Supplementary Information

Subcritical Water Digestion of Woody Biomass: Extraction of Cellulose Nanomaterials under Acid-Lean Condition

Ruby Osei-Bonsu,[a,b] Mahfuzul Hoque,[a,b] Philip S. McMichael,[a,b] and E. Johan Foster*[a,b]

a. Department of Chemical and Biological Engineering, University of British Columbia, 2385 East Mall, Pulp and Paper Centre, V6T 1Z4.
b. Bioproducts Institute, 2385 East Mall Vancouver, BC Canada V6T 1Z4.

Experimental

Yield analysis. The yield of CNCs produced via acid-catalyzed subcritical water hydrolysis was calculated using the equation below:

$$Y = \frac{(m_2 - m_1)V_1}{m_3 V_2} \times 100$$

where m_1, m_2, and m_3 correspond to the mass of the empty weighing dish, weighing dish and the dried material, and the freeze-dried material, respectively. V_1 is the total volume of the CNM suspension and V_2 is the volume of CNM suspension to be dried.

Polarized Optical Microscopy (POM). Optical microscopy was used to investigate the digestion of pulp fibers after subcritical water treatment. Subcritical water hydrolyzed pulp suspension was vortexed for about 5 minutes after 4–5 washes at high speed. A few (2–3) drops of the suspensions were dropped by using a Pasteur pipette onto a glass slide and images with a polarised optical microscope (Nikon Eclipse LV100N POL), without polarization.

Fourier Transform Infrared Spectroscopy. The chemical composition of subcritical hydrolyzed materials and bleach wood pulp was investigated using a Fourier transform infrared spectrometer from Perkin Elmer, USA (model: Frontier™) with an attenuated total reflectance (ATR) probe. Freeze-dried samples were placed on the surface of the diamond crystal and secured tightly with the attached anvil, ensuring maximum contact between the sample and the crystal. The spectra readings were taken between 4000–650 cm^{-1} at 4 cm^{-1}. A total of 16 scans were performed for each sample.

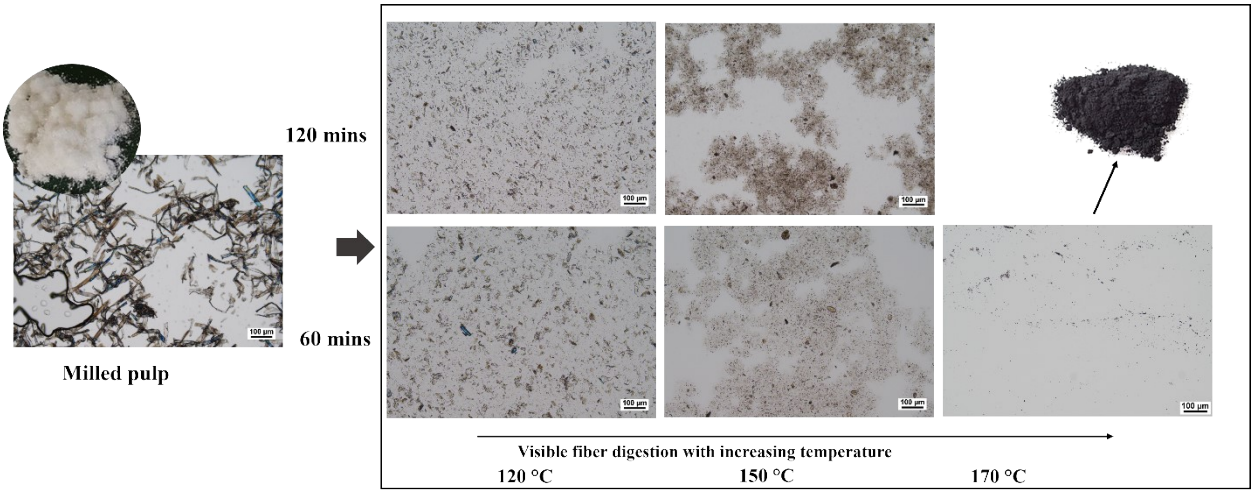
Conductometric Titration for surface charge measurement. Conductometric titration was used to estimate the surface charge content of subcritical water-hydrolyzed cellulose nanocrystals, affording to investigate the esterification ability low concentrated phosphoric acid employed in the reaction. The protocol used was adapted from CAN/CFA-Z5100-17 National Standard of Canada, 2017, with very slight modifications. For all materials investigated in this work, 0.15g of freeze-dried hydrolyzed material was initially redispersed (*via* ultrasonication) in 40 mL of ultra-pure water, diluted by adding 156.5 mL of ultra-pure followed by 2 mL of 0.1M NaCl solution and 1.5 mL of 0.1 M HCL solution, to make a total of 200 mL solution. After that, the solution was stirred continuously for about 5 minutes before titration.

