

Combined Production of Polymeric Birch Xylan and Paper Pulp by Alkaline Pre-extraction Followed by Alkaline Cooking

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S Supporting Information

ABSTRACT: Alkaline pre-extraction of birch wood was performed to isolate polymeric xylan and subsequently produce a paper-grade pulp. At 95 °C and 2.5 mol/L NaOH, 7% of wood was transferred to the E-lye as polymeric xylan with an anhydroxylose-lignin ratio of 6.5. Xylan with a weight-average molar mass of 20 kDa was quantitatively precipitated from the solution previously concentrated from 7.4 to 37 g/L. The anhydroxylose-lignin ratio in the carbohydrate fraction increased to 29 g/g upon precipitation. Enzymatic hydrolysis of the commercial birch xylan with Pentopan Mono PG resulted in a uniform xylooligosaccharide product with low xylose content at a yield of 61%. The pre-extracted pulp had excellent papermaking properties but its yield was 4.9% units lower than that of the reference pulp. Commercial potential of the modified process was discussed.

1. INTRODUCTION

Pulp market prices are currently driven by the modern production facilities in the Southern Hemisphere, benefiting from the low-cost raw materials, state-of-the art technologies, and economy of scale. Creating new value-added products from noncellulosic side streams could be the key to market success for pulp manufacturers in the Northern Hemisphere where hemicelluloses—branched wood polysaccharides—are seen as a potential raw material for such products. *O*-Acetyl-4-*O*-methylglucurono- β -D-xylan (hereafter referred to as xylan) is the dominating hemicellulose type in hardwood species. At present, typical commercial xylan-derived products include but are not limited to xylose, xylitol, and furfural. A variety of traditional and new products and applications of polymeric xylan, xylooligosaccharides (XOS), and xylose is summarized in a review by Deutschmann and Dekker.¹

Birch, the dominant hardwood species in Northern Europe, contains approximately 25% of xylan. However, its retention in the solid phase during kraft pulping is only about 50% of the initial xylan content, while in the case of *Eucalyptus globulus*, kraft pulp retains 75% of the xylan in the unbleached pulp when comparing both pulps at a kappa number of 18.6.² The high alkali stability of xylan in *E. globulus* has been connected to its substituted uronic acid units, which are absent in birch xylan.² A low contribution of wood hemicelluloses to the calorific value of the black liquor as well as a wide range of potential hemicellulose-derived products creates an interest in their isolation even before alkaline pulping.³

Isolation of hemicelluloses in alkaline,^{4,5} neutral,⁶ and acidic⁷ environments before or after pulping^{8,9} as well as isolation from kraft pulping liquor at the different stages of the cook¹⁰ has

returned to the forefront of science. In alkaline pre-extraction, a fraction of the hemicelluloses can be dissolved in polymeric form and reintroduced to the pulp in subsequent process stages, as demonstrated by Muguet et al.,¹¹ or utilized elsewhere in a polymeric or depolymerized form. The advantages of alkaline pre-extraction compared to other pretreatments include its compatibility with kraft or soda pulping, the absence of sticky lignin constituents formed in acidic conditions,¹² and the commercial availability of the membrane separation processes for extracted hemicelluloses.¹³ However, despite the mildness of the pre-extraction process, changes in the hemicellulose content may affect the yield of the pulp and its properties.

Soda-anthraquinone (SAQ) pulping is a sulfur-free alternative to the kraft process where AQ replaces sodium sulfide as a nucleophile. SAQ pulping has already proved its potential to produce high-quality pulps.¹⁴ The potential advantage of a sulfur-free process over the kraft process is that the resulting black liquor can be directly utilized in gasification producing over two times more electrical energy than a conventional recovery boiler-based energy recovery cycle. Recently, however, AQ has been removed from the list of recommended chemicals for the food packaging industry due to its carcinogenic potential according to the German Federal Institute for Risk Assessment (BfR).¹⁵

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In the present study, pre-extraction of polymeric xylan from birch chips was performed using concentrated NaOH solutions in the absence of a nucleophile to minimize the concomitant extraction of lignin. The process conditions were optimized aimed at the recovery of high-molecular weight xylan with low lignin coextraction. A membrane filtration technique was applied to the liquid pre-extract (E-lye) in order to concentrate the xylan solution and to separate NaOH for the process recirculation. Concentrated xylan was precipitated to obtain a potentially marketable product. Production of XOS as a potential xylan-derived product was evaluated by applying specific enzymatic hydrolysis. We also aimed for the production of a paper-grade pulp by the pre-extraction SAQ (E-SAQ) process with subsequent oxygen delignification (O-stage), comparable with a reference birch SAQ pulp.

2. MATERIALS AND METHODS

2.1. Pre-extraction, Pulping and O-Stage. Birch chips (*Betula pendula*) were screened (SCAN-CM 40:01; Scandinavian Standard, 2001) and stored at $-18\text{ }^{\circ}\text{C}$ before the experiments. The dry matter content of the chips was determined (SCAN-CM 39, 1994). For the small-scale optimization of alkaline pre-extraction (Figure 1) two fractions

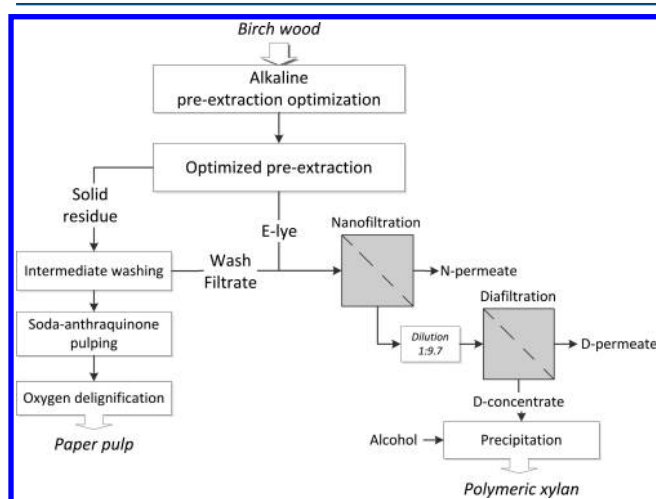


Figure 1. Block diagram of the performed process steps.

of wood chips in thickness ranges of 2–4 mm and 4–6 mm were separated. For the pre-extraction, the chips were placed in stainless steel bombs of 220 mL each and mixed with the corresponding NaOH solution in deionized water while a liquid–solid ratio (L:S) of 10 L/kg was maintained. The bombs were heated rapidly in a silicon oil bath to reach the target temperature. After 60 min at the target temperature, the bombs were cooled in cold water. The free E-lye was collected undiluted, after which the solid residue was washed with deionized water.

