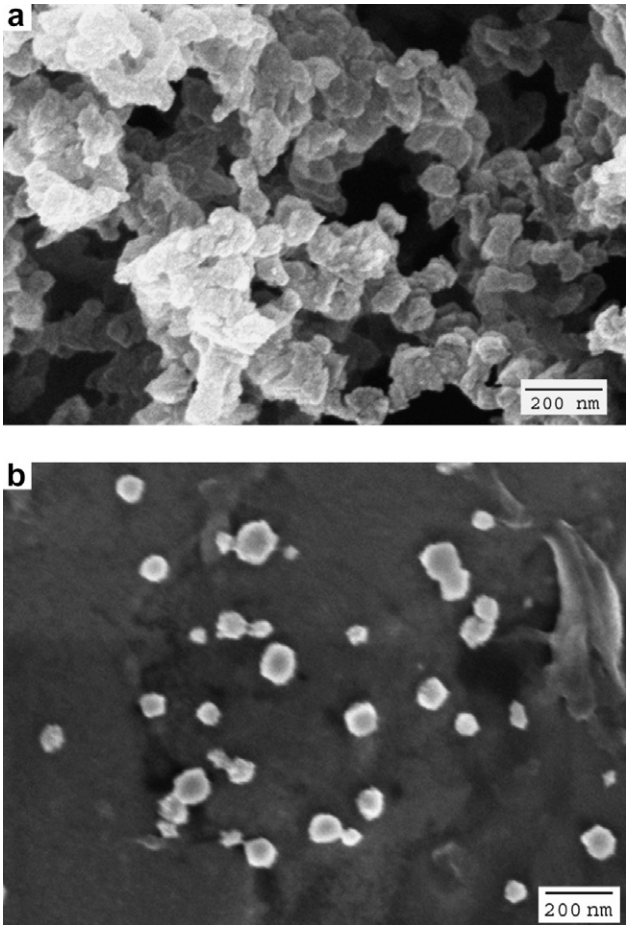

Fig. 4. SEM image (a) of aggregated cellulose nanoparticles with an average diameter of 80 nm, 10 kV, and SEM image (b) of a well distributed sample of the same sample.

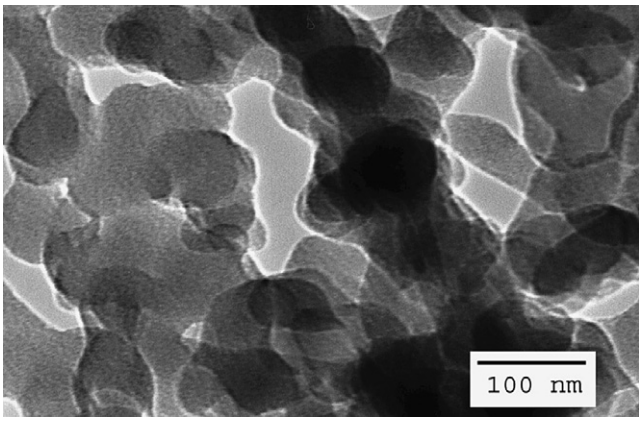

Fig. 5. TEM images of aggregated NCC with an average diameter of 80 nm.

a crystallinity index (C.I.) of 0.61. After the initial alkaline and DMSO preswelling treatment the cellulose fibers were converted from cellulose I to cellulose II and exhibited a reduced C.I. of 0.58. Subsequent acidic sonication yielded cellulose particles with increased crystallinity as summarized in Table 3. The changes in the degree of crystallinity during acid hydrolysis were due to degradation of amorphous cellulose.

XRD diffraction (Mansikkamaeki, Lahtinen, & Rissanen, 2005) analysis (data not shown) of the freeze dried 0.237 mm starting cellulose, NaOH–DMSO pre-treated fibers and cellulose nanoparticles with an average diameter of 80 nm indicated that the initial cellulose sample was cellulose I, while the pretreated cellulose and nano cellulose, with an average diameter of 80 nm, was cellulose II. The results were in agreement with the crystal index data obtained from CP/MAS NMR. Based on the experimental data a possible process for the production of nanocellulose can be hypothesized. Cellulose II was obtained after the alkaline pretreatment and the initial acid hydrolysis yielded a micrometer scale cellulose in which the amorphous regions are preferentially hydrolyzed. The micrometer scale cellulose was then converted into nanocrystalline cellulose upon additional acidic sonication. The obtained cellulose nanocrystals were predominately spherical in shape. This structural pattern of cellulose nanospheres bears resemblance to recent studies by Koga, Takenaka, Aizawa, Nakamura, and Hashimoto (2005). They demonstrated for carbon black fillers the primary aggregate was spherical in shape with a diameter of 27 nm and this unit would form higher aggregates that were linear in shape.

Table 3
Crystallinity index of hydrolyzed cellulosic particles as determined by ^{13}C CP/MAS NMR[a]

Cellulose particle dimension (nm)	Crystallinity index
~347000	0.58
~505	0.67
~460	0.71
~180	0.74
~80	0.82

[a] Particle dimension refers to the diameter of the nano cellulose with the exception of the initial value that refers to the length of the fiber.

The results in this paper provide a facile and direct means of producing spherical cellulose nanoparticles of controlled size from 60–570 nm in diameter. The methodology developed in this study provides a near linear relationship between cellulose nanoparticle size and treatment time. The hydrolyzed nanocellulose is predominantly cellulose II and relatively uniform in particle size. This methodology provides a new supramolecular cellulose nanostructure that complements the advances in the synthesis of cellulose whiskers and will undoubtedly lead to new applications for composites, drug delivery and as a template for the synthesis of spherical structures.

4. Conclusions

The present work demonstrates that nanoscale spherical cellulose particles could be synthesized from cellulosic fibers through a refined procedure, mechanical stirring was necessary to obtain a mono-mode distribution in the first place, weak acid sonication provided a near linear relationship between cellulose nanoparticle size and treatment time. These cellulose spherical structures were studied by various means including Ems, and AFM. The obtained cellulose particles were of cellulose II polymorph, there was also a tendency for the cellulose crystallinity index to increase as the particle sizes became smaller.

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