The experiment was performed with an auto titrator from Metrohm AG (model: *tiamo*™ with 856 conductivity module) against 0.01M NaOH solution with 0.05 mL increments. The surface charge content was calculated according to the following equation:

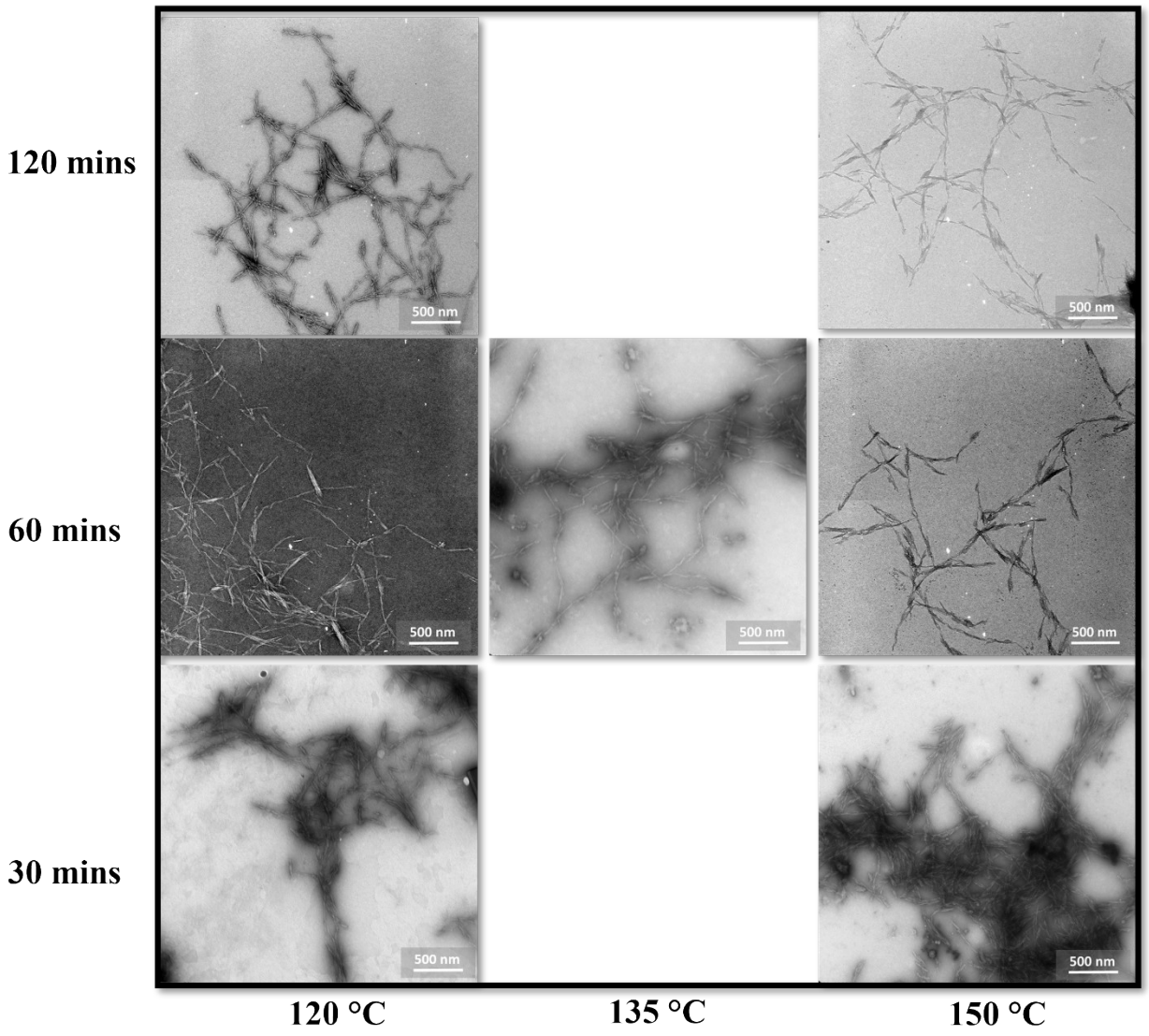
$$\frac{mmol\ HPO_4^-}{kg\ cellulose} = \frac{C_{NaOH} \times V_{NaOH}}{W_{CNC}} \times 10^6$$