Pre-extraction in optimized conditions for the subsequent pulping experiments (Figure 1) was carried out for wood chips of 2–6 mm thickness in a 10-L reactor with external circulation of the liquid phase from bottom to top. The L:S in the large-scale experiments was 9.3 L/kg. After completing pre-extraction, the process was terminated by draining off the E-lye through a cooling coil. The solid residue was washed with deionized water for 5 min at room temperature maintaining a L:S of 1.25 L/kg of oven-dried wood (o.d. wood) in order to reduce the alkalinity in the pulping stage and recover additional

amounts of extracted xylan. Subsequent SAQ pulping was performed without opening the reactor with the resulting effective alkali (EA) charge of 18.7% o.d. wood, an AQ charge of 0.075%, and the addition of deionized water to maintain a L:S of 3.5 L/kg, all values calculated on raw wood. Pulping was performed at $160\text{ }^{\circ}\text{C}$ with an H-factor of 550. The resulting pulp was washed in the reactor in three consecutive stages for 20 min at $60\text{ }^{\circ}\text{C}$, 20 min at $45\text{ }^{\circ}\text{C}$, and 10 min at room temperature; after collecting the pulp from the reactor, manual washing was performed. The pulps were then screened through a 0.35 mm Mänttä vibrational slit screen, after which the amount of rejects, pulp yield, kappa number (SCAN-C 1:00, 2000), and intrinsic viscosity (SCAN-CM 15:99, 1999) were determined. Reference SAQ pulping without pre-extraction was performed with an H-factor of 800, a NaOH charge of 22% EA, an AQ charge of 0.1%, a L:S of 3.5 L/kg, and a temperature of $160\text{ }^{\circ}\text{C}$.

O-stage was carried out in a batch air bath digester where unbleached pulp was mixed with 20 kg/t o.d. pulp NaOH and 0.5 kg/t Mg (as $\text{Mg}_2\text{SO}_4 \cdot 7\text{H}_2\text{O}$) and pressurized with oxygen to 0.9 MPa, heated to $90\text{ }^{\circ}\text{C}$, and treated for 60 min. The pulps after O-stage were subjected to washing as well as to yield, kappa number, and intrinsic viscosity determinations. The yields of the solid residues and pulps are reported as the ratio of the o.d. mass of the corresponding sample to the o.d. mass of the initial wood.

2.2. Nano- and Diafiltration. The objective of the nano- and diafiltration was concentrating the extracted xylan and separating most of the NaOH from the organic fraction. A mixture of the raw E-lye and the wash filtrate was used as feed in the nanofiltration experiments (Figure 1), while for the subsequent batch mode diafiltration the concentrate from the nanofiltration step (N-concentrate) was diluted by a factor of 9.7. The experiments were carried out using an Alfa Laval TestUnit M20 filtration unit (Nakskov, Denmark) with a 1-kDa cutoff membrane (NP 010; Microdyn-Nadir, Wiesbaden, Germany) at a cross-flow of 16 L/min, inlet pressure of 32 bar, and pressure loss of 2–4 bar at a temperature of $48\text{--}50\text{ }^{\circ}\text{C}$.

2.3. Xylan Precipitation. After diafiltration, the xylan in the concentrate (D-concentrate) was subjected to ethanol precipitation at an ethanol–concentrate ratio of 1 L/L. The mixture was then centrifuged for 15 min at a rotational speed of 4500 rpm. The supernatant was decanted and the sediment was redispersed in deionized water for purification. This was followed by isopropyl alcohol addition at a ratio of 1 L/L and then further centrifugation as described above. The solid fraction was freeze-dried and stored at room temperature for further analyses.

2.4. Enzymatic Hydrolysis of Polymeric Xylan. Preliminary studies on enzymatic hydrolysis were carried out on commercial alkali-extracted birch xylan (X-0502; Sigma, Germany) with the commercial food-grade endoxylanase, Pentopan Mono BG (Novozymes, Denmark), which is produced by *Aspergillus oryzae* carrying the gene encoding glycoside hydrolase (GH) family 11 endoxylanase from *Thermomyces lanuginosus*. The protein content of Pentopan Mono BG was analyzed by the commercial Bradford assay (Bio-Rad, Hercules, CA, USA) to be 13 mg/g of product. Before use, the commercial xylan was dissolved in a NaOH solution, neutralized with HCl, and dialyzed against water. The nondried xylan suspension (10 g/L) in 50 mM sodium acetate buffer (pH 5.0) was hydrolyzed using two enzyme dosages (0.65 and 6.5 mg of enzyme protein/g of xylan) at $60\text{ }^{\circ}\text{C}$ for 4

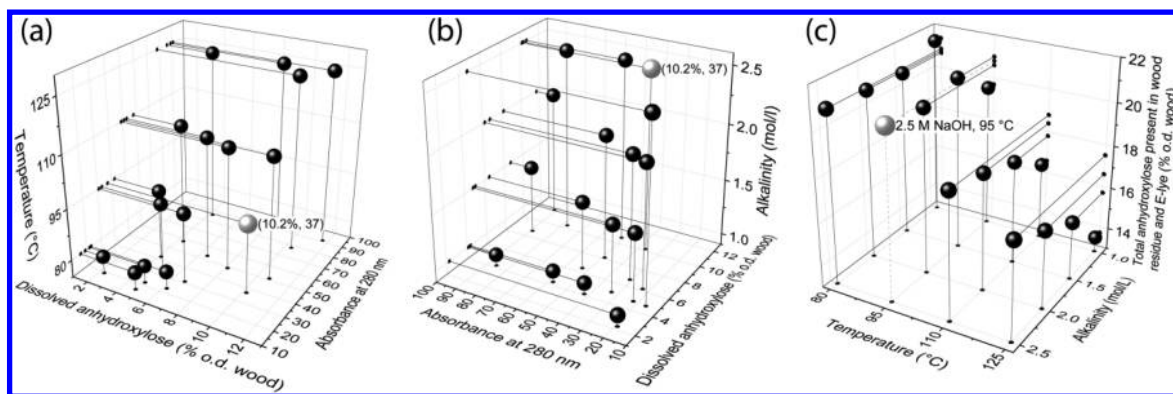


Figure 2. The effect of extraction conditions on the amount of anhydroxylose and the lignin absorbance at 280 nm in the E-lye (a and b) as well as on total anhydroxylose present in solid residue and E-lye after pre-extraction (c). Extraction was performed in 0.22-L reactor. (a) Effect of temperature. The alkali concentration was varied between 1.0, 1.5, 2.0, and 2.5 mol/L. (b) Effect of NaOH concentration. The temperature was varied between 80, 95, 110, and 125 °C. (c) Effect of the pre-extraction temperature and alkalinity on the total xylan balance in the E-lye and the solid residue. Selected optimum is marked by gray bullets.

h after which the reaction was stopped by keeping the samples in a boiling water bath for 10 min. The yields of the hydrolysis products were calculated based on the xylan content in the sample.

