

ULTRASOUND-CATALYZED TEMPO-MEDIATED OXIDATION OF NATIVE CELLULOSE FOR THE PRODUCTION OF NANOCELLULOSE: EFFECT OF PROCESS VARIABLES

Shree P. Mishra,* Jennifer Thirree, Anne-S. Manent, Bruno Chabot, and Claude Daneault

In this study application of ultrasound in oxidizing native cellulose for the production of nanocellulose has been explored for the first time. Bleached hardwood kraft pulp was oxidized with an ultrasound (US) catalyzed 2,2,6,6-tetramethylpiperidin-1-oxyl (TEMPO) system (US-TEMPO-system) at five different temperatures – 5, 15, 25, 35, and 45°C and two pH ranges, 8.5-9.0 and 10.0-10.5 – to obtain the optimum reaction conditions. The reaction pH and temperature have significant effect on the kinetics of the formation of carboxylate in the oxidized pulps and produce depolymerization at temperatures greater than 25°C. Formation of carboxylate on the cellulose chain is directly proportional to the NaBr concentration. The pulp oxidized by the US-TEMPO-system at 25°C had 10-15% more carboxyls and showed a ca. 10% increase in the nanocellulose yield when compared to the TEMPO-system without sono-catalysis.

Keywords: Nanocellulose; TEMPO; Native cellulose; Oxidation; DP_v; Carboxyl groups; Yield

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INTRODUCTION

Native cellulose is mainly present in lignocellulosic biomass (wood and agri-residues). Besides being paracrystalline in morphology (Atalla 1987), native cellulose obtained from wood always contains varying amounts of other amorphous constituents, including hemicelluloses, and lignins, depending upon the refinement process. The hemicelluloses found in wood are mainly xyloglucan, arabinoxylans, and mannans, and the primary precursors for lignins are coniferyl alcohol, synapyl alcohol, and p-coumaryl alcohol. Principal steps for the isolation of cellulose from woody raw materials are digestion or cooking, and further purification or bleaching, involving complex chemical reactions using a range of chemicals. The bleached pulp so obtained forms the basis for the vast paper, board, textile, and other related industries world wide.

Undoubtedly, the paper industry is the biggest consumer of bleached, delignified pulp and will continue to be so in the future. Recent stagnant or sluggish performance of the pulp and paper industries (Brown 2010) throughout the world has speeded up the campaign to use the lignocellulosic biomass for producing value-added products. Besides the biorefinery concept, which is one of the most sought-after projects (Cherubini 2010; Kamm and Kamm 2007; Luo et al. 2010), production of cellulose nanofibers from native cellulose has also been the subject of intensive investigation (Habibi and Dufresne 2008;

Henriksson and Berglund 2007; Iwamoto et al. 2005, 2007; Janardhnan and Sain 2006; Nakagaito and Yano 2004; Paakko et al. 2007; Saito et al. 2005, 2006a,b; Taniguchi and Okamura 1998; Wang and Sain 2007). Fundamentally, lignocellulosic biomass is made up of nanometer-scale cellulose building blocks that provide valuable properties (mechanical, optical, and other) to wood and other types of renewable lignocellulosic and cellulosic biomaterials. Similarly, the nanometer dimensions of the cellulose, lignin, and other components provide the origin for the unique properties of wood, in addition to a host of wood-based products including paper, paperboard, oriented strandboard, glulam beams, etc., (Klemm et al. 2005). These nanosized cellulose building blocks, which are networked, and irreversibly fixed in the supramolecular cellulose structure, and determines the product properties and functionality, have been described as nanocellulose (Klemm et al. 2005). While the relative mass of the nanofibrils and nanocrystalline cellulose are small, their surface area is large (Wegner and Jones 2009).

Cellulose nanofibers can be prepared by various ways and often bear different names such as “whiskers” (de Rodriguez et al. 2006; Oksman et al. 2006; Rånby 1952; Samir et al. 2005), “microfibrillar cellulose,” (MFC) (Stenstad et al. 2008), and “nanofibrillar cellulose,” (NFC) (Paakko et al. 2008). “Whiskers,” which are also called “nanorods,” “rod-like cellulose microcrystals,” or “nanowires,” are very long and straight crystals of cellulose, whereas MFCs and NFCs remain attached together for at least a portion of their length (Hubbe et al. 2008).

During the last few years efforts have been made to prepare cellulose nanofibers or nanocellulose from native cellulose by TEMPO-mediated oxidation using a TEMPO-NaBr-NaOCl system followed by mechanical treatment or with the help of ultrasonic waves (Johnson et al. 1999; Saito and Isogai 2005; Saito et al. 2007). The term TEMPO stands for 2,2,6,6-tetramethylpiperidin-1-oxyl. Oxidation of the primary alcohol groups (C6 hydroxyls in the cellulose chain) in the presence of secondary and tertiary hydroxyls to carboxylate is the main objective of TEMPO-NaBr-NaOCl treatments (de Nooy et al. 1995). Such regioselective conversion of the primary hydroxyl groups to carboxylate will make it possible to loosen the adhesion between cellulose fibrils by preventing the formation of strong interfibril hydrogen bonds (Saito et al. 2007). A general reaction scheme of the oxidation of C6 hydroxyl to the carboxyl group by TEMPO-system is shown in Fig. 1. The detailed reaction mechanism has been described in Li (2006).

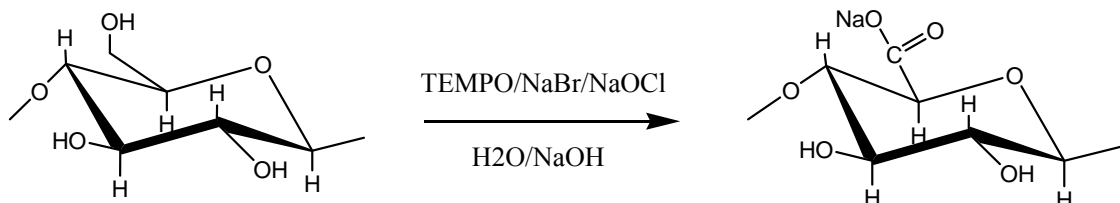
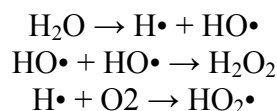


Fig. 1. A reaction scheme of the oxidation of C6 hydroxyl group of cellulose by TEMPO-system

The dimensions (width and length) of the cellulose nanofibers or nanocelluloses obtained by TEMPO mediated oxidation followed by mechanical treatment have been measured with transmission electron microscope (TEM). TEM is the most commonly used technique for the determination of the dimensions of the cellulose nanofibers. The widths of the nanocelluloses were in the range of 3 to 5 nm, and their lengths varied from

nanometers to a few microns (Saito et al. 2007). However, the geometrical dimensions of the cellulose nanocrystals (obtained by acid hydrolysis) vary widely depending on the source of the cellulosic material, the hydrolysis conditions, and on the measurement techniques used (Habibi et al. 2010).