Where W_{CNC} (g) is the weight of SC-CNCs titrated and V_{NaOH} corresponds to the volume difference of NaOH (L) needed to reach the first and second equivalence points.

Characterization



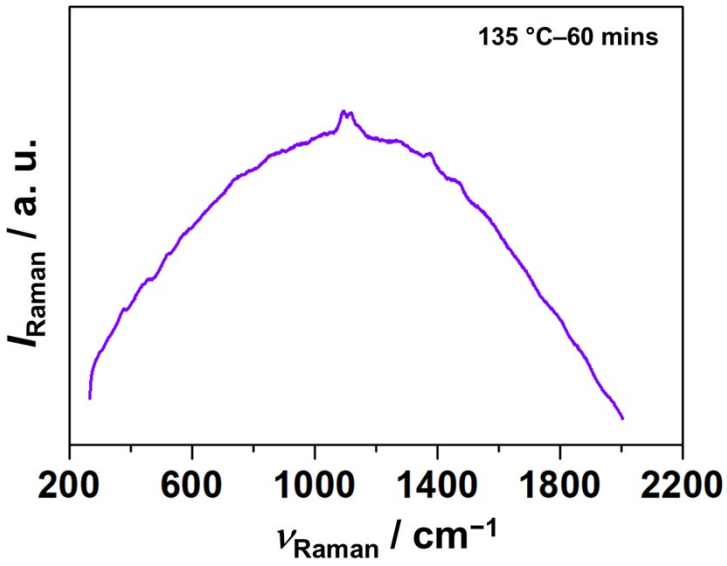
Supplementary Figure 1. Polarized optical microscopy images showing a reduction in fiber lengths and sizes after subcritical water treatment with increasing temperature from 120–170 °C for 60 and 120 minutes.



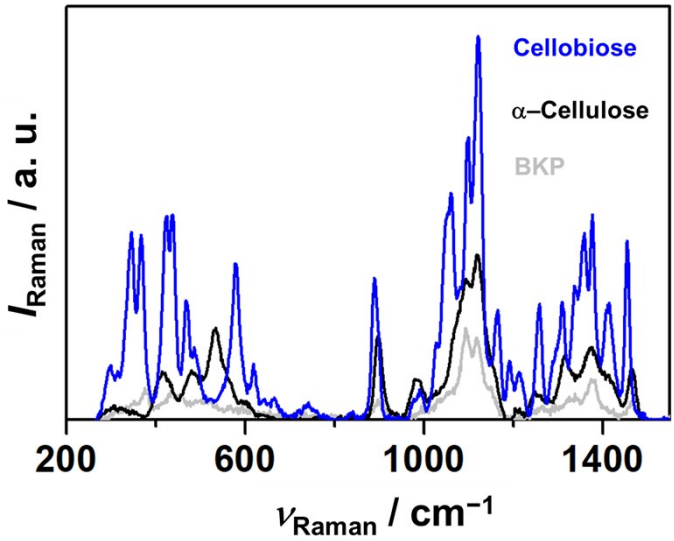
Supplementary Figure 2. TEM images of subcritical water hydrolyzed cellulose nanocrystals isolated at different subcritical temperatures and times.