2.5. Papermaking Properties. For the papermaking properties evaluation, the pulps were refined in a PFI mill at 10% consistency (ISO 5264-2:2002) after which the Schopper–Riegler drainability (ISO 5267-1:1999) was determined. Handsheets with the grammage approximately 60 g/m² were prepared using a KCL model machine (ISO 5269-1:2005). Tensile (ISO 1924-2:1994), tear (ISO 1974:1990), as well as dry and wet zero-span strength (ISO 15361:2000), brightness (ISO 2470:1999), grammage (ISO 536:1995), and density (ISO 534:1988) were measured. Specific values (indices) for the strength properties were calculated.

2.6. Analytical Procedures. The solid phase (birch wood, solid residue, and pulps) was air-dried to the dry matter content of 90–95% and ground in a Wiley mill. The Klason lignin, acid-soluble lignin (ASL) content, and carbohydrate composition (NREL/TP-510-42618; NREL, 2008) were determined. Carbohydrates were quantified using a Dionex ICS 3000 high performance anion exchange chromatography (HPAEC-PAD) (Dionex, Sunnyvale, CA, USA).

Residual alkali concentration in the E-lye and black liquor was determined according to SCAN-N 33:94 using an automatic Mettler Toledo DL53+ titrator (Greifensee, Switzerland). In the E-lye, lignin absorbance in the UV-region was measured at 280 nm. The obtained absorbance values were multiplied by the dilution factor and used as a relative measure of lignin content since no exact extinction coefficient was available for the performed treatment. The carbohydrates in the E-lye were quantified according to NREL/TP-510-42623, NREL 2008 with the chromatographic setup described in the preceding section. In the large-scale experiments after complete acid hydrolysis, low-molecular weight lignin (ASL) in the E-lye was quantified using UV spectroscopy at a wavelength of 205 nm according to Tappi UM 250 1991 and acid insoluble high molecular weight lignin (AISL) gravimetrically.

Molar mass distribution of the xylan samples isolated from the E-lye was determined by size-exclusion chromatography with a refractive index detection and 0.5 M NaOH as eluent. The chromatographic system consisted of a precolumn (MCX 1000, 10 m, 8 mm × 50 mm; PSS, Mainz, Germany) and two analytical columns in series (PSS MCX 1000, 10 m, 8 mm ×

300 mm; PSS) to improve separation. The system was calibrated against xylose (Merck, Darmstadt, Germany), cellobiose, maltotriose, stachyose (Fluka, Buchs, Switzerland), dextran (PSS): 1350, 5200, 11 600, 23 800, 48 600, and pullulan (Showa Denko, Tokyo, Japan): 5900, 11 800, 22 800, 47 300, 112 000, 212 000, 404 000.

The amounts of xylose and XOS after hydrolysis were determined by HPAEC-PAD with a CarboPac PA100 column (Dionex).¹⁶ Xylose (Merck), xylobiose, xylotriose, and xylotetraose (Megazyme, Wicklow, Ireland) were used as standards.

3. RESULTS AND DISCUSSION

3.1. Pre-extraction Optimization. The yield of the solid residue upon alkaline pre-extraction was strongly dependent on the pre-extraction temperature, while the effect of alkali concentration on the overall yield could not be clearly defined. The solid residue composition indicated that lignin was almost quantitatively preserved from the dissolution at 80 and 95 °C. At higher temperatures a notable delignification occurred through the cleavage of the ether bonds, and at 125 °C up to half of the wood lignin was transferred to the E-lye. The content of the principal wood components in raw wood, the solid phase, and E-lye after pre-extraction is summarized in Table S1 in the Supporting Information.

Similarly to lignin, the cellulose content remained stable in the wood under the low-temperature extraction conditions. At temperatures of 110 and 125 °C, cellulose started to degrade by alkaline peeling reactions at the reducing end followed by the formation of soluble isosaccharinic acid (ISA).¹⁴ ISA and its degradation products in the E-lye were not quantified; nevertheless, the involvement of peeling reactions was supported by the fact that under all applied pre-extraction conditions, the E-lye contained only up to 0.15% o.d. wood of anhydroglucose likely originating from the extracted polymeric glucomannan. The observed notable cellulose degradation at 110 and 125 °C could result in unacceptably low pulp yield and should be avoided.

Figure 2 demonstrates the effects of temperature (a) and alkali concentration (b) on the amount of extracted anhydroxylose and the absorbance of the E-lye at a wavelength of 280 nm. Xylan dissolution was clearly enhanced as a function of both alkalinity and temperature, showing that up to half of

Table 1. Lignin and Carbohydrate Composition of the Solid Samples (Wood, Solid Residue after Pre-Extraction, Pulps) and Liquid Pre-extraction Samples (E-Lye and Wash Filtrate). The Reactions Were Performed in a 10-L Autoclave

composition	solid samples				E-lye and wash filtrate					
	raw wood	solid residue	SAQ pulp ^a	E-SAQ pulp	E-lye		wash water		total recovered	total theoretical ^b
		% o.d. wood			% o.d. wood	g/L	% o.d. wood	g/L	% o.d. wood	% o.d. wood
Klason lignin	18.8	18.4	0.56	0.58	0.58	0.72	0.054	0.44	0.63	0.67
ASL	4.4	3.1	0.52	0.48	0.33	0.41	0.059	0.48	0.39	0.38
total lignin	23.2	21.5	1.1	1.1	0.9	1.1	0.11	0.92	1.0	1.1
xylose ^c					6.1	7.7	0.49	4.0	6.6	7.2
xylan ^d	24.2	12.2	13.6	9.2						
galactose ^c					0.046	0.58	0.0092	0.075	0.055	0.054
mannose ^c					0	0	0	0	0	0
glucomannan ^d	2.51	0.66	0	0						
glucose ^c					0	0	0	0	0	0
cellulose ^d	39.8	38.6	36.3	36.3						
arabinose ^c					0.015	0.020	0.00087	0.0071	0.016	0.019
total carbohydrates	66.5	51.5	49.9	45.5	6.2	8.3	0.50	4.1	6.7	7.3
yield	100	82.7 ^e /87.5 ^f	51.0	46.6						
Kappa number			17.3	17.9						

^aAn intermediate washing stage with a L:S of 1.2 L/kg was applied before pulping. ^bCalculated with the assumption that all E-lye is recovered. ^cAs anhydrosugars. ^dXylan, glucomannan, and cellulose content in the solid phase calculated using Janson formulas.¹⁷ ^eYield after one-stage washing (weight of residual NaOH excluded). ^fUnwashed yield (weight of residual NaOH excluded).

