Cationization of lignocellulosic fibers with betaine in deep eutectic solvent: facile route to charge stabilized cellulose and wood nanofibers

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KEYWORDS: cationization, betaine, deep eutectic solvent, cellulose nanofibers, wood nanofibers

ABSTRACT In this study, a deep eutectic solvent (DES [based on triethylmethylammonium chloride (TEMA) and imidazole]) was used as a reaction medium for cationization of cellulose fibers with trimethylglycine (betaine) hydrochloride in the presence of p-Toluenesulfonyl (tosyl) chloride. Cellulose betaine ester with a cationic charge up to 1.95 mmol/g was obtained at mild reaction conditions (four hours at 80 °C). The reaction was further demonstrated in the fabrication of cationic cellulose nanofibers (CCNFs) by a mild mechanical disintegration of cationized cellulose. In addition to CCNFs, cationic wood nanofibers (CWNFs) were produced directly from groundwood pulp (GWP) with a high lignin content (27 w%). Individualized CCNFs and CWNFs had a fiber diameter of 4.7 ± 2.0 and 3.6 ± 1.3 nm, respectively, whereas some larger fiber aggregates (diameter below 200 nm) were also observed, especially in the case of CWNFs.

Introduction

Chemical modification of biomass is an important part of the biorefinery concept where natural renewable materials are utilized as sources for novel chemicals and products. Although natural biomasses have many useful properties by themselves, chemical modifications can significantly improve their feasibility in scientific and industrial applications.(John & Anandjiwala, 2008; Laurichesse & Avérous, 2014; O’Connell, Birkinshaw, & O’Dwyer, 2008; Wan Ngah & Hanafiah, 2008) Using chemical modification, properties such as surface activity and solubility can be altered to meet the requirements of target applications.

Cellulose nanofibers (CNFs) are biomass-derived nanomaterials mainly obtained from delignified and bleached cellulose pulp (Kim et al., 2015; Klemm et al., 2011) and widely studied for example as reinforcement in composites. (J. Li et al., 2018; Satyanarayana, Arizaga, & Wypych, 2009) The high energy consumption of CNF production can notably be decreased by chemical modifications. Moreover, the modifications can be used to introduce desired functionalities on the surface of nanofibers.(Klemm et al., 2011) Most often these modifications are based on anionic carboxyl acid groups,(Isogai, Saito, & Fukuzumi, 2011) that create electrostatic repulsion and higher osmotic pressure within fibers, which both help the liberation of CNFs during the mechanical disintegration.(Lavoine, Bras, Saito, & Isogai, 2016)

In addition to anionic CNFs, cationic CNFs (CCNFs) have proven to be useful material for several applications, including flocculation(Liimatainen, Suopajarvi, Sirviö, Hormi, & Niinimäki, 2014; Suopajarvi, Sirviö, & Liimatainen, 2017a) and water purification.(H. Sehaqui, Larraya, Tingaut, & Zimmermann, 2015; Houssine Sehaqui et al., 2016) Cationization has been carried out using methods such as etherification with 2,3-epoxypropyl trimethyl ammonium chloride(Pei, Butchosa, Berglund, & Zhou, 2013; H. Sehaqui et al., 2015) and sequential periodate oxidation and imination.(Liimatainen et al., 2014; Suopajarvi et al., 2017a) These methods, however, rely

on toxic and expensive chemicals. Thus, chemicals, such as natural occurring betaine, have been proposed as alternative ways for cationization of cellulose(Ma, Yan, Meng, & Zhang, 2014); but these chemicals have rarely been used in CCNFs production.

In addition to use of bleached cellulose pulp for cellulosic nanomaterial production, use of less processed natural fibers (e.g., fibers where the lignin, hemicellulose and other materials are not removed) have lately been investigated.(Hassan, Berglund, Hassan, Abou-Zeid, & Oksman, 2018; Herrera et al., 2018; Rojo et al., 2015; Visanko et al., 2017; Winter et al., 2017) For this purpose, carboxylation using (2,2,6,6-Tetramethylpiperidin-1-yl)oxyl (TEMPO)(Okita, Saito, & Isogai, 2009) and nitric acid or a nitric acid-sodium nitrite mixture(Sharma, Joshi, Sharma, & Hsiao, 2017) have been used. However, these methods can simultaneously solubilize non-cellulosic materials (e.g. lignin) and lead to yield losses and alteration of CNF surface characteristics. To obtain wood nanofibers (WNF) containing most of the natural lignin intact, few mechanical methods have been utilized so far. Recently, anionic WNF was produced using succinylation of groundwood pulp (GWP) in deep eutectic solvents (DES).(Sirviö & Visanko, 2017) During the succinylation, most of the original lignin (and other non-cellulosic materials) was preserved after chemical modification.