Supplementary Table 1: Chemicals and utility cost analysis [1], [2]to produce cellulose nanocrystals. A comparison of the current study to previous studies and conventional methods.

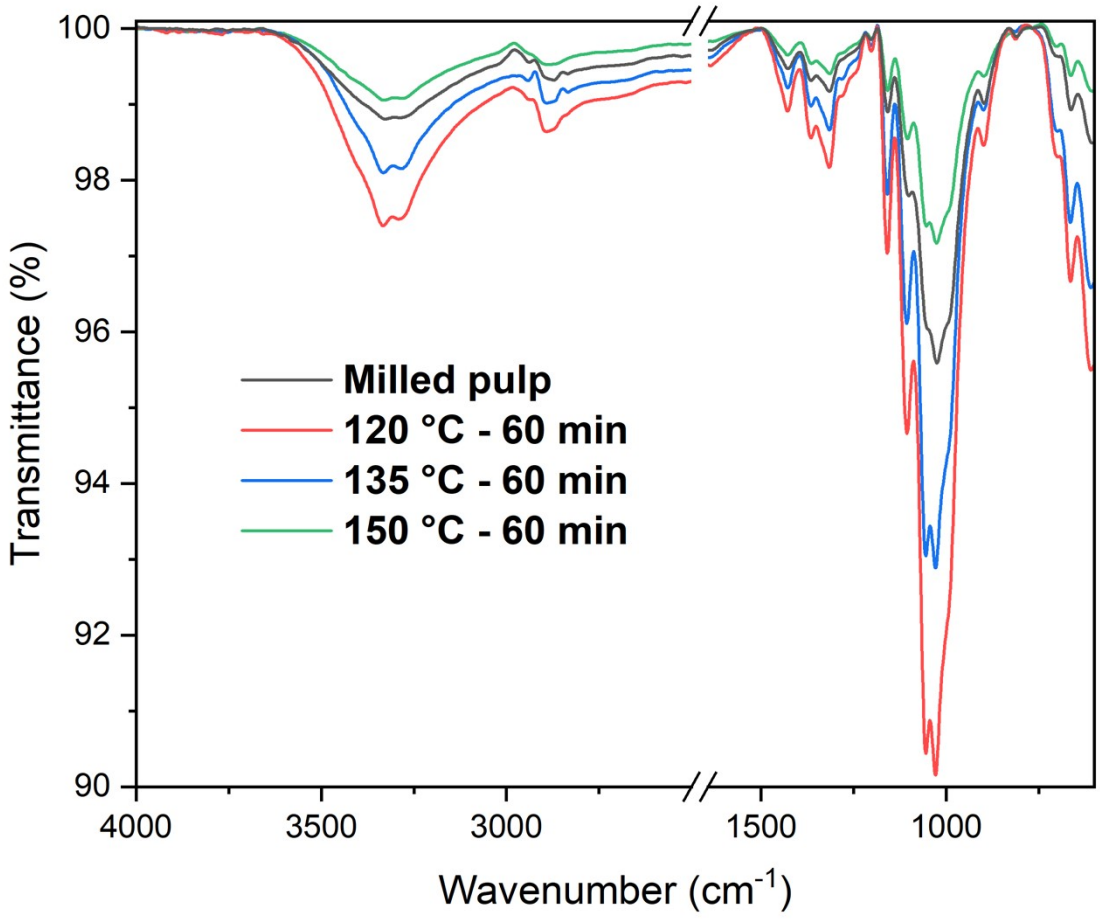
Input materials required	**Unit cost**	**This study** **Cost ($/kg CNC)**	**Previous study** [2]	**Conventional acid hydrolysis** **Cost ($/kg CNC)**
Process water ($/kg)	0.0003	**0.60**	0.30	1.308
Acid required ($/kg)	0.070	**0.70**	**-**	44.80
Cooling water ($/kg)	0.000001	0.0506	0.0506	-
Steam ($/kg)	0.002	0.3857	0.3857	0.0390
Total		1.7363	0.7363	46.147