the birch xylan could be transferred to the E-lye under the applied conditions. In addition to xylan, small amounts of arabinose (<0.1%), galactose (<0.2%), and glucose (<0.2%), reported as anhydrosugars, were detected in all E-lye samples. Importantly, xylan dissolution efficiency should be compromised with the amount of coextracted lignin, implying that lignin retention in the solid phase should be as high as possible at the maximum xylan extraction. Obviously, the highest efficiency of xylan extraction at minimum lignin coextraction falls in the area with a low temperature and a high pre-extraction alkalinity (Figure 2). On the basis of this observation, experiment 8, performed at 95 °C in 2.5 M alkali solution (Supporting Information, Table S1), had optimal selectivity toward xylan extraction, allowing 10.2% of anhydroxylose based on o.d. wood to be transferred to the liquid phase at a low level of lignin coextraction. The amount of the extracted anhydroxylose in the liquid phase was calculated by assuming that the E-lye could be fully recovered. The recovery of the E-lye is strongly dependent on the L:S in the pre-extraction and on the pre-extract release conditions.

The difference between the reduction in anhydroxylose content in the solid residue and its amount detected in the E-lye suggests that the extraction at 110 and 125 °C caused substantial xylan loss through its degradation in alkali. Despite the fact that xylan is stable to alkaline peeling due to the presence of α -(1 \rightarrow 2)-linked 4-O-methylglucopyranosyluronic acid (4-O-MeGlcA) residues,¹⁴ some degradation of xylan obviously may have occurred through secondary peeling resulting from the cleavage of glycosidic bonds.¹⁹ It was also evident that high alkalinity had a positive effect on the total xylan balance (Figure 2c), specifically, smaller amounts of xylan were degraded likely due to the higher preference toward the stopping reaction. Furthermore, Al-Dajani and Tschirner⁴ demonstrated that no monomers were present in the E-lye, which conforms to the nature of alkaline degradation of carbohydrates.

The effect of a reduced thickness of the chips in the studied range was small, in terms of xylan extractability; therefore, industrial scale pre-extraction is not restricted to thin chips.

3.2. Alkaline Pre-extraction at Larger Scale. The pre-extraction process conditions corresponding to experiment 8 (Supporting Information, Table S1) were applied in a 10-L digester. In contrast to the small-scale experiments, the yield of the solid residue in Table 1 was measured without washing (87.5%) and after a one-stage washing at a L:S of 1.2 L/kg (83.7%). The corresponding yield of experiment 8 after thorough washing was 75.5%.

The pre-extraction efficiency of the large-scale experiment generally corresponded to that in the optimization phase, which is reflected in the content of anhydroxylose in the solid residue (Table 1 and Supporting Information Table 1). However, the degradation of wood carbohydrates appeared to be somewhat more intensive at the large scale presumably due to the temperature gradient in the larger reactor setup. The reduction of the lignin content in the solid residue was mainly observed in the acid soluble lignin (ASL) corresponding to the low-molecular lignin fraction and only to a small extent in Klason lignin.

Only 80–85% of the E-lye could be separated in an undiluted form under the studied extraction conditions with the rest remaining in wood pores and partly in the pipelines of the reactor setup. Introducing a short washing stage of the solid residue after pre-extraction enhanced the recovery of the dissolved wood components by 8% compared to the recovery from the undiluted E-lye alone (Table 1). Furthermore, the wash filtrate contained anhydroxylose at a concentration comparable to that in the undiluted E-lye due to the restricted wash water amount (L:S 1.25 L/kg). The total xylan recovered from the E-lye and the wash filtrate was 6.1% (7.7 g/L) and 0.5% o.d. wood (4.0 g/L), respectively. The high-molar mass of the carbohydrate fraction in the E-lye of $M_n = 10.3$ kDa, $M_w = 19.8$ kDa, and $M_z = 35.8$ kDa confirmed a significantly lower

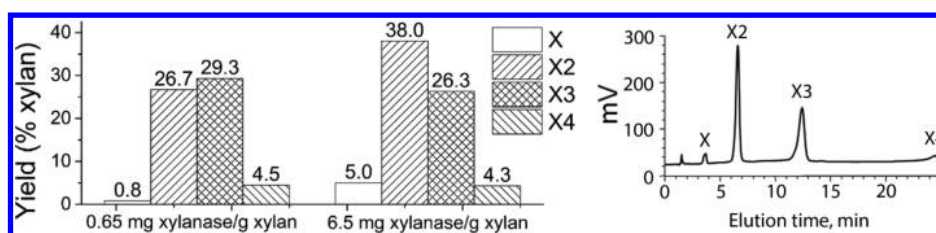


Figure 3. Production of XOS from X-0502 (xylan supplied by Sigma) by specific hydrolysis with Pentopan mono PG xylanase at 60° for 4 h. (Left) effect of xylanase charge; (right) product pattern after hydrolysis with 0.65 mg xylanase/g xylan. X-xylose numbers 2, 3, and 4 denote the number of xylopyranosyl units in an oligosaccharide.

degree of degradation compared to that of a xylan extracted from bleached pulp.⁸ The low polydispersity index of 1.9 was comparable to that reported by Sjöström²⁰ for native xylan.

Upon the complete acid hydrolysis of the E-lye comparable amounts of Klason lignin and ASL were measured which corresponded to the high- and low-molar mass lignin fractions, respectively. The total amount of the extracted lignin corresponded to 1% of o.d. wood with the concentration in the undiluted E-lye of 1.3 g/L.

Importantly, the mass reduction of the wood upon pre-extraction and one washing stage (17.3% o.d. wood) exceeded the total mass of the compounds measured in the E-lye and the wash filtrate (7.7% o.d. wood) by 9.6%. The cleaved xylan acetyl groups (4.1% o.d. wood recovered with the E-lye and wash filtrate assuming complete cleavage of total wood xylan of 4.75% o.d. wood¹⁸), uronic acid substituents in the extracted xylan (approximately 0.8% o.d. wood assuming a degree of substitution of 0.08¹⁸), and the extractive compounds (maximum of 2%¹⁸) contribute to the total amount of the organic constituents in the E-lye and the wash filtrate. Additionally, small amounts of carbohydrate degradation products, mainly organic acids originating from xylan and glucumannan, may be present in the liquid phase.

