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Supporting Information

Easily deconstructed, high aspect ratio cellulose nanofibres from *Triodia pungens*; an abundant grass of Australia's arid zone

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Measurement of length of spinifex fibre samples

Measuring the length of spinifex long and "rope-like" NFC was processed using two different methods of; cryo-TEM, a 3D tomography and measurement from TEM images by AutoCAD software.

1. Cryo-TEM

1.1. Plunge freezing protocol

4 µl of NFC dispersion in water was transferred onto TEM holey carbon grids (C-flat and lacey carbon), in an FEI Vitrobot Mark 3 (FEI Company, Eindhoven, the Netherlands), while the chamber was set to 100% humidity at room temperature (~22 ºC). Optimal blot time was 3-5 s, and then sample was plunged into liquid ethane.

1.2. Electron microscopy

Frozen/vitrified samples were viewed on a Tecnai F30 TEM (FEI Company) operating at 300kV, and imaged 23,000x magnification with a Direct Electron LC1100 4k x 4k camera (Direct Electron, San Diego, USA), using low-dose mode of SerialEM image acquisition software (http://bio3d.colorado.edu/SerialEM/). The reason that samples were subject to low dose conditions is the extreme sensitivity of the unstained cellulose nanofibres to beam damage. This consisted of using spot size 5, making focus and exposure adjustments outside of the image capture area, creating a map of grid locations at very low magnification where area selection was based on the quality of vitreous ice rather than sample morphology and performing subsequent high magnification imaging via an automated batch imaging function in SerialEM which the total electron dose was limited to 130 electrons/Å² or less. The tilt range was +/-60º with an increment of ~1.5º to 2.5º.

1.3. Image processing and analysis

125 2D images were captured in this instance and the raw image data was then processed using IMOD processing and modeling software (http://bio3d.colorado.edu/imod). This program allows contours to be manually drawn following the non-linear path of each cellulose nanofibre in xy space and contains tools for the subsequent calculation of contour length.

2. AutoCAD

Same as IMOD software, the small contours were manually drawn along the individual nanofibril or the bundles of nanofibrils in several adjacent high-resolution TEM images by AutoCAD, then the software was employed to calculate the sum of contour lengths automatically. It is easier to measure the length of a bundle of nanofibrils in the lower magnification TEM images, as the ends of fibrils can be readily located by the microscope operator.

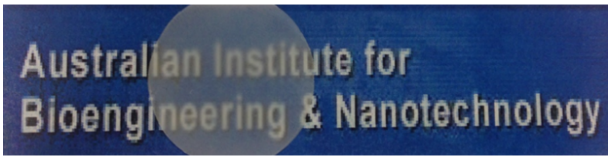

Figure S1. The optical transparency of spinifex nanopaper produced from homogenized NFC after vacuum filtration and drying with a hot press at 103 °C and 1000 psi for one hour

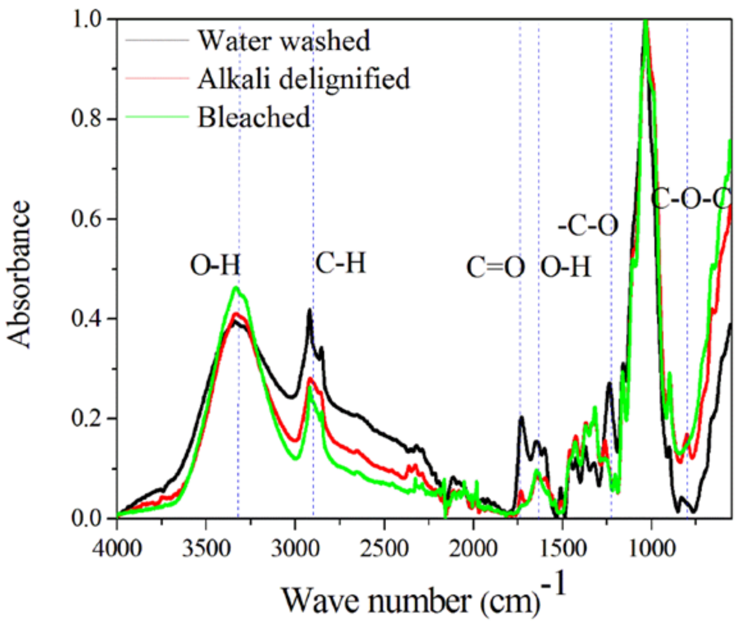

Figure S2. ATR FTIR spectra of *T. pungens* fibres before and after delignification and bleaching

Figure S3. Flowchart representing the procedures of nanofibrillated cellulose from *Triodia pungens*

Table S1. Crystallinity and crystalline domain size of water washed, delignified, bleached fibres and NFC of *T. pungens*

	Water washed fibre	Delignified fibre	Bleached fibre	NFC
Crystallinity index (%)	52	71	74	69
Crystalline domain size (nm)	2.9	4	4	4.3

Table S2. Comparison of the mechanical properties of nanopaper obtained from NFC prepared from the other sources of cellulose with spinifex NFC

Source of Cellulose	Mechanical Treatment	Young's Modulus (GPa)	Tensile Strain (%)	Tensile Stress (MPa)	Work at Fracture (MJ m^{-3})	Ref
Spinifex	Homogenization	3.2±0.2	18±0.2	84±5	12.3±2	
Sodium form washed wood pulp	Homogenization	11.2	7.2	230	12.3±2	1
TEMPO mediated wood pulp	Homogenization	1.4	16.6	83	10.5	2
Bleached soft wood	Homogenization	6.7±0.6	Tensile index (NM/g) 105.3±19		7.8	3
Bleached hard wood	Homogenization	6.3±0.6	Tensile index (NM/g) 91.7±25		5.7±2.1	3
Soft wood pulp with the DP=1100	Microfluidization	14.7	6.9	205	5.8±3.2	4
Beech-wood pulp	Homogenization	7.5	5	152	9.8	5
Bleached soft wood	Homogenization	2.3	11	80	-	6
Bleached hard wood	Homogenization	1	2	20	-	6

1. M. Osterberg, J. Vartiainen, J. Lucenius, U. Hippi, J. Seppala, R. Serimaa and J. Laine, *ACS Applied Materials & Interfaces*, 2013, **5**, 4640-4647.
2. H. Sehaqui, Q. Zhou, O. Ikkala and L. A. Berglund, *Biomacromolecules*, 2011, **12**, 3638-3644.
3. K. Spence, R. Venditti, O. Rojas, Y. Habibi and J. Pawlak, *Cellulose*, 2010, **17**, 835-848.
4. M. Henriksson, L. A. Berglund, P. Isaksson, T. Lindstrom and T. Nishino, *Biomacromolecules*, 2008, **9**, 1579-1585.
5. G.-A. Wolfgang, V. Stefan, O. Michael, T. Christian and K. Jozef, in *Functional Materials from Renewable Sources*, American Chemical Society, 2012, vol. 1107, ch. 1, pp. 3-16.
6. W. Stelte and A. R. Sanadi, *Industrial & Engineering Chemistry Research*, 2009, **48**, 11211-11219.