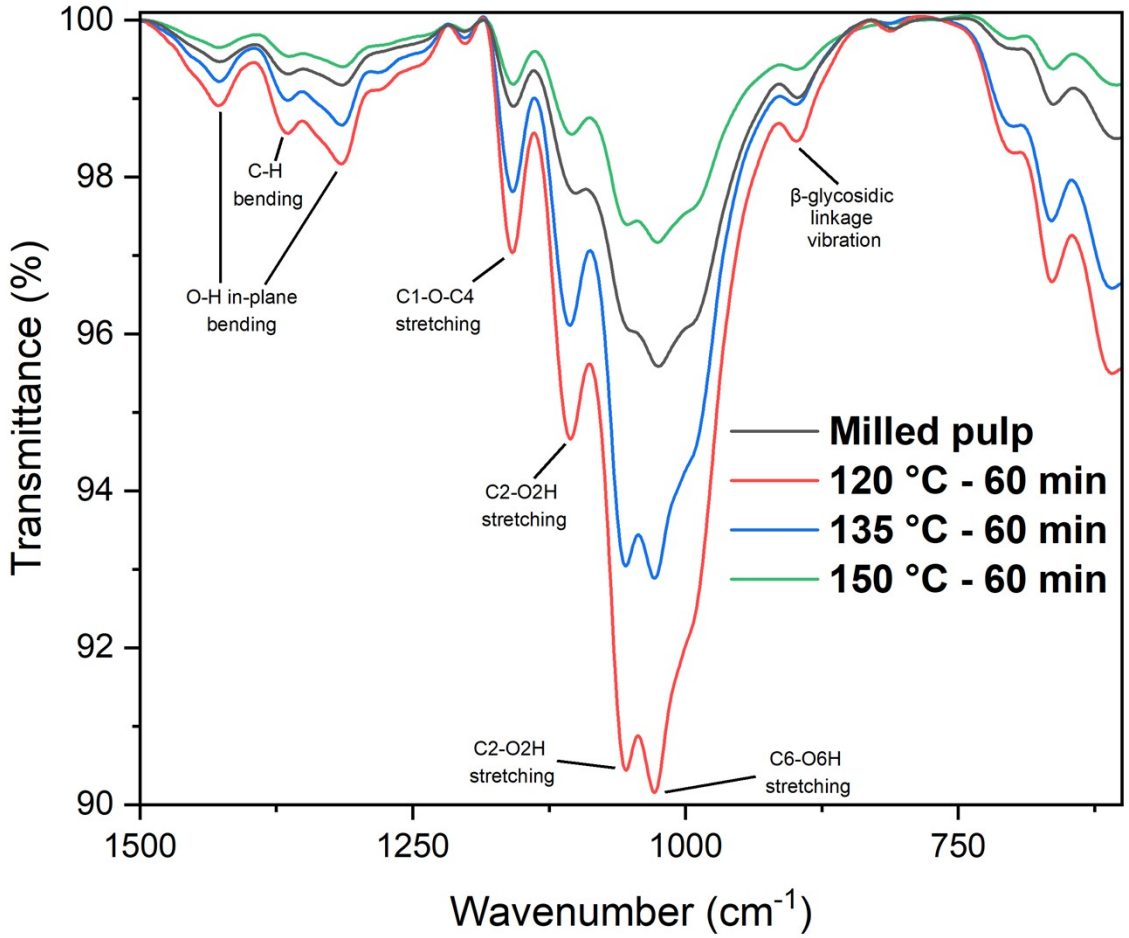
Supplementary Figure 3: Raman spectral region (1050–1175 cm^{-1}) covers the observable peaks corresponding to SCW-derived CNCs owing to large background fluorescence.



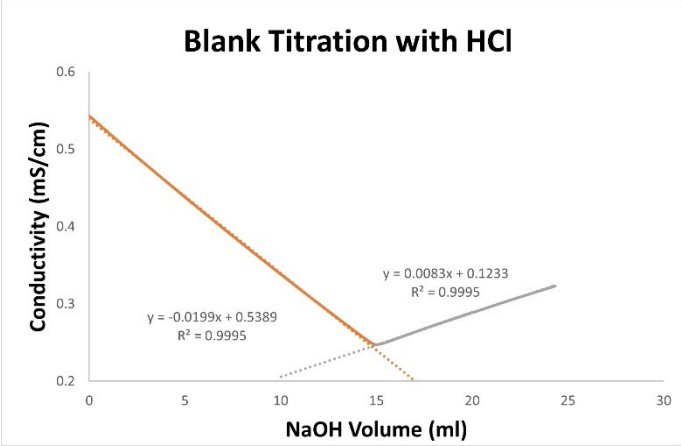
Supplementary Figure 4: The Raman spectral region (1050–1175 cm^{-1}) covers the observable peaks corresponding to SCW-derived CNCs owing to large background fluorescence.