3.3. Membrane Filtration and Xylan Precipitation.

Sequential nano- and diafiltration of the E-lye was performed in order to concentrate the xylan solution for the subsequent precipitation in ethanol (Figure 1). The resulting concentrates and permeates of the nano- and diafiltration were denoted with N- and D- prefixes, respectively. The feed containing 7.4 g/L anhydroxylose, 1.1 g/L lignin, and 80 g/L NaOH was concentrated in the nanofiltration step transferring 83% of the total NaOH to the N-permeate (Table S2 in Supporting Information). Approximately 14% of low-molecular lignin (ASL) and negligible amount of xylan passed through the membrane of a 1-kDa cutoff. This indicates that a very small share of the low-molecular weight xylan and lignin was present in the E-lye. For the diafiltration stage, the feed was diluted by a factor of 9.7. In this stage, the xylan solution was concentrated to approximately 40 g of anhydroxylose/L, while only 2% of the NaOH in the starting E-lye remained in the D-concentrate fraction (Supporting Information, Table S2). While high-molecular lignin was entirely retained in the D-concentrate, the content of ASL was reduced by 20% compared to that in the starting E-lye. As expected, membrane filtration was an efficient technique in concentrating the organic fraction and separating the alkali. However, the applicability of this technique to separate xylan from the coextracted lignin is limited due to the polydisperse nature of these polymers.

Alcohols are known to be very efficient precipitation aids for polysaccharides owing to their low solvation power.²¹ The addition of ethanol to the D-concentrate resulted in a

quantitative xylan precipitation. On the other hand, only 32% of the total low-molecular (ASL) and 10% of the high-molecular (Klason) lignin coprecipitated and remained in the solid phase after subsequent water and isopropyl alcohol purification stages. In contrast to polysaccharides, lignin is known to have higher solubility in alcohols, particularly after the degradation of aryl ether bonds occurring in alkaline environments.²² For that reason, precipitation of the carbohydrate fraction in alcohols could serve both as means of its purification and isolation as a final product. Further, isopropyl alcohol washing enabled extended removal of salt contaminants due to the lower dielectric constant of the alcohol compared to that of ethanol. The final product contained 2.6% lignin, 0.7% anhydrogalactose, and 0.2% anhydroglucose. The amount of anhydroxylose of only 75.5% was measured by HPAEC-PAD analysis after sulfuric acid hydrolysis. This amount may be somewhat underestimated because of the side reactions.²³ Additionally, the nonquantified fraction contains the side chains of xylan, mainly uronic acid, as well as salts and organic impurities that were not extracted with the solvents. Cleaved xylan acetyl groups did not contribute to the weight of the xylan. Part of the 4-*O*-MeGlcA residues in xylan are typically converted in an alkaline environment to hexenuronic acid (HexA) residues.²⁴ Both 4-*O*-MeGlcA and HexA residues contribute to the overall share of the xylan in the precipitate.

Conversion and applications of the xylan product is governed by its macromolecular properties, chemical structure, and purity. The absence of acetyl groups renders xylan less water-soluble, and together with the partial elimination of 4-*O*-MeGlcA substituents results in a nearly linear polymer. Its high molar mass (20 kDa) suggests the utilization as a polymer, however, probably not without further functionalization.⁸

3.4. Production of Xylooligosaccharides. We evaluated the possibility of producing a mixture of XOS with low xylose content using specific enzyme hydrolysis of an alkali-extracted xylan. Pentopan Mono BG demonstrated high potential for XOS production as only a low amount of xylose was concomitantly formed (Figure 3). An increase in the enzyme dosage facilitated the hydrolysis. The combined yield of xylobiose (X2), xylotriose (X3), and xyloetraose (X4) increased from 60.5% at 0.65 mg Pentopan Mono BG per 1 g of xylan to 68.6% after a 10-fold enzyme protein loading. The product pattern, however, shifted toward the lower molar mass, and the yield of monomeric xylose also increased from 0.8 to 5%. Similar behavior was marked when the hydrolysis time was increased to 24 h (not shown). The nonquantified products corresponded essentially to the oligosaccharides carrying 4-*O*-MeGlcA substituent (not shown), the main one being xyloetraose with 4-*O*-MeGlcA at the penultimate nonreducing xylopyranosyl residue.²⁵ Native birch xylan carries 4-*O*-MeGlcA

substituent on average on every 17th xylopyranosyl residue,²⁶ thus residual MeGlcAX4 will correspond to 24% of the total xylose in birch xylan. The yields of quantified products (X–X4) in the higher enzyme dosage hydrolysis summed up to 74%, meaning that no further yield could be obtained with Pentopan Mono BG. Falck et al.²⁷ reported production of XOS from alkali-extracted birch xylan. However, they used GH10 endoxylanase from *Rhodothermus marinus*, which is prone to produce xylose, and thus limited hydrolysis with low enzyme dosage was applied, resulting in an XOS yield of 20–25%. Metsämuuronen et al.²⁸ compared three commercial endoxylanases in the production of XOS from bleached birch kraft pulp. GH11 endoxylanase from *Bacillus* sp. (Pulpzyme HC) was found to produce XOS without xylose formation with up to 20% yield from pulp xylan. Interestingly, an enzyme product (Bio-Feed) with the same *T. lanuginosus* endoxylanase as is present in Pentopan Mono BG, was releasing a significant amount of xylose.

Enzymatic hydrolysis of alkali-extracted polymeric birch xylan with Pentopan Mono BG demonstrated a high potential to produce a uniform XOS mixture at a good yield. Selective elimination of the xylan side groups, 4-*O*-MeGlcA by α -glucuronidase and HexA by acid, would further increase the yield of linear XOS.

3.5. Pulp Production. The E-SAQ pulping process was optimized in terms of alkali charge, L:S ratio, and pulp-washing conditions. The optimization target was to obtain a pulp with the highest possible yield while maintaining kappa number, intrinsic viscosity, and alkali consumption comparable to those of a reference SAQ process. A careful control of the pulping EA charge, stipulated by the starting alkali charge in pre-extraction as high as 89% o.d. wood, appeared to be a key tool for maintaining pulp quality. To achieve a reliable alkali balance for the described reactor setup, the E-lye was first discharged by pressure relief, after which the EA of 65.6% o.d. wood was recovered. Further, pulping experiments were performed directly after the E-lye discharge and after an intermediate washing of the solid residue with the L:S of 0.6 and 1.2 L/kg, respectively. The pulp produced without intermediate washing had a yield of only 35.4% o.d. wood while introducing an intermediate washing stage resulted in significantly increased pulp yields of 44.7% and 46.6% at the lower and higher washing L:S, respectively. High EA concentration is known to promote random cellulose chain scission, particularly for SAQ pulping.¹⁴ Consequently, new reducing ends are formed which in turn initiate secondary peeling reactions and, subsequently, result in cellulose yield loss. Additionally, dissolution of xylan is enhanced when the alkali concentration is increased.²⁹ The intermediate washing allowed the reduction of the initial EA for pulping by 2.4% o.d. wood (L:S = 0.6 L/kg) and 4.3% o.d. wood (L:S = 1.2 L/kg), respectively.

