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1 Chemically extracted nanocellulose from sisal fibres by a simple and 2 industrially relevant process

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10 **Keywords:**

11 Sisal fibres; Cellulose nanofibres (CNFs); Cellulose films; acetylation, nanofibers

12 **Abstract:**

13 A novel type of acetylated cellulose nanofibre (CNF) was extracted successfully from sisal fibres using
14 chemical methods. Initially, a strong alkali treatment was used to swell the fibres, followed by a bleaching step
15 to remove the residual lignin and finally an acetylation step to reduce the impact of the intermolecular hydrogen
16 bonds in the nanocellulose. The result of this sequence of up-scalable chemical treatments was a pulp consisting
17 mainly of micro-sized fibres, which allowed simpler handling through filtration and purification steps and
18 permitted the isolation of an intermediate product with a high solid content. An aqueous dispersion of CNF
19 could be obtained directly from this intermediate pulp by simple magnetic stirring. As a proof of concept, the
20 dispersion was used directly for preparing a highly translucent CNF film, illustrating that there is no large
21 aggregates in the prepared CNF dispersion. Finally, CNF films with alkali extracts were also prepared, resulting
22 in flatter films with an increased mass yield and improved mechanical strength.

23 **1. INTRODUCTION**

24 Cellulose is the most abundant bio-derived polymer in the world, with a yearly production of about 10¹¹ tons
25 (Azizi Samir et al. 2005). This production originates mainly from plants, but there are other sources of cellulose
26 such as bacteria, tunicates and algae (Moon et al. 2011). Cellulose has the empirical formula (C₆H₁₀O₅) and is a
27 linear homopolysaccharide with hundreds to thousands of glucose units connected through 1-4-β-glucosidic
28 bonds. Cellulose is semicrystalline and therefore contains both amorphous and crystalline domains of various
29 types depending on the source of cellulose. Due to the recently increased focus on sustainability, lignocellulosic
30 materials in general, and cellulose in particular, have been investigated widely in search of novel application
31 fields such as biofuels (Baker and Keisler 2011), polymer reinforcement (Saheb and Jog 1999) and biomedical
32 applications (Czaja et al. 2007) (Lin and Dufresne 2014). A turning point in this development occurred when
33 Herrick et al. (Herrick et al. 1983) successfully isolated microfibrillated cellulose using mechanical methods to
34 break up the hierarchical structure of cellulose. The cellulosic fibres can be considered bundles of nanosized

35 cellulose fibres (CNFs) comprising of cellulose fibrils with a high aspect ratio, having diameters on the
36 nanoscale and lengths on the microscale. CNFs have been shown to have very interesting properties, such as a
37 specific Young's modulus that is 3.4 times higher than that of steel(Eichhorn et al. 2009). Research into
38 applications of the nanosized cellulose materials has increasingly caught the interest of the scientific
39 community(Lavoine et al. 2012), and the subject has been widely studied for applications such as
40 hydrogels(Chang and Zhang 2011), aerogels(Fischer et al. 2006), barrier coatings(Minelli et al. 2010) and
41 polymer reinforcement(Siró and Plackett 2010). Films containing such nanofibres are reported to have very
42 good mechanical properties(Siró and Plackett 2010), high transparency(Siró et al. 2011), good oxygen barrier
43 properties at low relative humidity as well as medium water vapour barrier properties(Lavoine et al. 2012) due
44 to the high water uptake of the nanofibres(Minelli et al. 2010).

45 In order to bring CNF applications to market, an industrially relevant method that can extract and break up the
46 strong association between fibres to prepare them for use in composites is required. Usually, the procedures for
47 obtaining CNFs are based on applying high shear forces to extracted fibres e.g. by grinding, micro fluidisation,
48 homogenisation or other similar techniques. These methods are usually highly energy demanding and are
49 performed in high dilution or using processes that are not directly up-scalable or cost-effective. Therefore
50 extensive research has been invested in finding methods for pre-treatment, such as enzymatic
51 treatments(Henriksson et al. 2007) or using (2,2,6,6-tetramethylpiperidin-1-yl)oxyl (TEMPO)(Saito et al. 2006)
52 for oxidation of the cellulose to decrease the energy consumption and weaken the hydrogen bonds between
53 fibres(Qing et al. 2013; Abdul Khalil et al. 2014). Here a method to obtain partially acetylated CNF, by
54 employing a simple chemical treatment followed by a low energy dispersion step, is presented. The intermediary
55 pulp achieved after the chemical treatments is easy to filter and dry, which makes it highly suitable for
56 transportation.

57 **2. EXPERIMENTAL**

58 **2.1 Materials and methods**

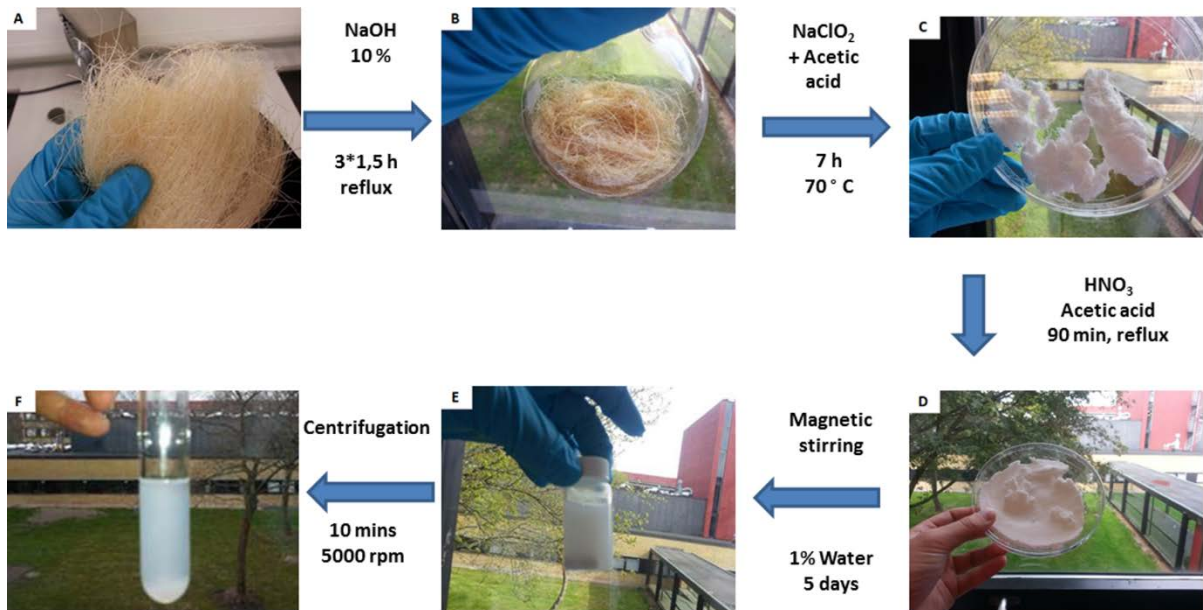
59 Cellulose nanofibres were extracted from sisal, which was kindly supplied by Expor Sisal S.L., while sodium
60 hydroxide, nitric acid (ACS reagent, 70%) and acetic acid (99%-100%) were purchased from Sigma Aldrich and
61 sodium chlorite (25 wt% in water) was obtained from Merck. All of the reagents were used as received. If not
62 specified, the analysis of CNF were done after centrifugation.