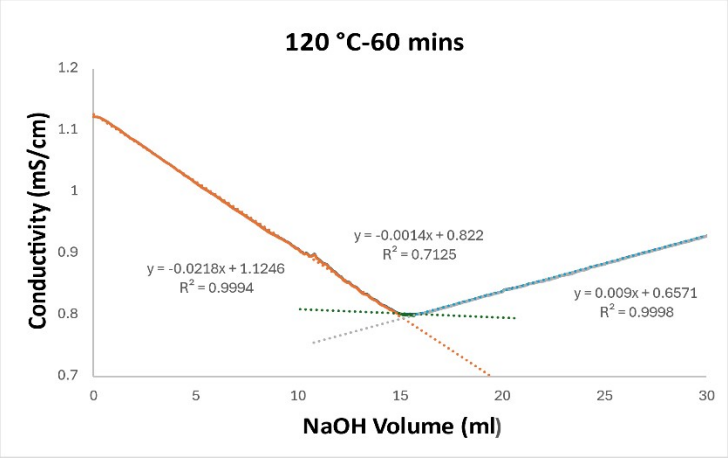
Supplementary Figure 5: ATR FT-IR spectroscopy of selected samples, showing characteristic –OH stretching vibration at ~3300–3250 cm^{-1}.



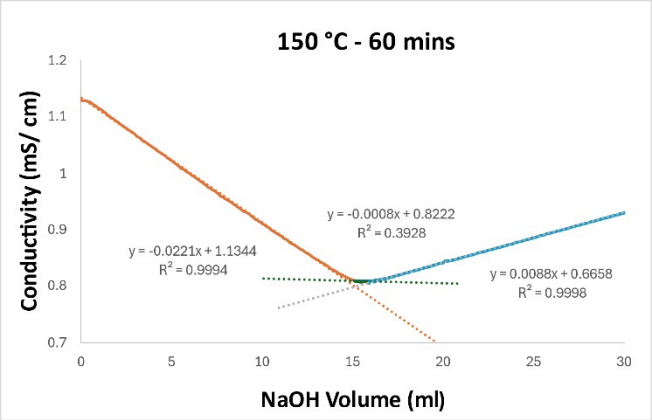
Supplementary Figure 6: ATR-FTIR spectroscopy of selected samples, showing key vibrational peaks. Samples treated at 135 ℃ and 150 ℃ showed increased peak intensity wavenumbers corresponding to various –OH stretches.



Supplementary Figure 7: Conductometric titration curve of neat medium (without CNCs) with HCl and NaOH.



Supplementary Figure 8: Conductometric titration curve of subcritical water hydrolyzed CNCs treated at 120 ℃ and 60 mins with HCl and NaOH



Supplementary Figure 9: Conductometric titration curve of subcritical water hydrolyzed CNCs treated at 150 ℃ and 60 mins with HCl and NaOH

REFERNCES

[1] N. Qureshi, B. C. Saha, M. A. Cotta, and V. Singh, "An economic evaluation of biological conversion of wheat straw to butanol: A biofuel," *Energy Convers. Manag.*, vol. 65, pp. 456–462, Jan. 2013, doi: 10.1016/J.ENCONMAN.2012.09.015.

[2] L. P. Novo, J. Bras, A. García, N. Belgacem, and A. A. S. Curvelo, "Subcritical Water: A Method for Green Production of Cellulose Nanocrystals," *ACS Sustain. Chem. Eng.*, vol. 3, no. 11, pp. 2839–2846, 2015, doi: 10.1021/acssuschemeng.5b00762.