The use of ultrasound for catalyzing chemical reactions has already been documented (Kardos and Luche 2001; Suslick 1995; Suslick and Price 1999). Several carbohydrate compounds have also been subjected to ultrasound-catalyzed oxidation (Kardos and Luche 2001; Zhang et al. 2008). Some studies have shown the effect of ultrasound on pulp preparation (fibrillation) (Iwasaki et al. 1962), and deinking of printed papers (Manning 2004). However, no chemical changes on the cellulose chain due to ultrasound have been reported so far (Thompson and Manning 2005). In liquid phase reaction medium, high intensity ultrasound waves function through the mechanism of acoustic cavitation (the formation, growth, and collapse of bubbles) (Leighton 1994) and consequently lead to a sonochemical effect. During cavitation, the bubbles collapsing produce intense local heating and high pressure for a very short lifespan. These transient, localized hot spots drive high energy chemical reactions, yielding temperatures of $\sim 5000^{\circ}\text{C}$, pressures of about 1000 atm, and heating and cooling rates above 10^{10} K/s (Suslick 1990; Suslick and Price 1999). An extensively studied reaction is the sonolysis of water, producing hydroxyl radicals and hydrogen atoms (Henglein and Kormann 1985).



The aim of this study was to use high-intensity ultrasound in combination with a TEMPO system to oxidize the primary hydroxyl groups on the cellulose polymer chains to carboxylate groups. We used high intensity ultrasonic waves at 68 kHz and 170 kHz frequencies along with the TEMPO system for the oxidation of once-dried bleached hardwood kraft pulp at various temperatures, and at different amounts of sodium bromide. We attempted to explain the correlation between the NaOH consumption, and the final pH to assess the extent of oxidation of the cellulose fibers. Cellulose nanofibers obtained from selected samples of different carboxylate contents using mechanical treatment were quantified gravimetrically.

EXPERIMENTAL

Materials

A mill-bleached, machine-dried hardwood kraft pulp was used as a source of native cellulose for the oxidation purpose. 4-acetamido-TEMPO (2,2,6,6-tetramethylpiperidin-1-oxyl), sodium bromide (NaBr), and sodium chlorite (NaClO_2) were obtained from Sigma-Aldrich and used as received. Sodium hypochlorite (NaOCl) was procured locally, and assayed for its molar concentration before use. The ultrasonic cleaning bath and ultrasonic generators were procured from Ultrasonic Power Corporation (USA). Two different ultrasonic generators were used to produce 68 kHz

and 170 kHz frequency ultrasonic waves. These were operated at 500, 750, and 1000 W ultrasonic power intensity. The cleaning bath was set at both frequencies.

Methods

Pulp oxidation by US (ultrasound)-TEMPO-NaBr-NaOCl-system

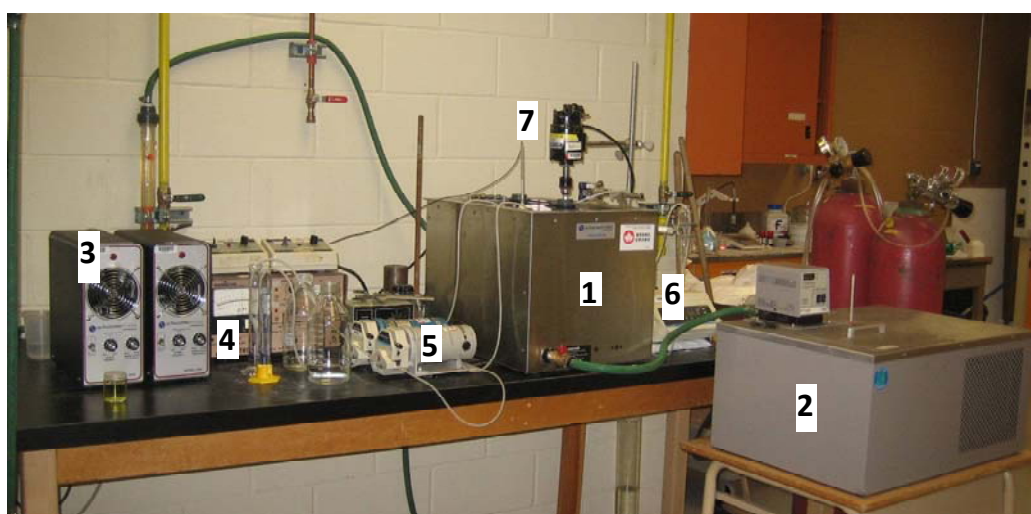
Oxidation of bleached hardwood kraft pulp was carried out batchwise using 20 g (wt. %) sample per batch. The pulp was pre-soaked overnight in de-ionized water, and disintegrated for 10 minutes in a laboratory disintegrator to obtain a uniform fiber suspension of about 1% consistency. The oxidation was conducted in a specially designed glass reactor that was placed in an ultrasonic cleaning bath to ensure that it was exposed to a uniform ultrasound effect. The frequencies of the ultrasonic waves used were 68 kHz, and 170 kHz at 1000 W ultrasonic power intensity unless otherwise mentioned. 4-acetamido TEMPO (0.46 g, 0.11 mmol per g cellulose fiber), and NaBr (1.27 g, 0.617 mmol per g cellulose fiber) were dissolved in 50 mL of de-ionized water before being added to the fiber suspension. The pH of the fiber slurry was adjusted to 10.0-10.2 with 0.5 M NaOH or 0.5 M HCl using a pH-stat as required. The TEMPO-mediated oxidation was started by adding the desired amount of the NaOCl solution (3.75 mmol NaOCl per g of cellulose fiber), at 25 °C. Deviations related to chemical charges are specified in the respective sections. The pH was maintained at 10.0 to 10.2 by adding 0.5 M NaOH using a pH-stat during the course of the reaction, and the amount of NaOH consumed was noted at 15-minute intervals. The reaction was stopped after 90 minutes by adding 50 mL of ethanol, and the final pH of the solution was adjusted to 7.0 by adding 0.5 M NaOH or 0.5 M HCl as required. The total volume of the TEMPO-oxidized fiber suspension was measured, and a known volume of which was filtered and washed with de-ionized water on a pre-weighed filter paper to quantify the yield after oxidation. The rest of the TEMPO-oxidized cellulose slurry was filtered, thoroughly washed with de-ionized water, and stored at 4 °C (to prevent from any kind of microbial attack) for further treatment, and analysis. Similar TEMPO-mediated oxidations were performed in the absence of ultrasonic waves. A photograph of the experimental setup for the US-TEMPO-system is shown in Fig. 2.

Measurement of carboxyl group content

The carboxylate content of the TEMPO-oxidized cellulose was determined using a conductometric titration method using a Dosimat 765 (Metrohm) titrator according to the technique of Katz et al. (1984). The sodium carboxylate groups in the TEMPO-oxidized celluloses were converted to the free carboxyl form by treating the sample with 0.1 M HCl solution three times and finally, thoroughly washed with de-ionized water to remove the excess acid. The oxidized pulp prepared in this way was transferred to a 600-mL beaker containing 450 mL of 0.001 N NaCl solution, and mixed well. Five mL of 0.1 N HCl was added to the fiber suspension before starting the titration of carboxylate groups with 0.1 N NaOH solution. The titration was controlled by use of automatic titration software developed in our institute. At the end of the titration the fibers were filtered, washed, and dried in an oven at 105 °C to determine the exact weight of the sample. The carboxyl content expressed in mmol/kg was calculated by the software.

Post-oxidation of the oxidized pulp

A post oxidation of the oxidized pulp with NaClO₂ in acetic acid / sodium hydroxide buffer (pH 5.0) at 70 °C was done to convert the aldehyde groups produced during the TEMPO-mediated oxidation of cellulose to carboxyl groups. In this process 2 g of the oxidized pulp (WT. %) was suspended in 160 mL of deionised water in a tightly closed 250-mL glass bottle (from Duran). This was followed by the addition and mixing, sequentially, of 20 mL of 34 g/L NaClO₂ solution and 20 mL of acetic acid/ sodium hydroxide buffer. The mixture was kept in a water bath at 70 °C for 2 h, after which it was cooled, filtered and washed thoroughly with water prior to further analysis. This method gave results similar to those yielded by the technique of Saito and Isogai (2004), and hence it was used in this work.