DESs are among the most promising new solvents and reagents for cellulose derivatization(Abbott, Bell, Handa, & Stoddart, 2005, 2006; Park, Oh, & Choi, 2013) and for production of various nanosized celluloses.(Hosseini, Annamalai, Wang, Martin, & Amiralian, 2017; P. Li, Sirviö, Haapala, & Liimatainen, 2017; Liu et al., 2017; Selkälä, Sirviö, Lorite, & Liimatainen, 2016; Sirviö, Visanko, & Liimatainen, 2015; Suopajarvi, Sirviö, & Liimatainen, 2017b) DESs can be produced by simply mixing two or more components together, resulting in a solution having a lower melting point than any of the original components.(Smith,

Abbott, & Ryder, 2014) In this work, DES, based on triethylmethylammonium chloride (TEMA) and imidazole, was investigated as reaction media for cellulose pulp cationization and production of CCNFs and CWNFs. For the reaction, trimethylglycine (betaine) hydrochloride was used as a natural-based cationization reagent, while p-Toluenesulfonyl (tosyl) chloride was accomplished as a coupling agent for the formation of an ester bond between the cellulose and betaine hydrochloride. The effect of the different reaction conditions, such as the reaction temperature and amounts of cellulose, and the betaine hydrochloride and tosyl chloride in reaction systems, were investigated. Diffuse reflectance infrared Fourier transform spectroscopy (DRIFT) and elemental analysis were used for the chemical analysis of cationized materials. The production of both CCNFs and CWNFs was then demonstrated by mechanical disintegration of cationized bleached cellulose pulp and GWP. The obtained nanofibers were characterized using transmission electron microscopy (TEM).

Materials and methods

Materials

Unbleached spruce GWP(Visanko et al., 2017) was obtained in never-dried form, whereas the softwood dissolving cellulose pulp(Sirvio, Hyvakko, Liimatainen, Niinimaki, & Hormi, 2011) was delivered as dry sheets. The materials were first disintegrated in water, then filtered and washed with ethanol, and dried at 60 °C for 24 h.

Imidazole, TEMA, and betaine hydrochloride were purchased from TCI (Germany). Tosyl chloride and phosphotungstic acid were obtained from Sigma Aldrich (Germany) and ethanol from VWR (Finland). All the chemicals were used as obtained without purification.

Cationization of cellulose

DES was first prepared by mixing imidazole and TEMA in a beaker at a molar ratio of 2:1. The beaker was then placed in an oil bath at the desired temperature (50 – 100 °C) and chemicals were mixed until a clear solution was formed. Next, desired amounts of cellulose fibers were added (2 – 4 % according to the mass of DES), followed by the addition of betaine hydrochloride and tosyl chloride (0 – 4 times molar excess compared to cellulose). The reaction was allowed to proceed for four hours after which the beaker was removed from the oil bath and followed by the addition of ethanol. The product was then filtered and washed with a large amount of ethanol. The final product was dried at 60 °C for 24 h.

Elemental analysis of cationic cellulose

The nitrogen and sulfur contents of the samples were analyzed using the PerkinElmer CHNS/O 2400 Series II elemental and LECO CS-200 carbon-sulfur analyzers, respectively. The degree of substitution (DS) of the cationic group was calculated using Equation 1:

$$DS = \frac{N \times 162.15}{1401 - (N * 136.6)}$$

where N is nitrogen content, 162.15 the molecular weight of the anhydroglucose unit of cellulose, and 136.6 was the molecular weight of the betaine group.

Cationicity was calculated from the nitrogen content of the product (one mole of nitrogen is added by one mole of betaine group).