63 **2.1.1 Extraction and isolation of the acetylated CNF pulp (SMBA)**

64 Sisal fibres (50 g) were cut and rinsed with an aqueous solution of sodium hydroxide (1.5 L, 2 wt%) at 23°C for
65 16 hours. The rinsed fibres were isolated by filtration and washed with distilled water until constant pH of the
66 washing water was achieved. The alkali treatment (mercerisation) was repeated three times (1.5 hours at boiling
67 temperature) with a stronger alkali solution (1.5 L, 10 wt%) followed by filtration, after which the pulp (SM)
68 was suspended in distilled water (1.25 L), and the temperature was increased to 70°C. Once this temperature
69 was reached, acetic acid (8 mL) followed by sodium chlorite (NaClO₂, 25 wt%, 40 mL) was added once every
70 hour for 7 hours. Finally, the wet bleached pulp (approximately 30 g on dry mass) was isolated by filtration and
71 washed with distilled water until a constant pH was reached (SMB). Thereafter, this pulp was suspended in a
72 mixture of nitric acid (150 mL) and acetic acid (900 mL), and the mixture was stirred at boiling temperature for
73 90 minutes. The mixture was cooled by dilution with cold distilled water (ratio 1:5) and the acetylated pulp was

74 isolated by filtration. The product was rinsed with distilled water until a constant pH level was achieved and the
75 acetylated pulp (SMBA) was used without further purification.

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77

78 **Figure 1.** Overview of the CNF extraction protocol. A) Sisal fibres (S); B) Sisal fibres after alkali treatments (SM); C) Sisal
79 fibres after mercerisation and bleaching (SMB); D) Sisal fibres after mercerisation, bleaching and acetylation (SMBA); E)
80 SMBA after dispersion in water (CNF) and F) Stable CNF dispersion after centrifugation.

81

82 2.1.2 Extraction of lignin and hemicellulose for compounding (residue solution)

83 Sisal fibres (35 g) were mixed with an aqueous solution of sodium hydroxide (300 mL, 10 wt%), and the
84 mixture was refluxed for 2 hours. The fibres were filtered off and the filtrate was cooled and dialysed in a
85 regenerated cellulose membrane (Cellu Sep T3, MWCO 12,000-14,000) against distilled water until pH was
86 constant. The dialysed residue containing cellulose, lignin and hemicellulose was used without further
87 purification.

88 2.1.3 Preparation of CNF films

89 An aqueous dispersion of CNF in water (1 wt%) was prepared by dilution of the SMBA pulp with distilled
90 water followed by magnetic stirring for five days in an Erlenmeyer flask. The dispersion was centrifuged at
91 5000 rpm for 10 minutes, and the supernatant (0.8 wt%) was transferred to a Teflon mould. A translucent CNF
92 film was formed by concentration of the solution in a climatic chamber for one week at 19°C and 65% relative
93 humidity (RH), resulting in a translucent film with a thickness of 20-30 µm.

94 2.1.4 Preparation of the CNF/residue films

95 CNF/residue films were prepared from the aqueous CNF dispersion as described above for the CNF films.
96 Additionally, the supernatant was mixed with the residue solution (extracted cellulose, lignin and hemicellulose
97 from the mercerisation) to obtain a mixture with 70 wt% CNF and 30 wt% residue. The mixture was cast in a

98 Teflon mould and dried in a climatic chamber for one week at 19°C and 65% RH, resulting in the formation of a
99 translucent CNF/residue film with a thickness of 20-30 µm.

100 2.2 Characterization

101 Thermogravimetric analysis (TGA) was performed on a thermal TGA Q500 (TA) instrument from 25-600°C
102 with a heating rate of 10 K/min under nitrogen flow. Fourier transform infrared spectroscopy (FT-IR) was
103 obtained by triplicate on a Thermo-Fisher is50 FT-IR spectrometer equipped with a universal attenuated total
104 reflection (ATR) sampling accessory with a diamond crystal at a resolution of 4 cm⁻¹ in the range of 500-
105 4000 cm⁻¹.

106 The rheological data was obtained with a TA Instruments AR2000 controlled stress rotational rheometer using a
107 cone-and-plate geometry. The aluminium cone had a diameter of 60 mm and a 1° cone angle. Viscosity
108 measurements were obtained in steady shear at 25°C.

109 Scanning electron microscopy (SEM) of the sisal and processed fibres was carried out using a Hitachi T3030
110 with a 5 kV field. The CNF was characterised by SEM on an FEI Quanta 200 ESEM FEG. Transmission
111 electron micrographs were obtained using a Hitachi HT770 microscope operating at 100 kV. A drop of an
112 aqueous CNF suspension (0.2 % CNF) was deposited on a 200 mesh carbon/formvar copper grid (TED PELLA,
113 USA) and imaged without the addition of staining agents or other chemicals.

114 X-ray diffraction was performed using a Panalytical X'Pert Pro MPD-Ray diffractometer with an Ni-filtered Cu
115 K α radiation ($\lambda=1.54 \text{ \AA}$) source, a voltage of 45 kV, a current of 40 mA and scans from 5° to 60°. The
116 crystallinity index of the extracted fibres was calculated using the Buschle-Diller and Zeronian equation.

$$117 I_c = 1 - \frac{I_1}{I_2}$$

118

119 Where I_1 is the peak at $2\theta=18.8^\circ$ (amorphous peak) and I_2 is the peak at $2\theta=22.8^\circ$ (crystalline peak), I_c , the
120 crystalline index.

121 Optical properties were measured in triplicates using a UV-Vis spectrometer (Polar Star Omega) in the range of
122 200 nm – 1000 nm and a Gardener Haze-Gard Plus to analyse the transmittance, haze and clarity of the
123 specimens. Transmittance is the percentage of light transmitted through the sample, haze the amount of
124 transmitted light that is scattered more than 2.5° and clarity the amount of transmitted light that is scattered less
125 than 2.5°.

126 The surface properties of CNF films were estimated by advancing contact angle (CA) measurements on a
127 Dataphysics Contact Angle System OCA20. Specimens of 2 x 1 cm were placed on a glass slide and a drop of
128 water (6 µL) was deposited on the specimen surface. Thereafter, the needle was placed into the drop and the
129 advancing CA was determined as the constant value obtained with a flow of 0.5 µL/s. The CA was determined
130 as an average of at least five measurements.

131 Water absorption analysis was performed in duplicates based on the mass increase of samples (100-300 mg)
132 conditioned in a climatic chamber overnight at 23°C with a relative humidity of 10%, 25%, 50% and 75%,
133 respectively.