1. Ultrasonic bath
2. Thermostated bath for cooling
3. Ultrasonic generators
4. pH controller
5. Peristaltic pumps for NaOH and HCl
6. Peristaltic pump for NaOCl
7. Motor for the agitator
8. Reactor for the oxidation

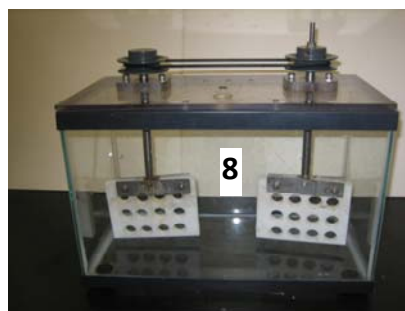


Fig. 2. Photograph of the experimental setup for the oxidation with TEMPO-, and US-TEMPO-systems

Measurement of viscosity

The relative viscosity of the TEMPO- and US-TEMPO-oxidized celluloses, as well as reference samples was measured as per SCAN standard methods SCAN-CM 15:88. The degree of polymerization (DP_v) was calculated as per the Sihtola method, using the following Mark-Houwink-Sakurada equation: $DP_v^{0.905} = 0.75 [\eta]$ (Sihtola and Kyrklund 1963). The DP_v calculation is based on the assumption that both cellulose and oxidized cellulose molecules have the same viscosity at the same DP.

NMR Measurements

Solid-state CP/MAS ^{13}C spectra were recorded on a Bruker Advance Spectrometer (Bruker Biospin Inc, Milton, ON, Canada) at 75 or 100 MHz (Magnetic fields of 7.0 or 9.4 Tesla). Samples were spun at 6 to 8 kHz in a 4mm ZrO_2 rotor at the magic angle, and at room temperature. A contact time of 1 ms, and a relaxation time of 1 s were accorded to an acquisition of 3000 transients to produce a good quality spectrum. A high power phase altered proton decoupling time-proportional-phase-increment (TPPI) was applied during free induction decay (FID) acquisition. ^{13}C chemical shifts were determined with reference to adamantane.

Production of nanocellulose

The oxidized cellulose sample (0.3 g) was suspended in water (300 mL) at 0.1 % concentration. The slurry was mechanically homogenized by means of a blender for a total time of 20 min (Saito et al. 2007). A pause of 15 s followed after each 45 s of mixing to prevent overheating of the blender. The disintegrated suspension was centrifuged at 10,000 g during 10 min to separate the supernatant containing nanocellulose from the unfibrillated and partially fibrillated fractions. The quantity of nanocelluloses was obtained by drying three samples of 40 mL each from the supernatant at 105°C. The slurry was stored at 4°C (to prevent from any kind of microbial attack) for further characterization.

TEM observation

The samples were examined using a Phillips EM 208S transmission electron microscope (TEM) operating at 80 kV. Images were acquired electronically with an installed camera. Diluted nanofibril suspensions were deposited on the formvar-coated copper grids (400 mesh). The excess liquid was absorbed with filter paper, and the adsorbed nanofibers were stained with 2% uranyl acetate negative stain before observation with TEM.

RESULTS AND DISCUSSION

This study was carried out with mill-produced bleached hardwood kraft pulp as a source of native cellulose. First of all, the pulp was subjected to an ultrasound (US) treatment under similar conditions as to the regular experiments using the US (ultrasound)-TEMPO system to determine its effect on the cellulose fibers, especially on the carboxyl groups. The carboxyl group content, yield, and viscosity obtained with US alone at 68 kHz and 170 kHz without the TEMPO-system are shown in Table 1. No change in the carboxyl group content was found when the oxidation was carried out with US alone without using TEMPO-system compared to the reference pulp, which confirms the previous findings on the subject (Thompson and Manning 2005). The degree of polymerization (DP_v), and yield of the US treated pulps were not much affected. Better yield, and DP_v were obtained at 170 kHz as compared to 68 kHz (Table 1). The carboxyl group content after post-oxidation was comparable to that of the reference pulp.

However, the use of ultrasound along with the TEMPO-system for the oxidation of pulp produced different results, which are presented in the following paragraphs.

Table 1. Effect of Ultrasonic Treatment on the Carboxyl Group Content, Yield, and DPv of the Pulp

Parameter	COO- (mmol/kg)	COO- Post-Oxidation (mmol/kg)	Yield (%)	DPv
Bleached pulp	61.1 (60.7, 61.4)	70.8 (72.7, 68.9)	100	997
PB - US 68 kHz (25°C)	60.4 (62.7, 58.1)	71.8 (77.8, 65.7)	96.4	936
PB - US 170 kHz (25°C)	61.2 (60.1, 62.2)	66.3 (66.6, 66.0)	98.8	981

Effect of Temperature (US-TEMPO-system)

It is well known that in aqueous medium ultrasound functions according to the principle of acoustic cavitation, which generates a lot of heat that could reach up to several degrees centigrade (Suslick and Price 1999). One may think that the extra heat generated during the US treatment may increase the rate of the reaction if the temperature of the reaction is not maintained. Our experiments were done at 5, 15, 25, 35 and 45°C, while keeping the other conditions constant, to find out the effect of the temperature on the reaction kinetics and advantages of using ultrasound in TEMPO- oxidation system. It should be noted that a lot of ice was needed to maintain the temperature during the US run. In fact, we were not able to conduct the US treatment at 5°C and, therefore, the oxidation of the pulp at 5°C was carried out in the absence of US treatment. The carboxyl group content, yield, and DPv of the oxidized pulp are plotted as a function of temperature, as shown in Figs. 3 through 5.

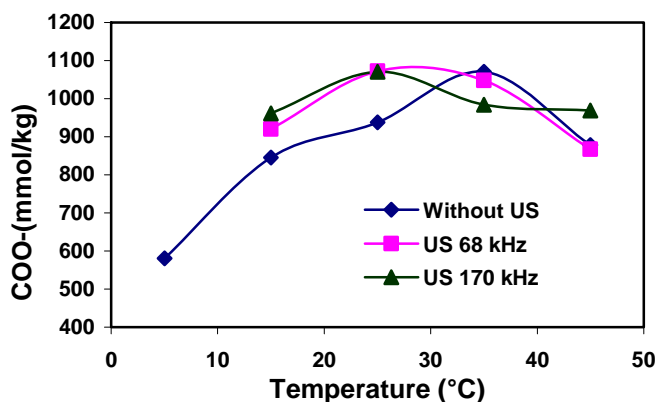


Fig. 3. Effect of temperature on the carboxyl group content in the US-TEMPO-system oxidized pulp

The carboxyl groups content increased with the increase in temperature up to 25°C, and it started to decrease at 35°C onwards in the case of US treatments, whereas without US it increased up to 35°C and then decreased. Figure 3 shows that a gain of about 15% was obtained with the presence of US treatment, i.e. 1071 mmol/kg versus 938 mmol/kg for TEMPO alone, at 25°C. The decrease in carboxyl group content at temperatures > 25°C in US-TEMPO oxidation and >35°C in TEMPO alone could be

related to the de-polymerization and simultaneous dissolution of the carboxylate-containing sugar moieties during the oxidation reaction, leading to a loss in yield. It could be seen from Fig. 4 that the yield after oxidation was less than 85% in all the samples at 45°C.