The alkali consumption during the pre-extraction and pulping stages was determined by measuring the residual alkali concentration in the discharged E-lye and black liquor. According to the measurements, 12.5% of EA on o.d. wood was partially consumed and partially reversibly absorbed³⁰ by wood during the pre-extraction stage. In the selected case where intermediate washing with L:S ratio 1.2 L/kg was applied, the overall EA consumption of the E-SAQ process was 15.7% o.d. wood considering a residual EA in the black liquor of 3.4% o.d. wood (11.5 g/L). Thus, the overall alkali consumption of the E-SAQ cook was slightly lower compared to that of the reference SAQ cook where the initial and the

residual alkali amounts corresponded to 20% and 2% o.d. wood (5.7 g/L), respectively. The lower total EA consumption of the E-SAQ process compared to the reference SAQ pulping may be attributed to the higher total retention of the carbohydrates of the former. The total amount of carbohydrates retained by the pulp and the E-lye was 46.7% and 48.4% for the SAQ (carbohydrates in the pulp only) and E-SAQ processes, respectively (Table 1).

Furthermore, the concentration of the residual alkali in the black liquor after the E-SAQ process (11.5 g/L) was higher than that after the reference SAQ pulping (5.7 g/L), which implies that a further reduction of the EA charge for an SAQ cook after an E-stage would be desirable to increase the pulp yield and viscosity. Since an intermediate washing step (between E and SAQ stages) will not be reasonable on a commercial scale, a more thorough drainage of the E-lye needs to be realized in practice.

We found that in the E-SAQ process a pulping H-factor of only 550 yielded a product with the same kappa number as that of the reference SAQ pulp where an H-factor of 800 was applied. We suggest that smaller amount of alkali consumed for carbohydrate degradation and thorough alkali impregnation in the pre-extraction stage facilitated delignification. Importantly, the amount of rejects in both processes was under 0.1% o.d. wood.

The screened yield of the unbleached E-SAQ pulp was 4.4% units lower (46.6%) than that of the SAQ pulp (51.0%), as shown in Supporting Information, Table S3, and a similar difference remained after O-stage. The yield decrease directly corresponded to the difference in xylan content between the E-SAQ and SAQ pulps (4.4% units) (Table 1). This indicated that the hemicelluloses that remained in the solid residue after pre-extraction were still susceptible toward extraction and alkaline degradation during the pulping stage. As discussed above, further reduction of the EA charge for pulping could positively contribute to the hemicellulose retention in the pulp.

Comparable cellulose content in both E-SAQ and SAQ pulps (Table 1) revealed that cellulose retention was not altered by the pre-extraction. However, slightly lower intrinsic viscosity of the E-SAQ pulp (Supporting Information, Table S3) indicated that a more severe hydrolytic degradation was associated with the higher alkali concentration in the E-SAQ process steps compared to the reference SAQ cook. Thus, further optimization of the alkali charge most likely also reduces the extent of chain scission by random hydrolysis.

3.6. Papermaking Properties. The reference and pre-extracted SAQ pulps after O-stage were evaluated as potential raw material for papermaking. Silva et al.³¹ demonstrated a positive correlation between beatability and xylan content in eucalyptus pulps. The beating degree values in Supporting Information Table S3 indicate that in the present study both pulps performed similarly in refining. The reduction of the xylan content in the E-SAQ pulp by 7% units (o.d. pulp) in relation to that of the reference SAQ pulp had no effect on pulp beatability. Apparently, the residual xylan content of the E-SAQ pulp was sufficient to maintain fiber beatability on a reasonable level. The overall increment of the beating degree as a function of the number of the PFI mill revolutions was limited by the adjustment of the PFI mill. The comparability of the PFI mill refining results with that of the industrial refining is limited, however, the method is suitable to elucidate the differences between pulps.

Wet and dry zero-span tensile indices were measured for the unbeaten pulps. The results of the wet zero-span test used to eliminate the effect of fiber bonding indicated that the strength of the pre-extracted fibers (0.120 kN·m/g) was inferior to that of the reference fibers (0.141 kN·m/g). However, as was demonstrated by Gurnagul and Page³² the strength of chemically damaged fibers may decrease upon rewetting and is not related to the strength of dry fibers. Indeed, the dry zero-span test revealed no difference between the tensile strength of the SAQ and E-SAQ fibers (Supporting Information, Table S3). However, in the case of the dry zero-span test, the effect of fiber bonding cannot be excluded.

According to Pulkkinen et al.³³ the apparent sheet density is mainly governed by interfiber bonding ability. Here, E-SAQ samples exhibited only slightly lower density than the SAQ samples. Similarly to the apparent density, slightly lower tensile strength was observed at all refining levels of the E-SAQ pulp. Tear strength of the E-SAQ pulp, on the other hand, was clearly superior to that of the reference. At the studied bonding levels, neither the tensile strength nor the tear strength can be correlated with the single fiber strength. Instead, the strength of fiber bonding is determinant, meaning that the fibers get pulled out of the fiber network rather than break at the applied tensile stress. Yet tensile and tear performance are affected differently by pre-extraction. Lower hemicellulose content, milder pulping intensity, and modification of the fiber wall structure are among the factors that collectively and discretely affect fiber interaction in the E-SAQ pulp network. One of the reasons for the enhanced tear strength of the E-SAQ pulp is likely due to its lower yield—the larger number of fibers in the test sample compared to that of the reference pulp at the same basis weight.

A number of studies performed on hardwoods using different pre-extraction and pulping techniques demonstrated results similar to the ones discussed in this work. Al-Dajani and Tschirner^{4,34} showed that alkaline pre-extraction of aspen combined with kraft, alkaline sulphite anthraquinone and alkaline sulphite anthraquinone methanol pulping also resulted in pulps with slightly lower tensile index and improved tear index compared to the corresponding reference pulps without pre-extraction. Pulp produced by alkaline pre-extraction of eucalyptus followed by SAQ pulping had an increased tensile index and similar tear index and zero-span index.⁵ Yoon, Tunc, and van Heiningen⁶ demonstrated that with near-neutral extraction with green liquor followed by kraft pulping of southern mixed hardwoods, pulps of comparable tensile index and improved tear vs tensile indices compared to those of the reference pulp could be obtained.

In papermaking, in addition to the strength, other pulp properties may be of importance. Surface charge is one such property responsible for the interaction with papermaking additives. Sjöström³⁵ confirmed that the charge of the wood pulp fibers mostly originates from uronic acid substituents of xylan. Therefore, the effect of the residual xylan content on the surface charge of pre-extraction pulps should be assessed in detail.