DS of the tosyl groups were calculated using Equation 2:

$$DS = \frac{S \times 162.15}{3200 - (S * 154.19)}$$

where S is sulfur content, 162.15 the molecular weight of the anhydroglucose unit of cellulose, and 154.19 was the molecular weight of the tosyl group.

Production of cationic cellulose and wood nanofibers

For the production of CCNFs and CWNFs, dissolving pulp and GWP were individually allowed to react with betaine hydrochloride in a similar manner described above (using two times molar excess of betaine hydrochloride and tosyl chloride at 70 °C for four hours). However, as a final step, the products were washed with water and stored in a non-dried state at 4 °C. The non-dried, cationized cellulose pulp or wood fibers were then diluted to a consistency of 0.5 wt% in deionized water, mixed for 30 seconds using an Ultra-Turrax (10,000 rpm). Then passed were two times at a pressure of 1000 bar through the 400 µm and 200 µm chambers and two times at 1500 bar through the 400 µm and 100 µm chambers of a microfluidizer (Microfluidics M-110EH-30, USA) to produce CCNFs and CWNFs.

Diffuse reflectance infrared Fourier transform spectroscopy

The chemical characterization of pristine and cationized cellulose and wood fibers was performed using DRIFT. The spectra were collected with a Bruker Vertex 80v spectrometer (USA) from freeze-dried samples. The spectra were obtained in the 600–4000 cm⁻¹ range and 40 scans were taken at a resolution of 2 cm⁻¹ from each sample.

Transmission electron microscopy

The morphological features of the fabricated CCNFs and CWNFs were analyzed with the Tecnai G2 Spirit TEM system (FEI Europe, Eindhoven, The Netherlands). Each sample was prepared by dilution with ultrapure water. A small droplet of the diluted CCNFs or CWNFs sample was placed on top of a carbon-coated and glow-discharge-treated copper grid. Then the excess of the sample was removed by touching the droplet with one corner of a filter paper. The samples were negatively stained with phosphotungstic acid (2% w/v at pH 7.3) by placing a droplet on top of each specimen. The excess phosphotungstic acid was removed as described above. The grids were dried at room

temperature and analyzed at 100 kV under standard conditions. The dimensions of the CCNFs and CWNFs were measured using the ImageJ measuring program (1.50i).

X-ray diffraction

The crystalline structure of the original and cationized dissolving pulp and GWP was investigated using wide-angle X-ray diffraction (WAXD). Measurements were conducted on a Rigaku SmartLab 9kW rotating anode diffractometer (Japan) using a Co K α radiation (40 kV, 135 mA) ($\lambda = 1.79030$ nm). Samples were prepared by pressing tablets of freeze-dried celluloses to a thickness of 1 mm. Scans were taken over a 2θ (Bragg angle) range from 5°–50° at a scanning speed of 10°/s, using a step of 0.5°. The degree of crystallinity in terms of the CrI was calculated from the peak intensity of the main crystalline plane (200) diffraction (I_{200}) at 26.2° and from the peak intensity at 22.0° associated with the amorphous fraction of cellulose (I_{am}), according to Eq. (1)(Segal, Creely, Martin, & Conrad, 1959):

$$CrI = \left(\frac{I_{200} - I_{am}}{I_{200}} \right) \cdot 100\%. \quad (1)$$

It should be noted that due to the Co K α radiation source, the cellulose peaks have different diffraction angles compared to results obtained using the Cu K α radiation source.

Viscosity

Viscosity of the CCNFs and CWNFs solution at concentration of 0.3% was measured by the TA Instruments Discovery HR-1 hybrid rheometer using cone-plate geometry (cone diameter of 40 mm and cone–plate angle of 1.999°). All measurements were conducted at 25 °C at a shear rate of 0.1–1000 1/s.