134 The mechanical properties were measured by duplicate in a DMA RSA3 (TA Instruments, USA) working in
135 tensile mode. The specimens (2 cm length and 0.5 cm width) were preconditioned at 23 °C and 50% RH for 24
136 hours prior to the measurement, which was carried at a speed of 1 mm/min with a distance between the fixtures
137 of 10 mm.

138

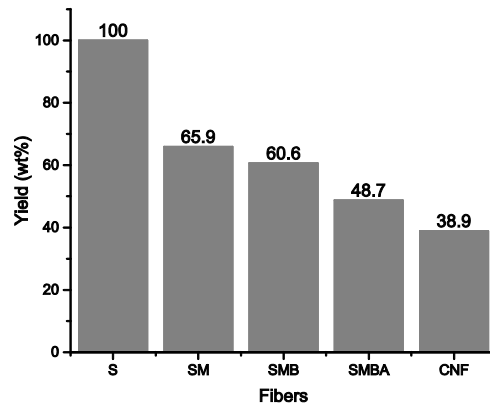
139 **3. RESULTS AND DISCUSSION**

140 **3.1 From raw materials to nanomaterials**

141 The developed method employs simple and industrially relevant processes for the conversion of sisal fibres into
142 CNF. Each of the steps is described in detail in the experimental section, as shown in Figure 1. An initial alkali
143 treatment was used to remove oil residues and impurities from the fibres. Three subsequent stronger alkali
144 treatments of sisal (S), to acquire mercerised fibres (SM, B in Figure 1), was used to swell the fibres and to
145 extract lignin and hemicellulose from the fibres. The repeated mercerisation minimized adsorption of these
146 impurities on the surface of the fibres compared to one longer mercerization step. After alkali treatment the
147 fibres were light-brown in colour, which is attributed to deposition of the extracted lignin on the surface of the
148 fibres. The deposited lignin was removed in the following bleaching step (SMB, C in Figure 1), and finally the
149 influence of hydrogen bonds between the fibres was reduced through acetylation of the fibres (SMBA, D in
150 Figure 1). This extraction protocol is based on two separate modifications of the fibres. Firstly, the alkali
151 treatment swells the fibres, breaks the strong association, due to hydrogen bonds, between the cellulose chains
152 and opens up the structure to additional chemical treatments (Mwaikambo and Ansell 1999). Secondly, in the
153 swollen state, the hydroxyl groups from the cellulose nanofibres are grafted with acetate groups, which thereby
154 permanently reduces the energy required to break the strong association between cellulose nanofibres in
155 subsequent processing steps.

156 The result of this sequence of chemical treatments was a pulp consisting mainly of modified micro-sized fibres,
157 which is easy to transport or store as a precursor for later preparation of CNF dispersions. An aqueous
158 dispersion of CNF was obtained directly from this intermediate by magnetic stirring (product E in Figure 1), and
159 any agglomerates were removed by centrifugation.

160 The yield of cellulosic material after each step of the extraction protocol is shown in Figure 2.



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Figure 2. Mass yield of the treated fibres after drying of a sample of the respective suspensions, where the CNF is the supernatant obtained after centrifugation in the last step (all results are on a dry basis).

164

The amount of extracted cellulose fibres depends strongly on the type of fibres used for the process. Sisal fibres are generally reported to consist of about 60-70 wt% cellulose, 10-15 wt% hemicellulose and 8-12 wt% lignin(Bismarck et al. 2001; Mondragon et al. 2014). After all of the chemical treatments and purification steps that removes the majority of the lignin and hemicellulose, 39 wt% of the original sisal fibres were converted into a stable CNF dispersion, which corresponds to an extraction of approximately 60 wt% of the total amount of cellulose from the sisal fibres. This compares to other extraction protocols, where e.g. 65 wt% of cellulose nanofibres have been extracted from cotton(de Morais Teixeira et al. 2010) or 50-60 wt% were extracted from softwood(Tejado et al. 2012).

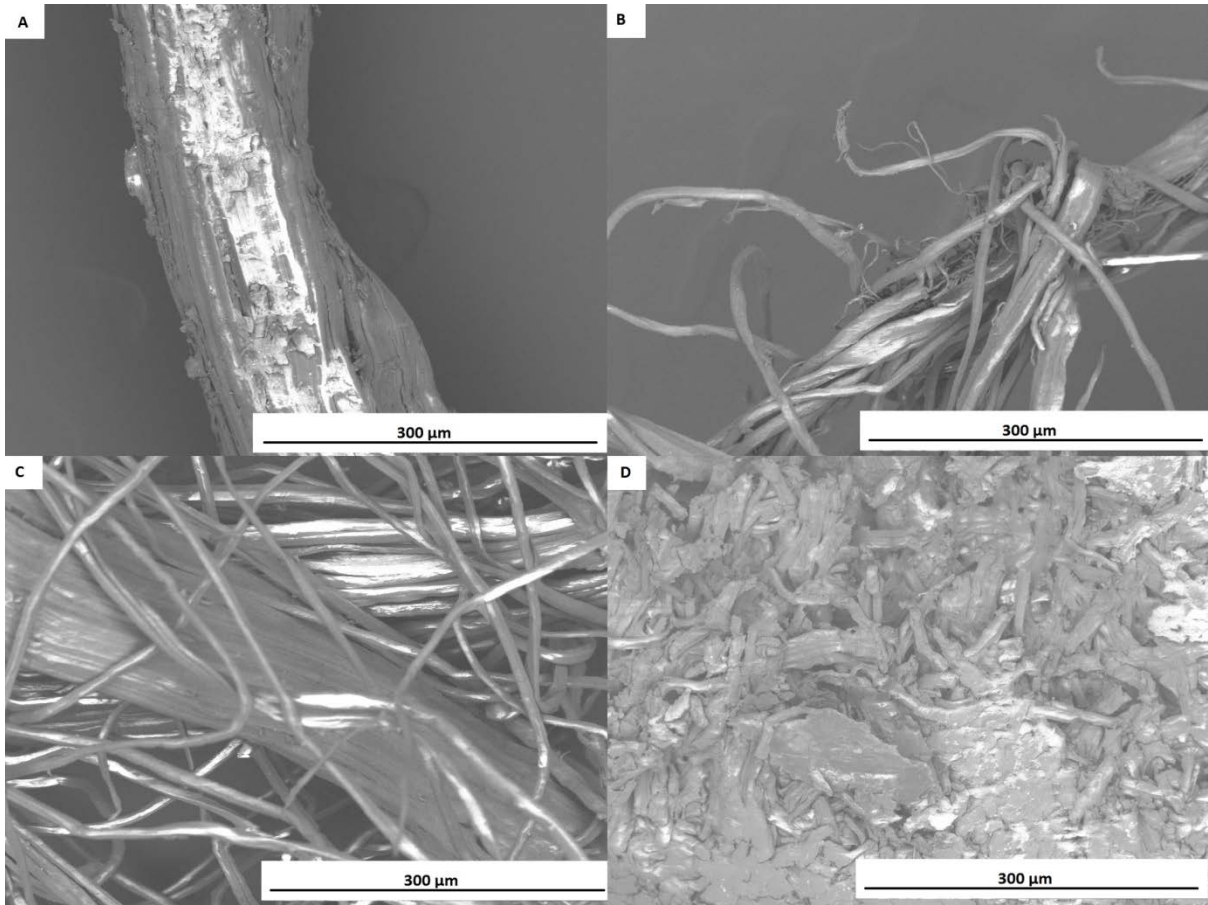
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172 **3.2 Characterization of the pulp**

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The isolated material was characterised by SEM after each step, in order to illustrate the effects of each of the treatments, as shown in Figure 3.