3.7. Commercial Potential of the Pre-extraction Pulping Process. Industrial feasibility of a modified pulping process is primarily driven by the production efficiency and the quality of the principal product as well as by the added value of the newly introduced products. Pulps with excellent papermaking properties could be produced from hardwood by alkaline pre-extraction followed by SAQ pulping. Yet, a detailed

application study of overall pulp performance in paper production is lacking.

The removal of xylan in an E-stage prior to SAQ cooking resulted in a reduction of the pulp yield after the O-stage by 4.9% units (Supporting Information, Table S3). As previously stated, a further reduction of the EA charge in the SAQ cook could further reduce the yield loss. According to this study, 2.24 t of birch wood is required to produce 1 oven dry tonne (ODT) of E-SAQ pulp (after O-stage). The lower pulp output compared to the reference process would alone result in a loss of the revenue; on the other hand, products derived from the E-lye could compensate for such loss (Table S4 in Supporting Information). At the moment, XOS appear to have the highest added value among the commercial xylan-derived products having a market price several times higher than the pulp price (assumed as 3500 EUR/t compared to the pulp price of 629 EUR/t (Dec. 2013)). According to this study, 145 kg of birch xylan (as anhydroxylose) can be produced along with 1 ODT of pulp. Products derived from the isolated xylan could generate notable revenue provided that a market for the specific product is available. Purified xylan could also be marketed as a paper strength additive; however, the market price would not likely exceed the pulp price. On the other hand, production of a barrier film from functionalized polymeric xylan could be an interesting alternative.

The implementation of a pre-extraction stage affects the material and energy balance of pulp production. First, a share of the organic matter is withdrawn from the process during pre-extraction. Second, after membrane filtration, some organic compounds are returned to the process together with the alkali-containing permeates. In case 1 of E-SAQ pulping where the wood intake is maintained at the same level as before the modification (SAQ), the pulp output is decreased by 10% and the potential of energy production from black liquor is reduced by 18%. Furthermore, the concentration of the dissolved solids in the black liquor is reduced, which will result in additional steam demand for the evaporation. Further, when the recirculation of the membrane filtration permeates is considered, the difference in the potential energy generation between the reference and the E-SAQ process is reduced from 18 to only 5%. On the one hand, such a decrease of the organic load and the generated energy leads to reduced electricity production; on the other hand, it could potentially debottle-neck the recovery boiler and allow increased pulp production.

In case 2, in order to maintain the target pulp output at the reference level the wood intake is increased from 167 to 187 t/h (considering an output of 2000 ODT pulp/day). The organic load of the black liquor still remains 14% lower when compared to the reference level, but the combined organic load of the black liquor and the permeates exceeds the reference level by 3%. In this case, sufficient capacity of both the fiberline and the energy recovery cycle is a prerequisite to maintain the pulp output at the target level.

The permeates generated in the nano- and diafiltration may involve difficulties in handling owing to the large volumes and the low concentrations of organic compounds (2.6 and 0.26 kg/m³ in the N- and D-permeate, respectively). The necessity for evaporating water contained by the permeates penalizes the production of electricity. To utilize the alkalinity, N-permeate containing 75 kg/m³ of NaOH can be recirculated to the pre-extraction stage. The dilute D-permeate could be utilized as washing liquor or alkaline bleaching liquor in O-stage and some bleaching stages.

Additional production costs originate from the chemicals required for the processing of the isolated xylan. The suggested ethanol/isopropyl alcohol xylan precipitation scheme could only be feasible provided that a recovery of the alcohols is performed. In the case of XOS production, the enzyme consumption of 0.65 kg/t xylan is considered (Supporting Information, Table S4).

Implementing the pre-extraction process in an existing pulp mill requires major investments depending on the target marketable products. A possibility to retrofit an existing impregnation vessel for pre-extraction purposes, if available, can be considered due to shorter pulping durations required in the modified process. In addition to an extraction vessel, a membrane filtration setup to isolate and concentrate polymeric xylan and recover alkali for the process recirculation is a principal process unit. Further isolating, purifying, and converting to products makes the process cost intensive; however, valorization of the extracted organic fractions is a prerequisite for cost effectiveness.

4. CONCLUSIONS

Alkaline pre-extraction at 95 °C and 2.5 M NaOH allowed isolating 6.6% of wood as polymeric xylan with a molar mass of 20 kDa. Membrane filtration was efficient in concentrating the polymeric fraction and separating NaOH, while alcohol precipitation was efficient in purifying xylan. This was demonstrated by the low lignin content in the precipitate. Enzymatic hydrolysis of xylan was efficient to produce a uniform XOS mixture with a low content of monomers. E-SAQ pulp demonstrated excellent papermaking properties, but its yield was 4.9% lower than that of the reference. The E-SAQ process proved to have a commercial potential.

■ ASSOCIATED CONTENT

Supporting Information

Tables including the data on the pre-extraction optimization, membrane filtration, properties of the produced pulps and calculations of the economic potential as well as a figure demonstrating the effect of chip size on the pre-extraction. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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Notes

The authors declare no competing financial interest.

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■ REFERENCES

(1) Deutschmann, R.; Dekker, R. F. H. From plant biomass to bio-based chemicals: Latest developments in xylan research. *Biotechnol. Adv.* **2012**, *30* (6), 1627–1640.

(2) Pinto, P. C.; Evtuguin, D. V.; Pascoal, N. C. Structure of hardwood glucuronoxylans: modifications and impact on pulp retention during wood kraft pulping. *Carbohydr. Polym.* **2005**, *60*, 489–497.

(3) van Heiningen, A. Converting a kraft pulp mill into an integrated forest biorefinery. *Pulp Pap—Can.* **2006**, *107*, 38–43.

(4) Al-Dajani, W. W.; Tschirner, U. W. Pre-extraction of hemicelluloses and subsequent kraft pulping. Part I: alkaline extraction. *Tappi J.* **2008**, *7*, 3–8.

(5) Schild, G.; Sixta, H.; Testova, L. Multifunctional alkaline pulping, delignification and hemicellulose extraction. *Cell Chem. Technol.* **2010**, *44* (1–3), 35–45.

(6) Yoon, S.-H.; Tunc, M. S.; van Heiningen, A. Near-neutral pre-extraction of hemicelluloses and subsequent kraft pulping of southern mixed hardwoods. *Tappi J.* **2011**, *10*, 7–15.

(7) Richter, G. A. Some aspects of prehydrolysis pulping. *Tappi* **1956**, *39* (4), 193–210.

(8) Alekhina, M.; Mikkonen, K. S.; Alen, R.; Tenkanen, M.; Sixta, H. Carboxymethylation of alkali extracted xylan for preparation of bio-based packaging films. *Carbohydr. Polym.* **2014**, *100*, 89–96.

(9) Borrega, M.; Sixta, H. Purification of cellulosic pulp by hot water extraction. *Cellulose* **2013**, *20* (6), 2803–2812.