Results and discussions

Cationization of cellulose was performed using betaine hydrochloride as a cationic reagent, tosyl chloride as a coupling agent, and DES, based on TEMA and imidazole, as a solvent. Dissolving pulp containing less than 4% of hemicellulose and trace amounts of other components (e.g., lignin) was used as a pure cellulose source to optimize the cationization reaction. Initial reactions were conducted using three times excess of tosyl chloride and betaine hydrochloride compared to cellulose at 80 °C for four hours. The cationic group content of cellulose increased when the amount of cellulose (relative to the mass of DES) increased from 2 to 3% (Table 1, entry 1 and 2). Based on the elemental analysis of the nitrogen content of products, DS increased from 0.14 to 0.43. The increase of cellulose content likely improves the interaction between reagent and cellulose, which results in a better reaction efficiency. Also, the amount of the possible side reaction may decrease. However, a further increase in the cellulose content (4%) showed only a minor increase in the DS. With this solution, fibrous cellulose could still easily be mixed with DES, yet after the addition of reagents, cellulose began to swell significantly, and after around 15 min, mixing became difficult with a magnetic stirrer. Therefore, no attempts to use higher cellulose contents were conducted. However, it is assumed that higher concentrations could be utilized using a special apparatus, such as a high consistency mixer.

Table 1. Reaction conditions, elemental contents (nitrogen and sulfur), cationicity and DS (betaine, Bet and tosyl, Tos) groups) of the cationic dissolving pulp.

Entry	Reaction time	Temperature	Cellulose concentration (%)	Excess of chemical		N%	S%	Cationicity	DS(Bet)	DS(Tos)
				Tosyl chloride	Betaine hydrochloride					

								(mmol/g)		
1	4	80	2	3	3	1.11 ± 0.015	0.22 ± 0.020	0.79 ± 0.015	0.14 ± 0.003	0.01 ± 0.001
2	4	80	3	3	3	2.74 ± 0.126	0.48 ± 0.025	1.95 ± 0.126	0.43 ± 0.038	0.02 ± 0.001
3	4	80	4	3	3	2.78 ± 0.030	0.57 ± 0.029	1.95 ± 0.030	0.44 ± 0.009	0.03 ± 0.001
4	4	80	4	2	3	2.30 ± 0.084	0.48 ± 0.003	1.69 ± 0.061	0.34 ± 0.016	0.02 ± 0.000
5	4	80	4	1	3	0.87 ± 0.007	0.26 ± 0.011	0.62 ± 0.005	0.11 ± 0.001	0.01 ± 0.001
6	4	80	4	0.5	3	0.55 ± 0.021	0.23 ± 0.003	0.39 ± 0.151	0.07 ± 0.003	0.01 ± 0.000
7	4	80	4	0	3	*	*	-	-	-
8	4	80	4	2	0	*	*	-	-	-
9	4	80	4	2	0.5	0.54 ± 0.000	*	0.39 ± 0.005	0.07 ± 0.000	-
10	4	80	4	2	1	0.95 ± 0.042	*	0.68 ± 0.030	0.12 ± 0.006	-
11	4	80	4	2	2	1.82 ± 0.057	*	1.31 ± 0.040	0.26 ± 0.010	-
12	4	80	4	2	4	1.64 ± 0.219	0.23 ± 0.015	1.17 ± 0.157	0.23 ± 0.036	0.01 ± 0.001

13	4	50	4	2	2	0.92 ± 0.00 7	*	0.65 ± 0.005	0.12 ± 0.001	-
14	4	60	4	2	2	0.55 ± 0.04 2	*	0.39 ± 0.030	0.07 ± 0.005	-
15	4	70	4	2	2	1.29 ± 0.12 0	*	0.92 ± 0.085	0.16 ± 0.029	-
16	4	90	4	2	2	2.00 ± 0.12 7	*	1.43 ± 0.091	0.29 ± 0.023	-
17	4	100	4	2	2	1.66 ± 0.00 7	*	1.18 ± 0.005	0.23 ± 0.001	-

*Below detection limit (0.2%)

One of the possible side reactions occurring during the cationization is the tosylation of cellulose. As a part of the DES, basic imidazole may catalyze the formation of a tosyl cellulose (In literature, cellulose tosylation is conducted in alkaline conditions.(Schmidt, Liebert, & Heinze, 2014)). Based on the elemental analysis of sulfur, DS of the tosyl group ranged from 0.01 to 0.03 when three times molar excess of tosyl chloride was utilized (Table 1, entry 1-3). To limit this small but detectable presence of a tosyl group, further study was conducted using different amounts of tosyl chloride. In addition to limiting the occurrence of side reactions, the decrease of chemical consumption can improve the environmental and economic feasibility of chemical modification, especially on an industrial scale. When the molar excess of the tosyl group toward cellulose was decreased from 3 to 2, only a modest effect on the cationic group content was observed (DS decreased from 0.34 to 0.44) (Table 1, entry 3 and 4). However, a further decrease of tosyl chloride content led to a significant decrease in the content of the betaine ester group. At the same time, a

small decrease in the DS of tosyl groups was noticed. No reaction took place without tosyl chloride, indicating that reaction is not just simple esterification of cellulose with betaine hydrochloride (Table 1, entry 7).