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Figure 3. SEM pictures of the fibres at the various stages of the process showing the transgression from large fibre bundles to the fully treated acetylated fibres in the final pulp. A) Sisal fibres B) fibres after alkali treatments C) fibres after bleaching D) fibres after acetylation.

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The micrographs in Figure 3 show how each step in the process affects the fibres. The strong alkali treatment swells the fibres and results in the formation of free individual fibres (SM). In the bleaching step even more separated fibres are produced (SMB) due to the removal of the remaining lignin, which reduces the cohesion between the fibrils. Finally after the acetylation step the structure of the macroscopic sisal fibres have been completely removed, resulting in the formation of a more uniform mass consisting of much smaller fibres. The results of the extraction protocol in terms of chemical and thermal properties can be seen in Figure 4.

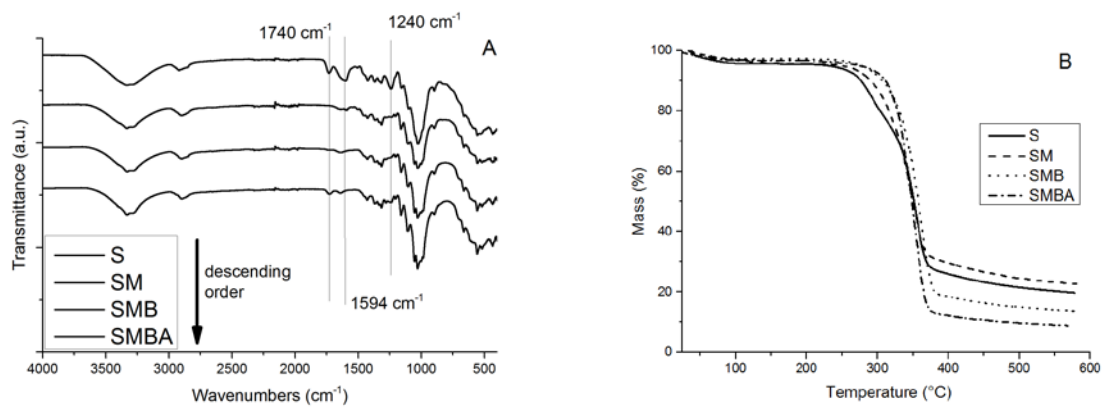


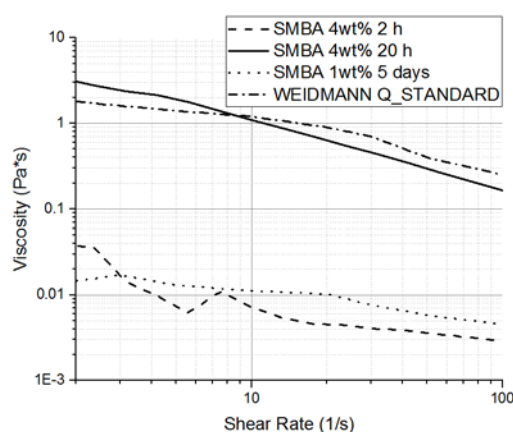
Figure 4. Analysis of the extracted CNF at each step in the process by FT-IR (A) and by TGA (B).

185 In Figure 4a the chemical changes to the fibres are illustrated by changes in the IR spectra. Here the removal of
186 the majority of hemicellulose during the first alkali treatment from S to SM can be seen through disappearance
187 of the peaks at 1740 cm^{-1} (which are related to acetyl and ester groups, characteristic of hemicellulose) and at
188 1240 cm^{-1} (C-O stretching vibration of the hemicellulose). In addition to this, the disappearance of the peak at
189 1594 cm^{-1} (related to the C-C in the plane symmetrical stretching of aromatic rings, characteristic of lignin)
190 from SM to SMB shows that the most part lignin has been removed from the fibre. This is corroborated by the
191 fact that the fibres are completely white after bleaching. The removal of a large part of the hemicellulose is
192 confirmed by TGA in Figure 4b. Hemicellulose usually degrades between $200\text{-}400^\circ\text{C}$, which results in an
193 increase in the onset of thermal degradation after both the alkali treatment and the bleaching step compared to
194 the pure sisal fibres.

195 Finally, acetylation of the pulp in the last step of the process is confirmed by reappearance of the peak at
196 1740 cm^{-1} , attributed to new acetate groups. The intensity of the carbonyl stretch indicates that only a partial
197 acetylation has taken place, which was also confirmed by the low degree of substitution (10%) determined using
198 the method published by Kim et al. (Kim et al. 2002). The extracted residues were also analysed (SI-Figure 1)
199 and it was found that the alkali treatments removed not only the majority of lignin and hemicellulose, but also
200 significant amounts of cellulose.

201 3.2 Characterization of the CNF

202 Introduction of acetate groups on the surface of the fibres, however, is not enough to fully separate the
203 nanofibres. This acetylation results in reduced cohesion between the nanofibres, but a very small amount of
204 energy is still required in order to separate the fibres. Magnetic stirring is sufficient to separate the nanofibres as
205 shown by the increase in viscosity of a 4 wt% SMBA dispersion stirred for respectively 2 and 20 hours, as
206 shown in Figure 5.