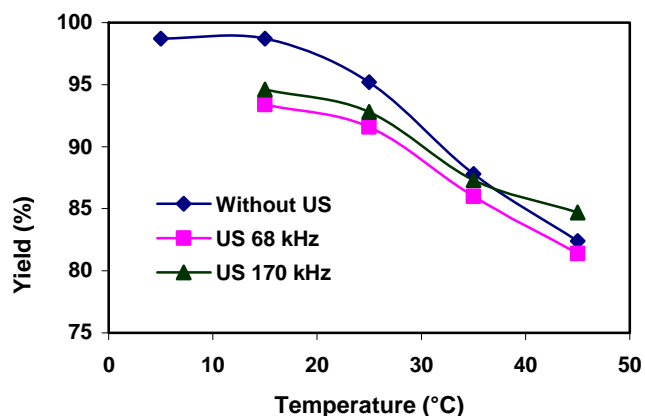


Fig. 4. Effect of temperature on the yield of the US-TEMPO-system oxidized pulp

Figure 4 shows that the yield decreased with increasing temperature in all the cases. Again, one reason could be the depolymerization of the cellulose and dissolution of the oxidized moieties with increase in the temperature, as mentioned above. Another possible reason could be the dissolution of the amorphous, non-cellulosic pulp constituents (hemicelluloses, pectins, and lignins) during the course of the US-TEMPO-mediated oxidation, in addition to the possibility that dissolution was further aggravated at higher temperatures. Isogai and Kato (1998) have also shown that the charge of TEMPO, reaction time, and temperature were the key factors controlling the depolymerization of cellulose in the case of TEMPO-mediated oxidation of regenerated or mercerized cellulose. At a given temperature the yield loss was more pronounced in the case of ultrasonic treatments as compared to the cases where no US treatment was involved. Note that the US treatment at 68 kHz gave lower yield than that at 170 kHz. One possible explanation for the greater loss in yield with US-TEMPO-system could be that the collapse of cavitation bubbles, the principal mechanism involved in the sonochemistry, in the reaction medium with the presence of oxidants facilitated the access of the reactive species to the reaction site with a greater ease increasing the reaction rate and yielded the related effects. However, simultaneous detachment and dissolution of the oxidized nanofibrils due to turbulent mixing and jets-streaming, a resultant effect of acoustic cavitation, might have led to the yield loss. The fact that cavitation effects are significant at lower US frequencies may be one reason for the observed greater yield loss at 68 kHz. From another viewpoint the observed effects of the ultrasound with the TEMPO-system could, possibly, be explained as follows: TEMPO-mediated oxidation of solid-state cellulose involves a great deal of difficulty in the separation of cellulose nanofibrils that strongly interact with each other via multiple hydrogen bonding. Thus, the ultrasonication is expected to have some contribution to loosening the cellulose nanofibrils, and to peeling the slightly carboxylated cellulose

chains/nanofibers from the solid cellulose sample. Considering the high carboxyl group content of 1071 mmol/kg and good yield of 93% obtained at 25°C, this temperature was found satisfactory in comparison to other temperatures studied.

The DP_v of the pulps was considerably affected during the US-TEMPO and TEMPO mediated oxidation. The observed DP_v of the oxidized pulps was about four times less compared to the DP_v of the original pulp. The DP_v of the oxidized pulps were in the range of 200-250 (Fig. 5), whereas the DP_v of the original pulps without TEMPO oxidation was in the range of 900 to 1000 (Table 1). The DP_v of the US-TEMPO and TEMPO oxidized pulps decreased with increase in the temperature (Fig. 5); however, at a given temperature there was no significant difference in the DP_v of the pulps oxidized with US-TEMPO and TEMPO system. The depolymerization of polysaccharides having glucuronic acid may occur to some extent by β -elimination during the TEMPO-mediated oxidation under alkaline conditions, e.g. pH 10.0 here (Kitaoka et al. 1999). However, besides the above mentioned reason, the depolymerization of the cellulose has also been attributed to the sodium hypochlorite oxidation of polysaccharides, leading to 2,3-scission of glucose units resulting in the formation of aldehyde and dicarboxylic structures (Besemer 1993).

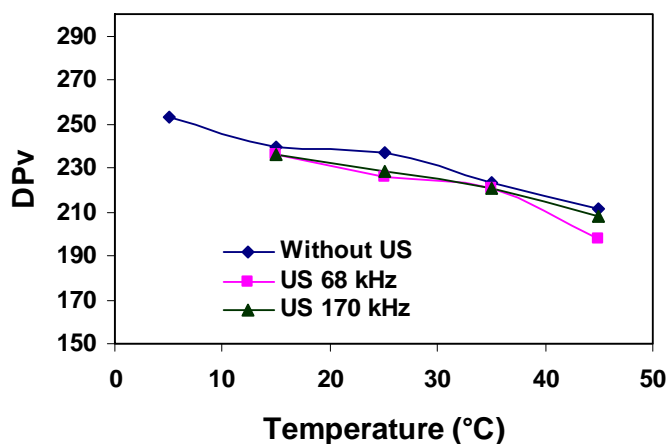


Fig. 5. Effect of temperature on the DP_v of US-TEMPO-system oxidized pulp

Aldehyde content at different temperatures

The oxidation of the C6 hydroxyls to carboxyl group proceeds through an intermediate aldehyde group (Bragd et al. 2004; de Nooy et al. 1995). The oxidation is seldom complete, and some amount of the aldehyde remains in the pulp, which could have a considerable influence on the behavior of the oxidized pulp (Saito and Isogai 2004). Aldehyde groups in the oxidized pulps were determined following an indirect method by which the aldehyde groups were converted to carboxylate by post-oxidation with NaClO₂ in acetate buffer at pH 5. The carboxyl group content in the post-treated pulp was re-determined. The difference in carboxyl contents before and after post oxidation was taken as a measure of the aldehyde groups present in the oxidized pulp.

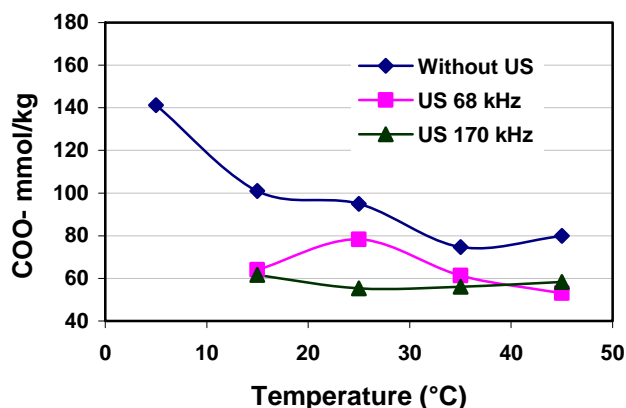


Fig. 6. Carboxyl group content (post oxidation - pre oxidation) as a measure of aldehyde content as a function of temperature

The aldehyde contents of the pulps oxidized with US-TEMPO-system at different temperatures are presented in Fig. 6. The aldehyde contents ranged from 50 to 100 mmol/kg of pulp. It could be observed that with increase in the reaction temperature, oxidation with TEMPO system proceeded towards completion, and lesser amounts of the aldehyde groups were formed. An exception was noted in the case where the pulp was oxidized at 5°C and yielded a high aldehyde content of 140 mmol/kg. Incomplete conversion of the aldehydes formed during the TEMPO-mediated oxidation reaction to carboxyls because of the lower temperature and shorter reaction times could be the possible reason for this phenomenon. From the results in Fig. 6, we can say that the US-TEMPO oxidation system, which gave less aldehyde content, was more efficient than the TEMPO oxidation system without US treatment.