The amount of betaine hydrochloride also had a significant effect on the reaction efficiency. DS gradually increased when the amount of betaine hydrochloride was increased (from 0.5 to two times excess compared to cellulose) (Table 1, entry 9-11). However, when four times excess of betaine hydrochloride was used, DS sharply decreased. Therefore, the maximum DS could be obtained using three times excess of betaine. However, to minimize the chemical consumption, two times excess was used for further studies. In addition, no nitrogen was detected when the reaction was performed without betaine hydrochloride (Table 1, entry 8). This indicates that the nitrogen is not due to the attachment of imidazole or TEMA-moieties on cellulose. Additionally, when using less than three times excess of betaine hydrochloride, the number of tosyl groups were below the detection limit of sulfur analysis.

The increase in temperature enhanced the reaction efficiency, and DS could be increased from 0.12 to 0.29 by changing the temperature from 50 to 90 °C (Table 1 entry 13-16). However, a slight decrease in DS was observed when the temperature was further changed to 100 °C (Table 1 entry 17). The decrease in the DS might indicate the occurrence of some side reaction hindering the reaction efficiency.

A possible reaction mechanism involves *in situ* formation of mixed anhydride *via* the reaction between a carboxylic acid group of betaine hydrochloride and tosyl chloride (Scheme 1). (Jasmani, Eyley, Wallbridge, & Thielemans, 2013) At first, basic imidazole (**2**) works as a catalyst by deprotonating the carboxylic acid group of betaine (**1**). Deprotonated betaine then reacts with tosyl chloride (**3**) to form mixed anhydride (**4**). Imidazole works as an acid scavenger by neutralizing

hydrogen chloride formed as by-product at this stage. In the next step, oxygen atom of hydroxyl group of lignocellulose (5) reacts mixed anhydride at carbonyl carbon to form intermediate (6). The intermediate is then deprotonated by the imidazole, resulting in the formation of cellulose betaine ester (7) as a product and p-toluenesulfonic (tosylic) acid as by-product. Tosylic acid is then neutralized of imidazole. As well, other reaction mechanism, such as the formation of the highly reactive intermediate acyl imidazole derivate of betaine can take place.(Wakasugi, Iida, Misaki, Nishii, & Tanabe, 2003)

Scheme 1. Possible reaction mechanism of the cationization of cellulose via imidazolium catalysed tosylation of betaine hydrochloride (imidazolium chloride ([IMI][Cl]) and tosylate ([IMI][Tos]) are formed as by-product).

The production of CCNFs from cationized cellulose was investigated with a specimen from the reaction of two times molar excess of tosyl chloride and betaine hydrochloride at 70 °C for four

hours. Furthermore, GWP was modified using the same chemical to fiber mass ratio at similar reaction conditions. Cationic group content of GWP was 0.70 mmol/g, which is slightly lower compared to cationized dissolving pulp (0.92 mmol/g). On the other hand, yield of cationic GWP was notable higher compared to cationized dissolving pulp (83% vs. 65%). Before the fibrillation, both samples were chemically characterized using DRIFT.