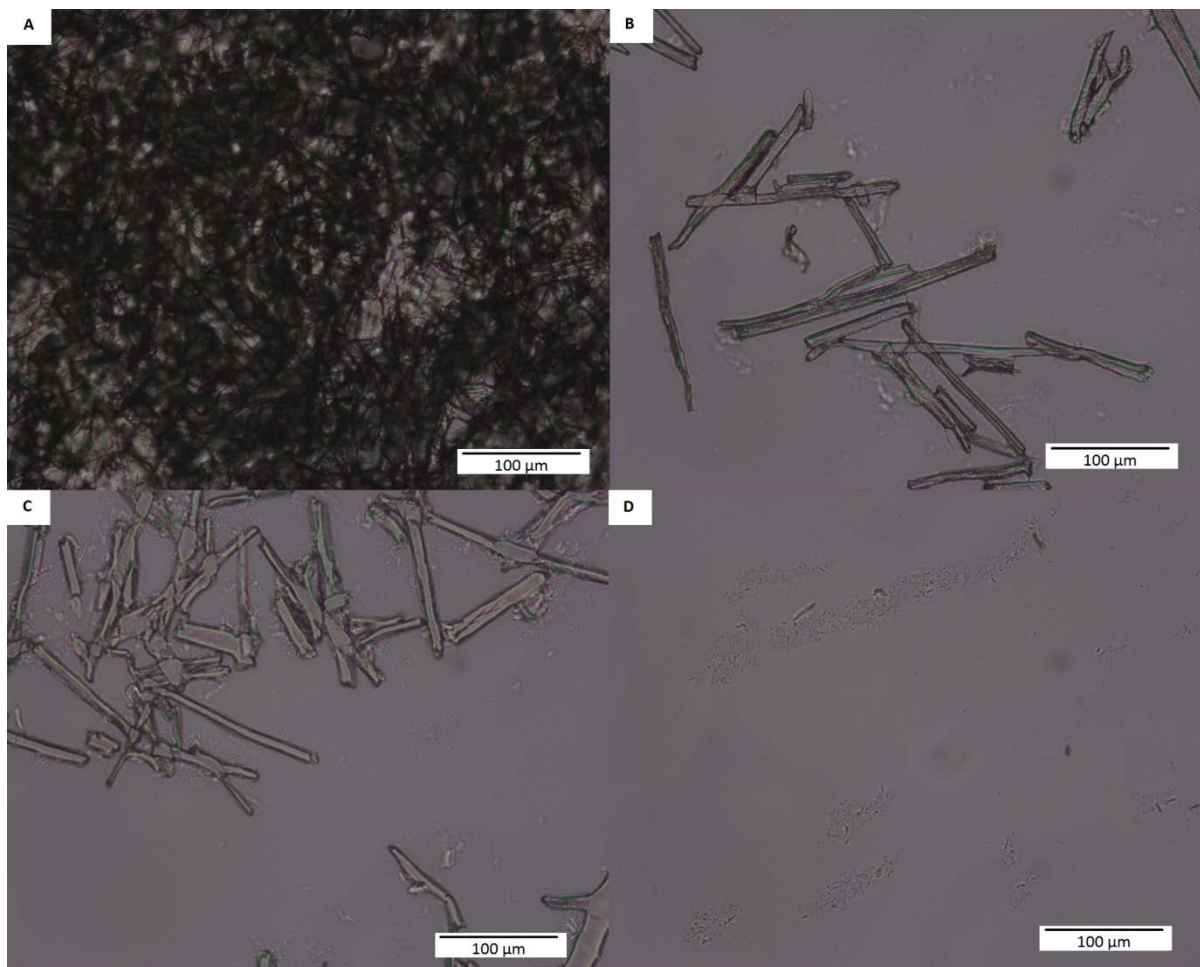


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208 **Figure 5.** Viscosity of a 4 wt% SMBA solution in water after magnetic stirring for 2 hours and 20 hours compared to a
209 commercial CNF (Weidmann Q standard) and to a 1wt% CNF solution that has been stirred for 5 days.

210 The viscosity is dramatically increased between 2 and 20 hours of magnetic stirring, due to dispersion of the
211 CNF in water. With continued stirring the fibres becomes gradually more and more separated and eventually the
212 sample reaches the rheological percolation threshold, resulting in formation of a CNF network and a significant
213 increase in viscosity. The viscosity of the resulting dispersion after 20 hours was similar to a commercially

214 available CNF produced by Weidmann (Q standard). The increased viscosity proves that the very soft
215 mechanical treatment successfully disperses the CNF. However, at 4 wt% CNF the viscosity was so high after
216 20 hours that magnetic stirring was no longer powerful enough to efficiently stir the suspension (it formed a
217 hydrogel), which prevents the complete dispersion of the CNF. Therefore the concentration was reduced to
218 1 wt%, which it was possible to stir for 5 days without reaching the rheological percolation threshold. Samples
219 of this solution were analysed by optical microscopy after different stirring times as shown in Figure 6.
220

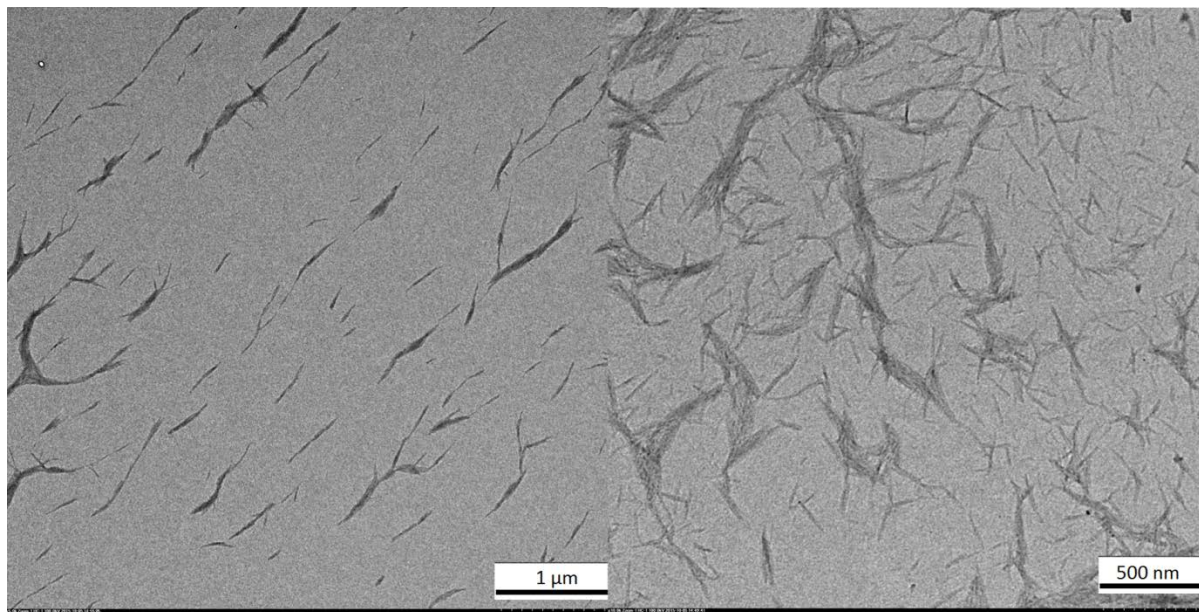


221
222 **Figure 6.** Optical micrographs of a 1wt% mixture of the acetylated pulp (SMBA) after magnetic stirring in water for 0 min
223 (A), 15 min (B), 1 hour (C) and 48 hours (D).

224 Simple magnetic stirring breaks up the aggregates and ultimately results in a stable aqueous dispersion of the
225 nanofibres after 48 hours. After two days of magnetic stirring, it is no longer possible to see large fibres in the
226 optical microscope, thus suggesting that the majority of the fibres are below microscale in size.