The DP_v calculated based on the viscosity of the pulp after post-oxidation increased by about two-fold when the oxidation was performed at 5, 15, and 25°C in comparison with the DP_v of the oxidized pulp samples before post-oxidation (Fig. 7). A possible reason to this effect could be the presence of aldehyde groups generated during the oxidation process, which may have rendered the cellulose polymers vulnerable to depolymerization in the highly alkaline CED solution used for the viscosity determination. A post-oxidation treatment converted the aldehyde groups to carboxyls and thus eliminated any kind of possible damage due to the highly alkaline conditions during CED viscosity measurement. Celluloses containing carbonyl groups have been found to be depolymerised by CED solution during the viscosity measurement (β -elimination) (Chandra and Gratzl 1985; de Nooy et al. 1996; Godsay and Pearce 1984; Isogai and Kato 1998). This undesirable effect occurred in addition to the cellulose depolymerization caused by sodium hypochlorite during the TEMPO-mediated oxidation. It has also been shown that the carbonyl groups on polysaccharides could lead to alkali-induced β -alkoxy elimination reactions (Calvini et al. 2004; Gratzl 1987; Gratzl 1990; Samuelson 1970; Theander 1975). The β -elimination causes chain rupture, decreasing the pulp viscosity and hence the DP_v. The increase in DP_v after post-oxidation was less pronounced when the pulps were oxidized at higher temperatures such as 35 and 45°C. This phenomenon could be attributed to the fact that at higher temperatures (e.g. >35°C)

the pulp underwent substantial depolymerization, damaging considerably the intrinsic strength of the cellulose polymer and thus leading to the loss in DP and dissolution of the oxidized fraction. Hence the DPv of the oxidized pulp could not be restored to the same extent as was observed in the case of pulps oxidized with US-TEMPO or TEMPO-system at temperatures less than or equal to 25°C. Elimination or conversion of the aldehyde groups either by reduction back to the hydroxyl groups with sodium borohydride (Chirat et al. 1993; Du Manoir 1980) or by oxidation to carboxyls with sodium chlorite as found in this study could be an efficient way to reduce the risk of further depolymerisation of the oxidized pulp during the viscosity measurement using CED solution. If we neglect the other unknown reactions, alkali-induced β -elimination reactions due to aldehyde groups may be one possible reason for such depolymerisations, as only carbonyl groups were concerned during the post-oxidation with sodium chlorite. Thus, the DPv of the pulp after post-oxidation can be considered as close to the true DP of the TEMPO oxidized pulps with CED method.

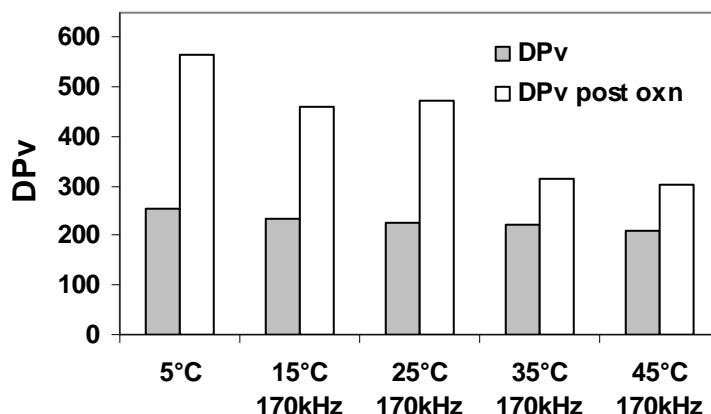


Fig. 7. DPv before and after post oxidation of the US-TEMPO (170 kHz), and TEMPO oxidized pulps at various temperatures

Trend of NaOH consumption in US-TEMPO-system at different temperatures

The trend of the NaOH consumption versus time at all the temperatures studied is shown in Fig. 8. One of the major observations during the oxidation with US-TEMPO-system and TEMPO-system was the consumption of the NaOH, and the final pH after stopping the reaction with ethanol at the end of the reaction time. It was found that the consumption of NaOH increased gradually during the reaction. At a given point of time the consumption of the NaOH was more significant in the case of US-TEMPO-system compared to TEMPO-system alone, which provides indirect evidence of the efficacy of the US-TEMPO-system. At all the temperatures studied the consumption of NaOH was apparently higher, in most of the cases, when oxidation was carried out with US-TEMPO-system at 170kHz compared to US-TEMPO-system at 68kHz and TEMPO alone without US. This would mean that US-TEMPO-system with high frequency ultrasound is more efficient in oxidizing native cellulose compared to one with the lower frequency at the same intensities.