The chemical modification of fibers during the cationization is apparent in the DRIFT spectra (Figure 1). The appearance of a new peak at 1756 cm^{-1} (the C=O stretching) confirms the formation of ester bond to the cellulose backbone (Ma et al., 2014). In Figure 1, ester peak can be easily observed in spectrum b, whereas no peak is detectable at spectrum a. The wavenumber of ester differs from the C=O stretching of the carboxylic acid group of betaine (appears at 1735 cm^{-1}) indicating that betaine is attached to cellulose via ester bond and not by physical absorption.. In the case of GWP, the original carbonyl peak at 1739 cm^{-1} , originating from C=O stretching of the ester from the acetyl group of hemicellulose and ester linkage of the carboxylic groups in the ferulic and p-Coumaric acids of lignin/hemicellulose, (Jonoobi, Niska, Harun, & Misra, 2009) was shifted to a higher wavenumber of 1749 cm^{-1} during the cationization. In addition, the intensity of carbonyl peak increased, indicating the successful formation of lignocellulose betaine ester.

Figure 1. DRIFT spectra of dissolving pulp and GWP before (a and b) and after (c and d) of cationization (dashed line indicates the betaine ester bond formed during the chemical modification in DES).

Both cationized cellulose and wood fibers disintegrated into long nanofibers with a diameter below 10 nm (Figure 2, more TEM images with different magnification can be found in Supporting Information, Figure S1 and S2) when passed through a microfluidizer. The average diameter for CCNFs and CWNFs were 4.7 ± 2.0 and 3.6 ± 1.3 nm, respectively, indicating that they mostly consisted of individual elemental fibrils (diameter of elemental fibrils is reported to be 3.5 nm). However, along with individualized elemental fibrils, some large aggregates were noted in both samples. In case of CCNFs, the diameter of nanofiber aggregates ranged between 20 to 70 nm, whereas aggregates with diameters close to 200 nm were observed with CWNFs. In addition to the larger nanofiber aggregates, coarse worm-like nanoparticles were observed in the TEM images of CWNF (indicated with the red arrow in Figure 2). These particles had a diameter around 20 nm. As these particles were absent in TEM images of CCNFs, they may be formed due to the presence of lignin (It can be noted that although hemicellulose might also have some contribution in the

255 formation of these particles, they are hardly reported in cases of bleached pulp containing
256 hemicelluloses.). These nanoparticles differed from the nanofiber aggregates with similar diameter
257 as no internal fine structure could be observed (In the larger nanofiber aggregates, individual
258 nanofibrils could be observed.). However, more observations are requested to determinate the
259 composition and formation mechanism of these particles.

Figure 2. TEM images of the a) CCNFs and b) CWNFs. Possible lining based nanoparticles are indicated by the red arrow.

Previously, the introduction of the cationic group content of 0.79 mmol/g on cellulose resulted in the formation of thin, homogenous CCNFs, whereas some aggregates were observed with a lower charge density.(Pei et al., 2013) On the other hand, charge density as low as 0.354 mmol/g has been reported to produce nanofibers as thin as 0.8 – 1.2 nm with the average diameter of 4 nm.(Olszewska et al., 2011) Although a direct comparison between different methods is not entirely meaningful due to the wide variety of conditions, chemicals, and starting materials used, it can be concluded that the method introduced here is suitable to produce similar albeit slightly

more heterogeneous CCNFs compared to many previous studies. Yet, there is a scarcity of the information on the production of CWNFs. However, compared to the anionic WNFs(Sirviö & Visanko, 2017), CWNFs appear as more homogenous in size, and previously observed large aggregates with diameters close to the micrometre were absent in this study. The CWNFs produced here were more in line with CNFs obtained from the acid-hydrolyzed unbleached hardwood chemical pulp, with an initial lignin content of 17.2%, where average diameters of 9.6 and 7.1 nm were observed.(Bian, Chen, Dai, & Zhu, 2017)

Based on XRD measurement, no rearrangement of the cellulose crystalline structure took place during the chemical modification or nanofibrillation (Figure 2). Both, cationized cellulose and wood fibers exhibited characteristic main peaks of cellulose I close to angles of 18.3° and 26.1° related to the 1–10, 110 (appears as superimposed peak at 18.3°) and 200 crystalline planes, respectively.(French, 2014) The weak peak of 003 crystalline plane can be seen close to angle of 40° (sharp peaks in diffractogram of CCNF close to 10° and 35° originates from the sample holder (Liimatainen et al., 2014)). Previous, the use of same DES showed similar results, and it can be concluded that no, or only minimal dissolution and regeneration of cellulose occurred at used reaction conditions. The CrI of the cationized cellulose and wood fiber were 63% and 46%, respectively. As the initial CrI of dissolving pulp and GWP were 65% and 52%, respectively, it indicates that cationization has only minor effect on the amount of the crystalline domains in the original pulps. Previously, succinylation of fibers in same DES led to the more notable alteration crystallinity (e.g. 27% decrease of the CrI in case of GWP).(Sirviö & Visanko, 2017)