227 In order to evaluate the dimensions of the prepared nanofibres, a drop of a 0.6 wt% CNF dispersion was casted
228 on an aluminium film, resulting in the formation of a thin film with a film thickness of around 100 nm. The
229 prepared film was sputtered and investigated by SEM, which showed a uniform distribution of nanofibres on the

230 surface (SI-Figure 2). The size of the cellulose nanofibres were additionally investigated using transmission
231 electron microscopy (TEM) as shown in Figure 7.

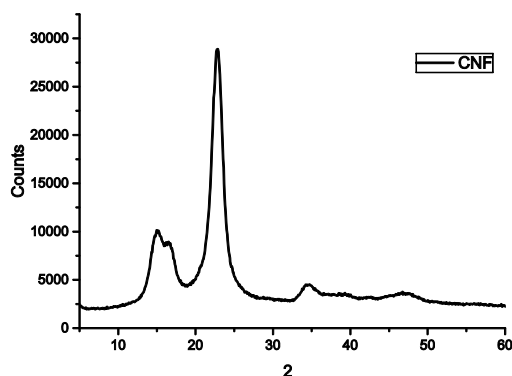


232

233 **Figure 7.** Analysis of the structure of the isolated CNF by TEM at different magnifications.

234 From both SEM and TEM it is clear that no large fibres are present in the film casted from the dispersion. The
235 fibres are estimated to have a diameter of 27 ± 13 nm and a length of 658 ± 290 nm with minor aggregates of
236 approximately 160 ± 75 nm in diameter and 0.90 ± 0.42 μm in length. The fibres are shorter than what has
237 been obtained when CNFs are prepared by using, for example, TEMPO-mediated oxidation and mechanical
238 methods such as homogenisation, ultrasound or grinding (Moon et al. 2011), which is attributed to the harsh
239 conditions employed during the extraction process.

240 The processing of cellulosic materials is known to affect the crystallinity, and it was therefore investigated by
241 X-ray diffraction (XRD), as shown in Figure 8.



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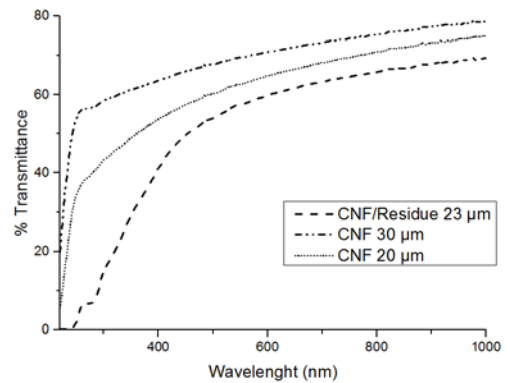
243 **Figure 8.** XRD analysis of the purified CNF.

244 The XRD spectrum shows the expected peaks from a cellulose material with peaks at $2\theta=15.13$ and $2\theta=22.88^\circ$.
245 The degree of crystallinity of the extracted fibres was calculated based on the peak at 22.88° , and it was
246 determined to be 84.2%. This value is comparable to the crystallinities reported in the literature where sisal-

247 based CNF was reported to have a crystallinity of 93% (Siqueira et al. 2010) and from TEMPO oxidized CNF
248 having a crystallinity of 59-92% (Lavoine et al. 2012).

249 3.3 CNF film properties

250 The CNF dispersion was used to prepare large CNF films by solution casting of either the prepared CNF
251 dispersion directly or by combination of the CNF dispersion and the extracted residues. The prepared films have
252 a high clarity and a good transparency as shown in Figure 9.



253 **Figure 9. Left: Optical image of the prepared film (pure CNF on the left and CNF with residue solution on the right);**
254 **Right: Transmittance of the CNF films determined by UV-vis spectroscopy.**

255 The film prepared from the pure CNF is fully translucent, and has a high clarity (69,8% clarity, 26,7% haze)
256 whereas the film with the added residues (extracted cellulose, lignin and hemicellulose from the mercerisation
257 step) resulted in a light-brown but more uniform film with a slightly reduced clarity (44,6% clarity, 36,2% haze)
258 (see Table 1, supporting information). The CNF/Residue film additionally had a decreased UV transmittance,
259 due to the presence of lignin that acts as a UV absorber and as antioxidant. Ultimately, the CNF are intended for
260 use in composites for packaging materials and for this application a reduced UV transmittance as well as the
261 antioxidative properties of the lignin are interesting properties. The residues were also incorporated into the
262 film, in order to investigate if they could potentially decrease the water sorption of the films as well as to
263 investigate any effects on film forming. The cross-sectional view of the films by SEM can be seen in Figure 10.

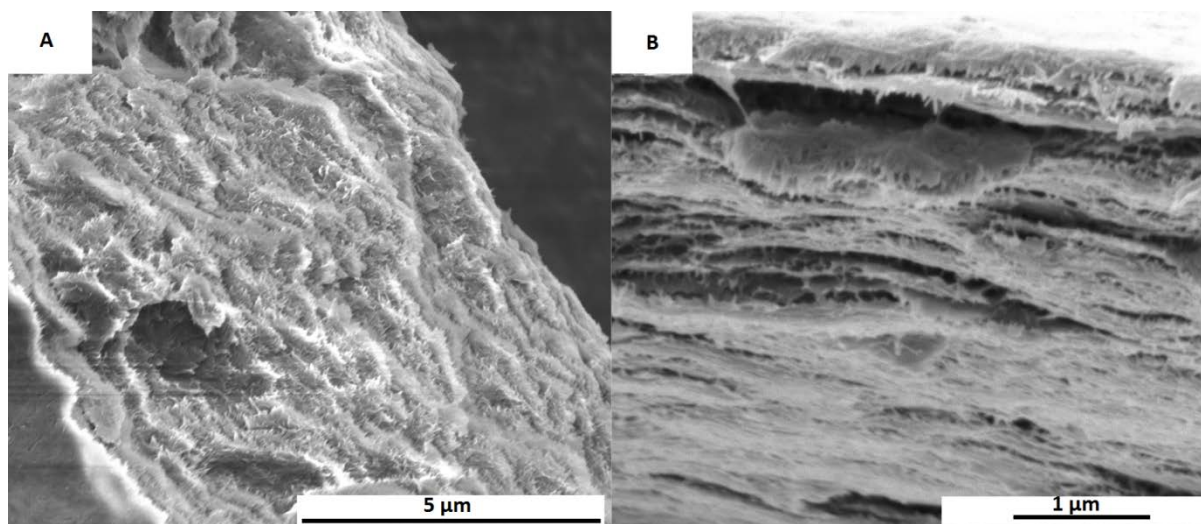


Figure 10. SEM analysis of a fractured cross section of a) Neat CNF film and B) CNF/Residue film.