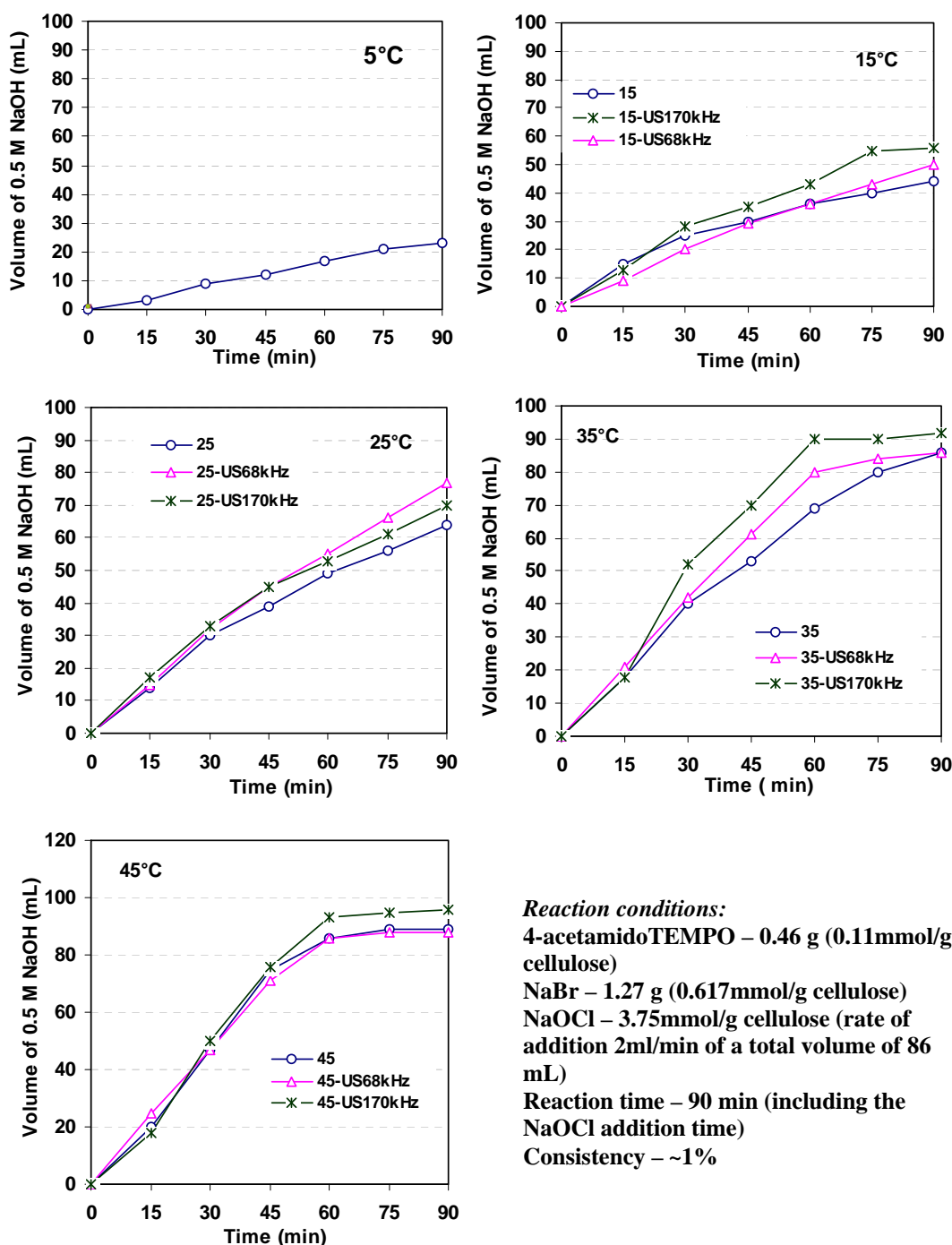


Fig. 8. Consumption of 0.5 M NaOH solution versus reaction time at various temperatures

Several conclusions could be drawn from the results shown in Figs. 8 and 9. First, at a given temperature the NaOH consumption, which can be considered as an indication of the extent of carboxylate formation on the C6 carbons, a faster reaction occurred in the case of oxidations with US-TEMPO-system compared to TEMPO-system alone. The NaOH was consumed during the oxidation in order to neutralize the generated carboxylic

acid groups and to maintain the reaction pH 10.0. However, when the oxidation was conducted at 45°C, the kinetics was mainly driven by the temperature. Second, after a certain time the consumption of NaOH stopped, remaining nearly constant for the rest of the reaction time. This indicates the complete consumption of NaOCl during the reaction; i.e. NaOCl became limiting in this particular case. The consumption of NaOH was stagnant when the oxidation was carried out at 35°C with 170 kHz or at 45°C after about 90 mL of 0.5 M NaOH. In such cases the reaction could be stopped after 60 minutes instead of 90 minutes because all the available sodium hypochlorite was consumed within 60 minutes.

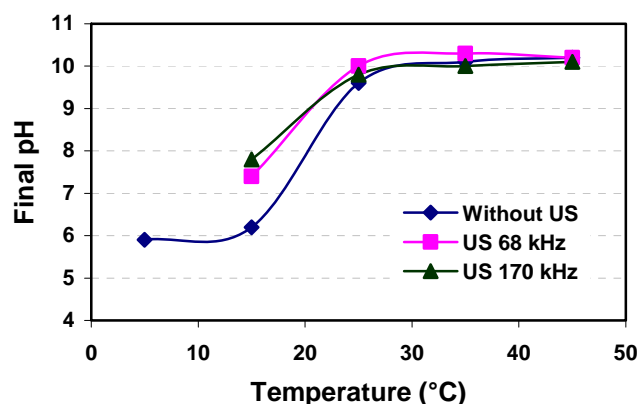


Fig. 9. Final pH versus temperature of the pulp suspension after stopping the oxidation reaction with EtOH)

Third, the oxidation reaction was complete at 35 and 45°C in all the cases. It was nearly completed at 25°C when the US treatments were applied under the given experimental conditions (observed final pH ~10) (Figs. 8 and 9). In contrast, at low temperatures (e.g. 5 and 15°C) there was a lot of unreacted NaOCl, and ethanol was added to quench the reaction. As a result, a lot of acidic compounds were formed. This led to the in situ formation of acetates which in turn caused a lowering of the reaction pH because of the buffering effect. It should be noted that the final reaction pH was adjusted to 7.0 at the end of reaction and before filtration. Saito and Isogai (2004) have also reported a similar correlation between NaOH consumption and the formation of carboxylate and aldehyde groups during TEMPO-mediated oxidation of cellulose cotton linters at pH 10.5.

Oxidation of Pulp with US-TEMPO-System at pH 8.5-9.0

Bragd et al. (2004) reported low pH optima, ca 8.0 for a bromide free 4-acetamidoTEMPO/NaOCl oxidation system of carbohydrates. To find out whether similar pH optima would work for the 4-acetamidoTEMPO/NaBr/NaOCl system with native cellulose, pulp samples were subjected to US-TEMPO-system oxidation at pH 8.5-9.0. The results were compared with those obtained at pH 10.0, as shown in Fig. 10. The pulp oxidized at pH 8.5-9.0 had lower carboxyl content compared to that oxidized at pH 10.0. However, the DP_v and yield after oxidation showed better selectivity at pH 8.5-9.0 compared to pH 10.0. De Nooy et al. (1996) have also reported better selectivity (less fiber degradation due to β-elimination) at pH 9.2-9.7 in a study on the oxidation of

pullulan with TEMPO system. Despite the higher DPv and yield at pH 8.5-9.0, the oxidation of native cellulose with US-TEMPO or TEMPO system alone would be preferred at pH 10.0-10.2, because the yields of the cellulose nanofibers are directly proportional to the carboxylate content in the oxidized pulp. This will be discussed in later sections.

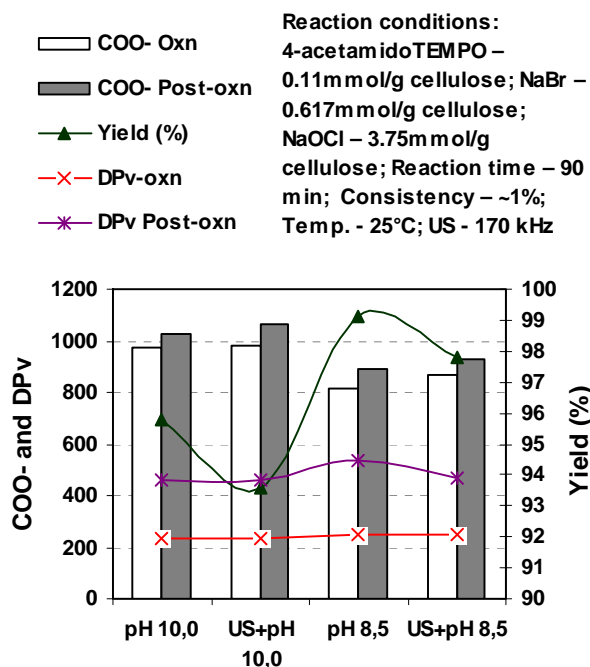


Fig. 10. Carboxyl groups, DPv, and yield versus reaction pH for the US-TEMPO and TEMPO oxidized pulp