(10) Niemelä, K.; Tamminen, T.; Ohra-Aho, T., Black liquor components as potential raw materials. *Tapssa J.* **2008**, Available online http://www.tappsa.co.za/archive3/Journal_papers/Black_Liquor_Components/black_liquor_components.html.

(11) Muguet, M. C. d. S.; Pedrazzi, C.; Colodette, J. L., Xylan deposition onto eucalypt pulp fibers during oxygen delignification: 11th EWLP, Hamburg, Germany, August 16–19, 2010. *Holzforchung* **2011**, *65*, 605–612.

(12) Leschinsky, M.; Zuckerstaetter, G.; Weber, H. K.; Patt, R.; Sixta, H. Effect of autohydrolysis of *Eucalyptus globulus* wood on lignin structure. Part 2: influence of autohydrolysis intensity. *Holzforchung* **2008**, *62*, 653–658.

(13) Schlesinger, R.; Götzinger, G.; Sixta, H.; Friedl, A.; Harasek, M. Evaluation of alkali resistant nanofiltration membranes for the separation of hemicellulose from concentrated alkaline process liquors. *Desalination* **2006**, *192*, 303–314.

(14) Sixta, H.; Potthast, A.; Krottschek, A. W., Chemical pulping processes. In *Handbook of Pulp*; Sixta, H., Ed.; Wiley-VCH Verlag GmbH & Co. KGaA: Weinheim, Germany, 2006; Vol. 1, pp 109–509.

(15) BfR BfR Removes Anthraquinone from Its List of Recommendations for Food Packaging; Federal Institute for Risk Assessment (BfR): Berlin, 2013.

(16) Rantanen, H.; Virkki, L.; Tuomainen, P.; Kabel, M.; Schols, H.; Tenkanen, M. Preparation of arabinoxybiose from rye xylan using family 10 *Aspergillus aculeatus* endo-1,4- β -D-xylanase. *Carbohydr. Polym.* **2007**, *68* (2), 350–359.

(17) Janson, J. Calculation of polysaccharide composition of wood and pulp. *Pap. Puu* **1970**, *52*, 323–329.

(18) Testova, L.; Chong, S.-L.; Tenkanen, M.; Sixta, H., Autohydrolysis of birch wood: 11th EWLP, Hamburg, Germany, August 16–19, 2010. *Holzforchung* **2011**, *65*, (4), 535–542.

(19) Lehto, J.; Alen, R. Alkaline pre-treatment of hardwood chips prior to delignification. *J. Wood Chem. Technol.* **2013**, *33*, 77–91.

(20) Sjöström, E. *Wood Chemistry: Fundamentals and Applications*, 2nd ed.; Academic Press: San Diego, 1993; p 293.

(21) Dudkin, M. S.; Gromov, V. S.; Vedernikov, N. A.; Katkevich, R. G.; Chernov, N. K. *Hemicelluloses*; Zinatne: Riga, 1991; p 488.

(22) Brauns, F. E. *Chemistry of Lignin*. Dekker: New York, **1952**; Vol. 1, pp 409–539.

(23) Willför, S.; Pranovich, A.; Tamminen, T.; Puls, J.; Laine, C.; Suurnäkki, A.; Saake, B.; Uotila, K.; Simolin, H.; Hemming, J.; Holmbom, B. Carbohydrate analysis of plant materials with uronic acid-containing polysaccharides—A comparison between different hydrolysis and subsequent chromatographic analytical techniques. *Ind. Crops Prod.* **2009**, *29*, 571–580.

(24) Teleman, A.; Harjunpää, V.; Tenkanen, M.; Buchert, J.; Hausalo, T.; Drakenberg, T.; Vuorinen, T. Characterization of 4-deoxy- β -L-

threo-hex-4-enopyranosyluronic acid attached to xylan in pine kraft pulp and pulping liquor by carbon-13 and proton NMR spectrometry. *Carbohydr. Res.* **1995**, *272*, 55–71.

(25) Puchart, V.; Biely, P. Simultaneous production of endo- β -1,4-xylanase and branched xylooligosaccharides by *Thermomyces lanuginosus*. *J. Biotechnol.* **2008**, *137*, 34–43.

(26) Chong, S.-L.; Koutaniemi, S.; Virkki, L.; Pynnönen, H.; Tuomainen, P.; Tenkanen, M. Quantitation of 4-O-methylglucuronic acid from plant cell walls. *Carbohydr. Polym.* **2013**, *91*, 626–630.

(27) Falck, P.; Precha-Atsawan, S.; Grey, C.; Immerzeel, P.; Staalbrand, H.; Adlercreutz, P.; Nordberg, K. E. Xylooligosaccharides from Hardwood and Cereal Xylans Produced by a Thermostable Xylanase as Carbon Sources for *Lactobacillus brevis* and *Bifidobacterium adolescentis*. *J. Agric. Food Chem.* **2013**, *61*, 7333–7340.

(28) Metsämuuronen, S.; Lyytikäinen, K.; Backfolk, K.; Siren, H. Determination of xylo-oligosaccharides in enzymatically hydrolysed pulp by liquid chromatography and capillary electrophoresis. *Cellulose* **2013**, *20*, 1121–1133.

(29) Rydholm, S. A. *Pulping Processes*; John Wiley & Sons, Inc.: London, 1964; p 1300.

(30) Pu, Q.; Sarkanen, K. Donnan equilibria in wood-alkali interactions. Part II. Effect of polysaccharide ionization at high alkalinities. *J. Wood Chem. Technol.* **1991**, *11* (1), 1–22.

(31) Silva, T. C. F.; Colodette, J. L.; Lucia, L. A.; Oliveira, R. C. d.; Oliveira, F. v. N.; Silva, L. H. M. Adsorption of chemically modified xylans on eucalyptus pulp and its effect on the pulp physical properties. *Ind. Eng. Chem. Res.* **2010**, *50* (2), 1138–1145.

(32) Gurnagul, N.; Page, D. H. The difference between dry and rewetted zero-span tensile strength of paper. *Tappi J.* **1989**, *72*, 164–7.

(33) Pulkkinen, I.; Fiskari, J.; Alopaeus, V. New model for predicting tensile strength and density of eucalyptus handsheets based on an activation parameter calculated from fiber distribution characteristics. *Holzforschung* **2010**, *64*, 201.

(34) Al-Dajani, W. W.; Tschirner, U. W. Pre-extraction of hemicelluloses and subsequent ASA and ASAM pulping: comparison of autohydrolysis and alkaline extraction. *Holzforschung* **2010**, *64*, 411–416.

(35) Sjöström, E. The origin of charge on cellulosic fibers. *Nord. Pulp Pap. Res. J.* **1989**, *4*, 90–3.