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265

266 The SEM micrographs show that there are no large agglomerates on the fractured interphase from the films,
 267 (SEMs of the surface view of the films can be seen in the supporting information in SI-Figure 3 and 4). The
 268 micrographs also show a clear differences in the layered structures of the two films, where the CNF/Residue
 269 film is clearly much more compact compared to the pristine CNF film, which appears to have a more open
 270 structure. This feature of the CNF/Residue film is attributed to the lignin and hemicellulose in the residue that
 271 work as binders and make a stronger bonding with CNF through hydrogen bonding, which results in a reduced
 272 swelling of the CNF during the drying stage.

273 The mechanical properties of the films were investigated (SI-Table2), showing that in particular the pure CNF
 274 film had lower tensile strength and elongation at break compared to other nanocellulose films(Henriksson et al.
 275 2008; Siró et al. 2011). This is attributed to the partial acetylation of the fibres, which results in less hydrogen
 276 bonding between the fibres in the neat film. However, the degree of substitution is not high enough to affect the
 277 transparency of the film, as would be expected from a fully acetylated CNF. The pure CNF films were very
 278 brittle and had an uneven surface, which made the mechanical analysis very difficult. In contrary to this, the
 279 CNF/Residue films were more uniform and easier to handle, which resulted in a significant increase in tensile
 280 strength and elongation. CNF films, apart from their brittleness, have been reported to have good properties at
 281 low relative humidity, but their barrier properties dramatically decreases with increasing relative humidity(Aulin
 282 et al. 2010; Minelli et al. 2010). For this reason, any process that could decrease water sorption would be very
 283 useful for new applications of CNF films. The CNF nanomaterials prepared here are partially acetylated, which
 284 could potentially affect water sorption. In an effort to elucidate this, the advancing water contact angles of the
 285 prepared films were determined. Both films have comparable water contact angles of $48.8^\circ \pm 2.7$ for the CNF
 286 film and $52.4^\circ \pm 1.1$ for the CNF/Res film, which is similar to other non-functionalised nanocelluloses (41.2°
 287 (Rodionova et al. 2010), $50-60^\circ$ (Siqueira et al. 2010), 50° (Wu et al. 2014)). Apparently the partial acetylation
 288 does not significantly increase the hydrophobicity of the thin films, which corroborates the low degree of
 289 substitution (10%). The actual water sorption of the films can be seen in the supporting information (SI-Figure
 290 5). The observed water sorption, approximately 2,6% at 23°C and 50% rel. humidity for both types of films, was
 291 slightly decreased compared to other nanocellulose films, where CNF and nanocrystalline cellulose have been

292 shown to have water sorption of 6.5% at 25°C and 50% rel. humidity (Belbekhouche et al. 2011), or 4% at 35°C
293 and 50% rel. humidity for enzymatically pre-treated CNF(Minelli et al. 2010).

294 The mechanical properties of these materials are too poor to allow a direct application of these materials.
295 However, the process produces partially acetylated CNFs, which makes these amphiphilic materials easier to
296 disperse in organic media, and thereby makes them well suited for use as reinforcement agents in e.g. poly(lactic
297 acid) (PLA). Recently CNF composites in PLA were shown to have superior thermomechanical resistance and
298 enhanced barrier properties (PLA/CNF 1% showed a 64% of decrease on oxygen transmission rate, a 46% of
299 decrease on water vapour transmission rate)(Trifol et al. 2016a), which were greatly improved when both CNF
300 and a commercially available clay (C30B) were used to reinforce PLA (PLA/CNF 5%/C30B 5% showed a a
301 reduction of up to 90% in OTR and a further reduction in the water vapour transmission rate (WVTR) of up to
302 76% (Trifol et al. 2016b).

303 **4. CONCLUSIONS**

304 In this study a method is presented whereby a partially acetylated CNF can be prepared by employing a
305 chemical treatment protocol. This method of extracting cellulose nanofibres has several advantages. The raw
306 pulp produced after the chemical treatments is produced in a high yield of 48% and can be easily filtered and
307 purified. The isolated SMBA pulp does not form hydrogels, and can be reduced to a water content of 50%,
308 resulting in a potential reduction in transportation costs of the pulp. The chemicals used in the process are very
309 common and not particularly expensive. Moreover, it is possible through well-established processes in the paper
310 industry to reclaim these chemicals. The process directly affords acetylated fibres, which results in easy
311 dispersion by low energy magnetic stirring. The prepared dispersions were seen to contain nanofibres, with no
312 large aggregates present, as illustrated by both SEM and TEM as well as through preparation of highly
313 translucent CNF films. In an attempt to improve the film forming properties of the CNF films the alkali residue
314 was reintroduced into the CNF films, which lead not only to an overall higher mass yield, but also to flatter
315 films, which illustrated the potential of the residue as a novel cementing agent for the CNF films. Additionally,
316 the inclusion of the alkali residue resulted in a significant reduction in the UV transmittance.

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325 **5. REFERENCES**

- 326 Abdul Khalil HPS, Davoudpour Y, Islam MN, Mustapha A, Sudesh K, Dungani R, Jawaid M (2014) Production
327 and modification of nanofibrillated cellulose using various mechanical processes: a review. *Carbohydr*
328 *Polym* 99:649–65. doi: 10.1016/j.carbpol.2013.08.069
- 329 Aulin C, Gällstedt M, Lindström T (2010) Oxygen and oil barrier properties of microfibrillated cellulose films
330 and coatings. *Cellulose* 17:559–574. doi: 10.1007/s10570-009-9393-y