Effect of NaBr

Although catalytic amounts of NaBr and TEMPO are required in the TEMPO oxidation system, these two reagents have significant influence on the outcome of reaction if they are insufficiently dosed, for example yielding longer reaction times and lower desired functionalities. In the present work the effect of NaBr on the carboxyl groups, DPv, and yield of the pulp oxidized by the US-TEMPO-system was examined. The carboxyl group content and DPv before and after post-oxidation and yield after oxidation with US-TEMPO and TEMPO alone are presented as a function of NaBr charge and shown in Fig. 11. Under the given experimental conditions the amount of carboxyl groups increased, and DPv and yield decreased with increasing the amount of NaBr. There was a clear-cut advantage in the case of the US-TEMPO-oxidation system in terms of carboxyl content. The carboxyl content of the samples oxidized in the US-TEMPO-system was higher compared to TEMPO-system without US at all NaBr charges. At the maximum charge of NaBr (1.23 mmol/g) a carboxyl content of 1350 mmol/kg and about 88% yield were obtained in the US-TEMPO-system, while the TEMPO system gave a slightly lower carboxyl content of 1300 mmol/kg and a higher yield of ~92%. Analyses of the pulp after post-oxidation with NaClO₂ showed similar

trends for the DP_v, as previously discussed in the case of experiments conducted at different temperatures. This further showed that besides the loss in DP_v during the oxidation with US-TEMPO or TEMPO-systems, the oxidized cellulose fibers were further depolymerized during the course of viscosity determination with CED solution. It seemed that the carboxyl content fell somewhat during the post-oxidation when the pulps contained high carboxyl content (e.g. oxidation at 24.69 mmol NaBr /20g pulp with US). The carboxyl content after post-oxidation was slightly inferior to that of the oxidized pulp in this case. However, the presence of carbonyl function was evident from the fact that the DP_v after post-oxidation was almost doubled in all the cases (Fig. 11).

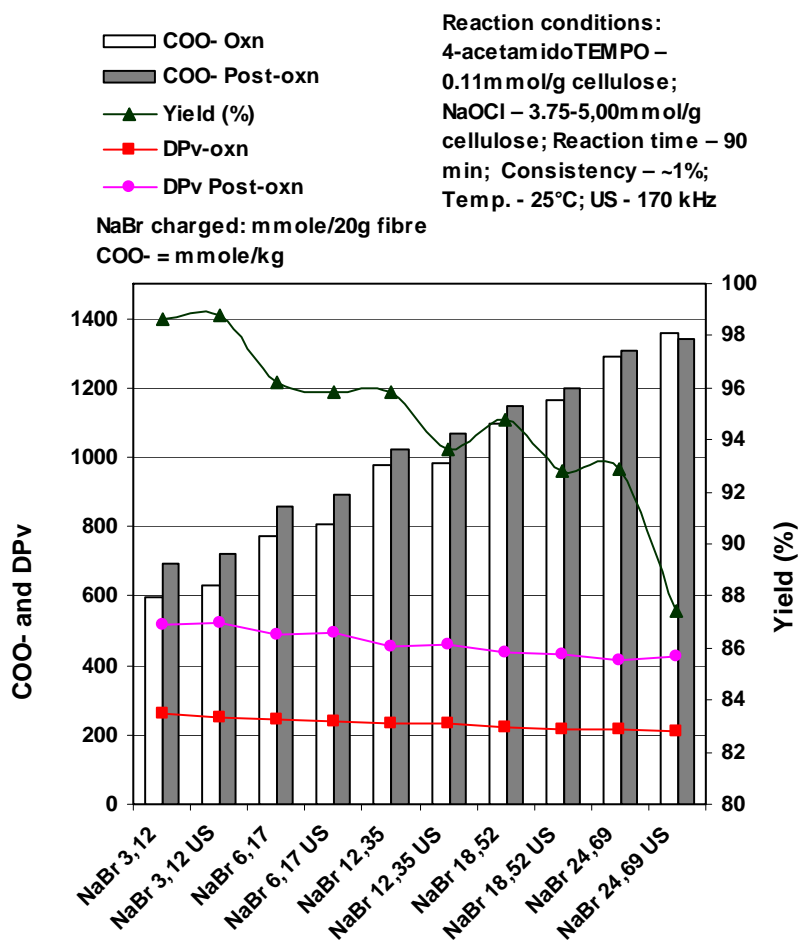


Fig. 11. Carboxyl groups, DP_v, and yield versus NaBr charge on the US-TEMPO and TEMPO mediated oxidation of pulp. The numbers associated with NaBr on the X-axis indicates the dose of NaBr (mmol/20g pulp) applied with and without US.

de Nooy et al. (1995) found a linear relationship between the rate of oxidation of methyl α -D-glucopyranoside, and the concentration of TEMPO and NaBr. It was also shown that OBr⁻/HOBr is more reactive oxidant than OCl⁻, and that in the absence of NaBr the reaction rate was decreased significantly (de Nooy et al. 1995). Same reaction rates can be obtained by decreasing the concentration of TEMPO, and increasing the concentration of NaBr (Anelli et al. 1987; de Nooy et al. 1995). The finding of this study

was in agreement with the results of de Nooy et al.; however, a detailed mechanism to explain this effect is unknown. An increased number of redox cycles at higher concentrations of Br^- (or TEMPO) in the presence of excess of NaOCl (which is normally the case in the TEMPO-mediated reaction) may be a possible explanation. This would mean that the selection of the amount of NaBr in the US-TEMPO- and TEMPO-oxidation system can be optimized according to the desired target properties (e.g. carboxyls content, DP_v and yield, etc.).

Production of Nanocellulose

Cellulose nanofibers were prepared from samples oxidized with US-TEMPO and TEMPO-system alone. These oxidized pulps contained different levels of carboxyl content and were disintegrated by mechanical treatment. The nanocelluloses obtained were observed under Transmission Electron Microscope (TEM) to verify their nano-size structures. A TEM micrograph of the cellulose nanofibers is shown in Fig. 12.

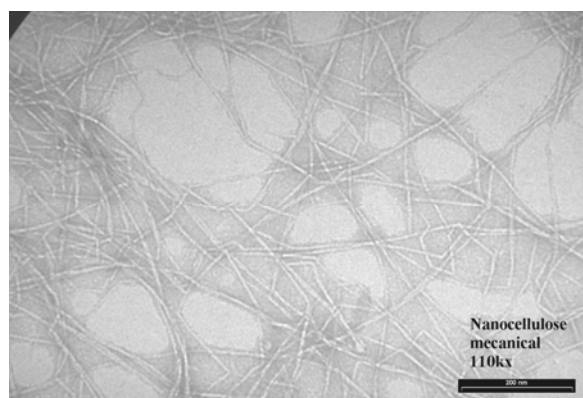


Fig. 12. TEM micrograph of the cellulose nanofibers prepared by mechanical treatment of the pulp oxidized with TEMPO-system

The TEM micrograph shows a number of the cellulose nanofibers with apparent kinks in them. The diameters of the individual nanofibers were measured, and were found to be in the range of 3 to 6 nm with varying length (not measured). The yield of the nanocellulose was quantified gravimetrically, as described in the experimental section. The yield of nanocellulose as a function of carboxyl content is presented in Fig. 13, which shows that the yield of nanocellulose increased with increasing carboxyl content. There was a clear correlation between the yield of cellulose nanofibers and carboxyl content. It was also found that at a given carboxyl content the US-TEMPO-system gave higher nanocellulose yield compared to TEMPO-system without US. For example, at a carboxyl content of ca. 1000 mmol/kg the US-TEMPO-system yielded about 10% more nanocellulose than the TEMPO alone did. However, the difference narrowed down when the carboxyl content reached 1100 mmol/kg, as revealed in Fig. 13.