Figure 3. XRD diffraction patterns of original fibers and cationized fibers after nanofibrillation

Both cationic nanofiber solutions (0.3%) exhibited typical shear thinning behavior of cellulosic nanofibers solutions.(Lasseguette, Roux, & Nishiyama, 2008) At low shear rate, CCNF solution showed viscosity similar to those of high viscose cellulose solutions reported in literature.(Moberg et al., 2017; Naderi, Lindström, & Sundström, 2014) On the other hand, significantly lower viscosity was observed for CWNF solution. For example, viscosity of CCNF solution was 1.34 Pa.s at shear rate of 1 s^{-1} whereas at same shear rate, the viscosity of CWNF was 0.12 Pa.s. Previously, similar trend was observed with WNF (lignin content of 27.4%) produced by heat-intensified disc nanogrinding process as around five-times higher consistence was used for WNF compared to reference CNF obtained from bleached cellulose fibers to obtain comparable viscosity values.(Visanko et al., 2017) Although it is generally accepted that higher viscosities of CNF solution are caused by longer and thinner fibers(Lasseguette et al., 2008), when comparing viscosities of lignin-poor and lignin-rich, other properties, such as the stiffness of the WNF, a lower water adsorption, and a reduced polarity caused by presence of lignin has to be taken in

account. It can be concluded that use of bleached cellulose fiber for production cationic nanofibers are desired for application, such food and cosmetic thickeners, where high viscosity is requested.

Figure 4. Viscosities of the CCNF and CWNF solutions at concentration of 0.3% as a function of shear rate (0.1–1000 s⁻¹).

Although a general approach based on DES has been introduced here as an efficient reaction media for esterification of cellulose with a cationic group containing carboxylic acid (betaine), several points need further investigation to prove in full the feasibility of the current method. Both DES components have not been reported to possess severe toxicity, although imidazole is a basic and corrosive chemical. However, it has been reported that formation of DES between two components can change their toxicity profile.(Juneidi, Hayyan, & Ali, 2016) Some DES systems have been reported to exhibit lower toxicity compared to their individual starting materials(Wen, Chen, Tang, Wang, & Yang, 2015), whereas in some cases, toxicity was increased when two components were combined(de Moraes, Gonçalves, Coutinho, & Ventura, 2015). In addition, recycling of DES is highly important when considering economic and environmental feasibility. The accumulation of chloride and tosyl salts of imidazolium in DES is one of the main obstacles

during recycling, as they cannot be evaporated out of the solution. Therefore, separation methods, such as selective precipitation of ionic species or the use of ion-selective membranes should be investigated in future studies.

Conclusions

Cationization of cellulose was accomplished using betaine as a reagent, tosyl chloride as a coupling agent, and DES, based on TEMA and imidazole, as a reaction medium. By adjusting the reaction conditions, such as the amount of cellulose and reagent and temperature, the DS could be varied from 0.07 to 0.44. The feasibility of the current cellulose cationization method in the material application was demonstrated by producing CCNFs. In addition to nanofibers produced from bleached wood pulp, lignin-containing wood fibers (non-bleached) were also cationized, and CWNFs were produced thereafter. CWNFs exhibited similar dimensions compared to CCNFs (average diameter being around 5 nm), whereas some larger nanofiber aggregates were more visible in CWNFs. In addition to the typical cellulose-based nanofibers, some coarse, worm-like nanoparticles were observed in CWNFs. These particles were assumed to originate from lignin. In addition to the cationization of cellulose, this method can be assumed as suitable for the esterification of lignocellulosic materials with other components, such as fatty acids.

ACKNOWLEDGMENT

Elisa Wirkkala and Dr. Mika Kaakinen are acknowledged for elemental analysis and help with TEM. Assoc. Prof. Henrikki Liimatainen is gratefully acknowledged for his help with the manuscript writing and fruitful discussions.

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