- 331 Azizi Samir MAS, Alloin F, Dufresne A (2005) Review of recent research into cellulosic whiskers, their
 332 properties and their application in nanocomposite field. *Biomacromolecules* 6:612–26. doi:
 333 10.1021/bm0493685
- 334 Baker E, Keisler JM (2011) Cellulosic biofuels: Expert views on prospects for advancement. *Energy* 36:595–
 335 605. doi: 10.1016/j.energy.2010.09.058
- 336 Belbekhouche S, Bras J, Siqueira G, Chappey C, Lebrun L, Khelifi B, Marais S, Dufresne A (2011) Water
 337 sorption behavior and gas barrier properties of cellulose whiskers and microfibrils films. *Carbohydr*
 338 *Polym* 83:1740–1748. doi: 10.1016/j.carbpol.2010.10.036
- 339 Bismarck A, Aranberri-Askargorta I, Springer J, Mohanty AK, Misra M, Hinrichsen G, Czaplá S (2001) Surface
 340 characterization of natural fibers; surface properties and the water up-take behavior of modified sisal and
 341 coir fibers. *Green Chem* 3:100–107. doi: 10.1039/b100365h
- 342 Chang C, Zhang L (2011) Cellulose-based hydrogels: Present status and application prospects. *Carbohydr*
 343 *Polym* 84:40–53. doi: 10.1016/j.carbpol.2010.12.023
- 344 Czaja WK, Young DJ, Kawecki M, Brown RM (2007) The future prospects of microbial cellulose in biomedical
 345 applications. *Biomacromolecules* 8:1–12. doi: 10.1021/bm060620d
- 346 de Moraes Teixeira E, Corrêa AC, Manzoli A, de Lima Leite F, de Oliveira CR, Mattoso LHC (2010) Cellulose
 347 nanofibers from white and naturally colored cotton fibers. *Cellulose* 17:595–606. doi: 10.1007/s10570-
 348 010-9403-0
- 349 Eichhorn SJ, Dufresne A, Aranguren M, Marcovich NE, Capadona JR, Rowan SJ, Weder C, Thielemans W,
 350 Roman M, Renneckar S, Gindl W, Veigel S, Keckes J, Yano H, Abe K, Nogi M, Nakagaito A. N,
 351 Mangalam A, Simonsen J, Benight A. S, Bismarck A, Berglund L A., Peijs T (2009) Review: current
 352 international research into cellulose nanofibres and nanocomposites. *J Mater Sci* 45:1-33. doi:
 353 10.1007/s10853-009-3874-0.
- 354 Fischer F, Rigacci A., Pirard R, Berthon-Fabry S, Achard P (2006) Cellulose-based aerogels. *Polymer*
 355 47:7636–7645. doi: 10.1016/j.polymer.2006.09.004
- 356 Henriksson M, Berglund L A., Isaksson P, Lindström T, Nishino T (2008) Cellulose nanopaper structures of
 357 high toughness. *Biomacromolecules* 9:1579–1585. doi: 10.1021/bm800038n
- 358 Henriksson M, Henriksson G, Berglund LA, Lindström T (2007) An environmentally friendly method for
 359 enzyme-assisted preparation of microfibrillated cellulose (MFC) nanofibers. *Eur Polym J* 43:3434–3441.
 360 doi: 10.1016/j.eurpolymj.2007.05.038
- 361 Herrick FW, Casebier RL, Hamilton KJ, Sandberg KR (1983) Microfibrillated Cellulose: Morphology and
 362 Accessibility. *J Appl Polym Sci Appl Polym Symp* 37:797–813.
- 363 Kim D, Nishiyama Y, Kuga S (2002) Surface acetylation of bacterial cellulose. *Cellulose* 361–368.
- 364 Lavoine N, Desloges I, Dufresne A, Bras J (2012) Microfibrillated cellulose - its barrier properties and
 365 applications in cellulosic materials: a review. *Carbohydr Polym* 90:735–64. doi:
 366 10.1016/j.carbpol.2012.05.026
- 367 Lin N, Dufresne A (2014) Nanocellulose in biomedicine: Current status and future prospect. *Eur Polym J*
 368 59:302–325. doi: 10.1016/j.eurpolymj.2014.07.025
- 369 Minelli M, Baschetti MG, Doghieri F, Ankerfors M, Lindström T, Siró I, Plackett D (2010) Investigation of
 370 mass transport properties of microfibrillated cellulose (MFC) films. *J Memb Sci* 358:67–75. doi:
 371 10.1016/j.memsci.2010.04.030
- 372 Mondragon G, Fernandes S, Retegi A, Peña C, Algar I, Eceiza A, Arbelaz A (2014) A common strategy to
 373 extracting cellulose nanoentities from different plants. *Ind Crops Prod* 55:140–148. doi:
 374 10.1016/j.indcrop.2014.02.014
- 375 Moon RJ, Martini A, Nairn J, Simonsen J, Youngblood J (2011) Cellulose nanomaterials review: structure,
 376 properties and nanocomposites. *Chem Soc Rev* 40:3941–94. doi: 10.1039/c0cs00108b
- 377 Mwaikambo LY, Ansell MP (1999) The effect of chemical treatment on the properties of hemp, sisal, jute and
 378 kapok for composite reinforcement. *Die Angew Makromol Chemie* 272:108–116. doi:
 379 10.1002/(SICI)1522-9505(19991201)272:1<108::AID-APMC108>3.0.CO;2-9

- 380 Qing Y, Sabo R, Zhu JY, Agarwal U, Cai Z, Wu Y (2013) A comparative study of cellulose nanofibrils
381 disintegrated via multiple processing approaches. *Carbohydr Polym* 97:226–34. doi:
382 10.1016/j.carbpol.2013.04.086
- 383 Rodionova G, Lenes M, Eriksen Ø, Gregersen Ø (2010) Surface chemical modification of microfibrillated
384 cellulose: improvement of barrier properties for packaging applications. *Cellulose* 18:127–134. doi:
385 10.1007/s10570-010-9474-y
- 386 Saheb DN, Jog JP (1999) Natural fiber polymer composites: A review. *Adv Polym Technol* 18:351–363. doi:
387 10.1002/(SICI)1098-2329(199924)18:4<351::AID-ADV6>3.0.CO;2-X
- 388 Saito T, Nishiyama Y, Putaux JL, Vignon M, Isogai A (2006) Homogeneous suspensions of individualized
389 microfibrils from TEMPO-catalyzed oxidation of native cellulose. *Biomacromolecules* 7:1687–1691. doi:
390 10.1021/bm060154s
- 391 Siqueira G, Bras J, Dufresne A (2010) New process of chemical grafting of cellulose nanoparticles with a long
392 chain isocyanate. *Langmuir* 26:402–411. doi: 10.1021/la9028595
- 393 Siró I, Plackett D (2010) Microfibrillated cellulose and new nanocomposite materials: a review. *Cellulose*
394 17:459–494. doi: 10.1007/s10570-010-9405-y
- 395 Siró I, Plackett D, Hedenqvist M, Ankerfors M, Lindström T (2011) Highly transparent films from
396 carboxymethylated microfibrillated cellulose: The effect of multiple homogenization steps on key
397 properties. *J Appl Polym Sci* 119:2652–2660. doi: 10.1002/app.32831
- 398 Tejado A, Alam MN, Antal M, Yang H, van de Ven TGM (2012) Energy requirements for the disintegration of
399 cellulose fibers into cellulose nanofibers. *Cellulose* 19:831–842. doi: 10.1007/s10570-012-9694-4
- 400 Trifol J, Plackett D, Sillard C, Hassager O, Daugaard AE, Bras J, Szabo P (2016a) A comparison of partially
401 acetylated nanocellulose, nanocrystalline cellulose, and nanoclay as fillers for high-performance
402 polylactide nanocomposites. *J Appl Polym Sci* 133:43257. doi: 10.1002/app.43257
- 403 Trifol J, Plackett D, Sillard C, Szabo P, Bras J, Daugaard AE (2016b) Hybrid poly(lactic
404 acid)/nanocellulose/nanoclay composites with synergistically enhanced barrier properties and improved
405 thermomechanical resistance. *Polym Int.* 65:988-995. doi: 10.1002/pi.5154
- 406 Wu C-N, Saito T, Yang Q, Fukuzumi H, Isogai A (2014) Increase in the Water Contact Angle of Composite
407 Film Surfaces Caused by the Assembly of Hydrophilic Nanocellulose Fibrils and Nanoclay Platelets. *ACS*
408 *Appl Mater Interfaces* 6:12707–12712. doi: 10.1021/am502701e
- 409