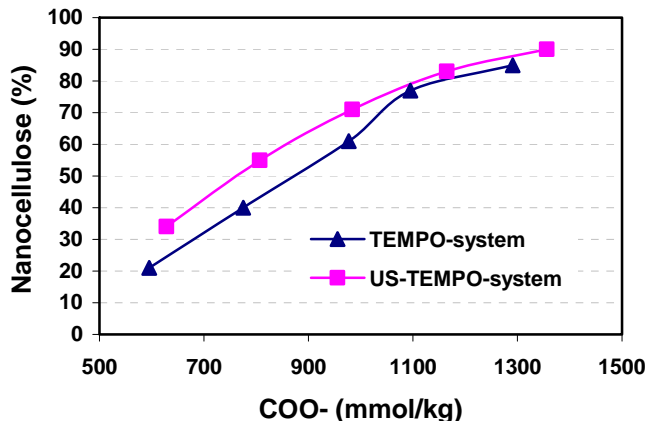


Fig. 13. Nanocellulose versus carboxyls content

The fact that the US-TEMPO-system produced higher carboxyl content and a great yield of nanocellulose in comparison to the usual TEMPO system could be attributed to the physical effects of the US treatment on the cellulose polymer chains. According to Suslick and Price (1999) there exist four physical effects responsible for the high-energy phenomena of the ultrasonic cavitation in liquid-solid systems. They are: (1) improvement of mass transport from turbulent mixing and acoustic streaming, (2) the generation of surface damage at liquid-solid interfaces by shock waves and microjets, (3) the generation of high velocity interparticle collisions in slurries, and (4) the fragmentation of friable solids to increase surface area. As we have mentioned earlier, the US treatment alone did not give any increase in the carboxyl content (Table 1), so the above mentioned phenomena could rightly be applicable for explaining the effects observed with cellulosic fibers. The collapse of cavitation bubbles in the reaction medium with the presence of oxidants facilitated the access of the reactive species to the reaction site (turbulent mixing and mass transfer), C6 hydroxyls in this case, with a greater ease, increasing the reaction rate and, consequently, increasing the carboxyl content. Meanwhile, the simultaneous damage to the fiber surface caused by the shockwaves and microjets facilitated the separation of nanofibers by mechanical treatment.

The results in Table 1 were obtained with ultrasound alone without the oxidants and did not represent the same chemical environment as the oxidations with TEMPO systems. Whether the oxidation of cellulose with the TEMPO-system in the presence of ultrasound will still be regioselective at C6 is not known. Hydroxyl radicals generated by sonolysis of water in the presence of TEMPO/NaBr/NaOCl may cause a possible oxidation of the C2 and C3 hydroxyls. The pulps oxidized with TEMPO- and US-TEMPO-system were subjected to ^{13}C NMR analysis to confirm the presence of any possible C2 and C3 carbonyls or carboxyls. The ^{13}C NMR spectra of the reference, TEMPO- and US-TEMPO-system oxidized pulps are shown in Fig. 14.

Besides the resonance peaks for C1-C6 carbons (usually observed in ^{13}C NMR of cellulose) in all the samples a peak at 177 ppm for the C6 carboxyls was obtained in the case of pulps oxidized with TEMPO- and US-TEMPO-systems. However, no peaks for the carbonyl carbons of ketones (200-210 ppm) which may form at C2 or C3 (Isogai and

Kato 1998) by the US-TEMPO or TEMPO-mediated oxidation were detected. Another observation is that the resonance peaks at 177 ppm were very small despite a carboxyl content of about 1000 mmol/kg on dry pulp. Therefore, any small contribution due to the C2 or C3 carboxyls was either not present or beyond the detection limit of the NMR analysis in this case. Another observation is that there was no additional ^{13}C NMR peak in the samples oxidized with US-TEMPO-system compared to the TEMPO-system. This confirms our assumption that, like TEMPO-system, the oxidation with US-TEMPO-system regioselectively occurs at C6 primary hydroxyl groups.

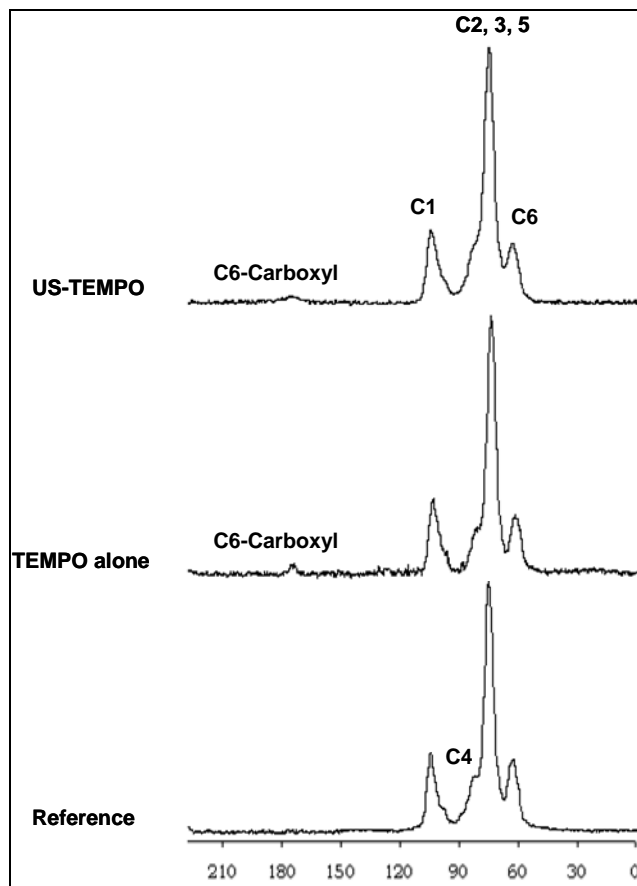


Fig. 14. NMR spectra of the reference and TEMPO- and US-TEMPO-system oxidized pulps

Compared to TEMPO alone, the US-TEMPO-system gave lower yield for the oxidized cellulose when the pulps were oxidized to high levels of carboxyl content. This nature needs further investigation, and experiments are being conducted to analyze the inherent pulp constituents before and after US-TEMPO oxidation. This will allow us to better understand the composition of the nanocellulose fraction and the fraction that remains unchanged after mechanical treatment. It is known that the bleached hardwood kraft pulp still contains at least 10-15% of hemicelluloses, which are amorphous in nature. The loss of hemicelluloses during the US-TEMPO oxidation could be one possible reason for the yield loss. As the oxidation of the cellulose proceeds, the interlinked hemicellulose components could be easily accessible to the oxidants and

consequently, could be eliminated by dissolution. The analysis of soluble fraction after US-TEMPO oxidation is also envisaged.

CONCLUSIONS

1. An ultrasound-assisted TEMPO-system can facilitate the oxidation of native cellulose for the production of nanocellulose. This system gave a 10 to 15% increase in carboxyl content and 10% augmentation in nanocellulose yield under optimized conditions.
2. Reaction temperature and pH have significant affect on the properties of oxidized pulp. To obtain acceptable carboxyl content, yield and DP_v of the oxidized pulps, the following conditions may be employed for the US-TEMPO-system: pH 10.0 to 10.5 and temperature 25°C.
3. Measurement of NaOH consumption at regular intervals and end pH after the termination of reaction with ethanol can offer valuable information on the status of the US-TEMPO- oxidation system and possible outcomes.
4. The dosage of NaBr plays a crucial role in oxidation with or without US treatment in terms of carboxyl content of the oxidized pulp; it can be optimized to obtain the desired properties.
5. From the NMR analysis data it can be concluded that like TEMPO-system, oxidation with US-TEMPO-system is also regioselective, and occurs mainly at C6 primary hydroxyl groups.

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