Jari Heinonen[1,*]
Juha Tamper[2]
Markku Laatikainen[1]
Tuomo Sainio[1]

Chromatographic Recovery of Monosaccharides and Lignin from Lignocellulosic Hydrolysates

The chromatographic recovery of monosaccharides and lignin from lignocellulosic hydrolysates was studied at laboratory and pilot scale. A weak cation-exchange resin in sodium form and a water eluent gave good separation efficiency. Scale-dependent phenomena, especially viscous fingering resulting from the large viscosity and density differences between the hydrolysate feed and eluent, were observed. The issue was resolved in the pilot scale with appropriate selection of the flow direction, and a high productivity was achieved at 95 % recovery yield. The pH value of the feed was found to have no effect on the actual separation; however, the resin was significantly less colored at a higher pH value.

Keywords: Chromatographic separation, Lignin, Lignocellulosic hydrolysate, Monosaccharides, Weak cation-exchange resin

Received: August 17, 2018; *revised:* September 24, 2018; *accepted:* September 27, 2018

DOI: 10.1002/ceat.201800412

1 Introduction

A wide range of platform chemicals, such as monosaccharides, lignin, aliphatic carboxylic acids (mainly formic acid and acetic acid), and furans (mainly furfural and 5-hydroxymethylfurfural HMF) can be produced from lignocellulosic biomasses. The monosaccharides can be further upgraded, e.g., to alcohols, carboxylic acids, amino acids, aldehydes, ketones, esters, ethers, polymers, alkanes, and alkenes [1–5]. Lignin can be used as a raw material for aromatic fine chemicals, dispersing agents, aromatic polymers, activated carbon, carbon fibers, composites, polyphenolic glues, biofuels, and additives [6, 7]. Furan derivatives, such as furfuryl alcohol (a precursor of levulinic acid) and 2,5-furandicarboxylic acid, can be produced from furfural and HMF [7].

Monosaccharides can be produced from lignocellulosic biomass by hydrolysis during which the polysaccharides, i.e., cellulose and hemicelluloses, in the biomass are cleaved into monosaccharides with mineral acids, e.g., sulfuric acid, or enzymes as the catalysts [8–12]. During acidic hydrolysis, a number of other components are also formed or dissolved from the biomass, and end up to a monosaccharide-rich solution. Acetic acid is formed because the acetyl groups in hemicellulose chains are cut off [10, 11]. Furans and formic acid are formed as result of monosaccharide degradation [10, 13, 14]. In addition, lignin in the biomass is partly dissolved when dilute acid is used as catalyst. Prior to the downstream processing of the monosaccharides, these byproducts must be removed because they are known to act as inhibitors in monosaccharide fermentation or to otherwise hinder the processing [10, 15–17]. The recovery of these byproducts as pure components would be beneficial because of their value as platform chemicals.

In the case of hydrolysates with low lignin concentrations, e.g., concentrated acid hydrolysates, the separation of the monosaccharides from the carboxylic acids and acid catalyst can be done by ion exclusion chromatography (IEC) [18, 19]. Separation of the furans is also achieved, albeit not efficiently because they are strongly adsorbed on the separation material, i.e., a strong cation-exchange resin [18, 20]. Efficient separation of the furans from such hydrolysates can be done by adsorption onto polymeric adsorbents [15, 21–24].

With the hydrolysates produced via dilute acid and enzymatic hydrolysis, the separation of lignin from the monosaccharides is also essential [11, 25]. A number of methods have been proposed for the removal of lignin from aqueous solutions. For example, the separation can be accomplished with ionic liquids [26, 27], by membrane filtration [28–30], or by adsorption [25, 31–38]. Of the studied methods, adsorption has received the most attention [25, 31–38]. Polymeric adsorbents are an efficient option instead of activated carbon because the adsorbed lignin can be easily desorbed from the adsorbent. Recently, an adsorption-based method for lignin recovery from lignocellulosic hydrolysates

[1]Dr. Jari Heinonen, Dr. Markku Laatikainen, Dr. Tuomo Sainio
jari.heinonen@lut.fi
Lappeenranta University of Technology, School of Engineering Science, Skinnarilankatu 34, 53850 Lappeenranta, Finland.
[2]Dr. Juha Tamper
UPM-Kymmene Corporation, North European Research Center, Biochemicals, Paloasemantie 19, 53200 Lappeenranta, Finland.

was presented [25]: 80 % lignin could be recovered, with a 95 % monosaccharide yield. However, the other components in the hydrolysates, namely, formic acid, acetic acid, and furans, could not be separated from the monosaccharides [25]. Thus, an additional step is required to purify the monosaccharides. In addition, the desorption of lignin from the adsorbent requires 50 wt % aqueous ethanol solution, and this increases the capital and operating costs [25]. The separation of carboxylic acids and furans from the monosaccharides could be done by adsorption onto activated carbon, but the desorption of these compounds requires large amounts of ethanol [22].

Here, an efficient chromatographic method for the recovery of monosaccharides from lignocellulosic hydrolysates produced under slightly acidic conditions is presented. With the proposed method, the monosaccharides are effectively recovered through the use of water alone as the eluent in the separation. No organic solvents are needed in this process on the contrary to adsorption-based processes. The study compares appropriate separation materials and examines the factors affecting separation efficiency at both laboratory scale and pilot scale. It also includes a discussion on the production of all of the main components in these hydrolysates, i.e., monosaccharides, lignin, carboxylic acids, and furans, as pure components or pure mixtures of similar components.

2 Experimental Methods

2.1 Chemicals and Separation Materials

Ultrapure water produced with CENTRA R 60/120 (ELGA LabWater) and sodium hydroxide (≥ 99.0 %, pellets for analysis, Merck KGaA) were used in the experiments. Blue Dextran 2000 (GE Healthcare) and sodium chloride (99.5 % AnalaR Normapus ACS, VWR Prolabo) were employed in the resin bed porosity measurements.

Two gel-type acrylate-based weak acid cation-exchange (WAC) resins, CA10GC (acrylate–DVB matrix; 5.0 wt % DVB; d_p = 300 µm) and CA16GC (8.0 wt % DVB; d_p = 300 µm) in Na^+ form (both from Finex/Johnson Matthey), served as separation materials. CA16GC resin with d_p = 250 µm was also applied in the separation experiments.

2.2 Lignocellulosic Hydrolysates

Two authentic lignocellulosic hydrolysates were selected as feed solutions (Tab. 1). It has been established that WAC resins are highly selective towards protons (H^+). To avoid the exchange of the ionic form of the resins to H^+ during the experiments, the pH values of the originally slightly acidic hydrolysates were adjusted to 6 or 10 with 50 wt % NaOH solution. Afterwards, the hydrolysates were filtered with a cord filter to remove possible remaining lignin precipitates.

The WAC resins used here are highly selective towards di- and trivalent cations [39]. This leads to the exchange of sodium (Na^+) ions with the di- and trivalent cations. To prevent this, an ion-exchange column should be placed in before the chromatographic step in order to remove the di- and trivalent cations from the hydrolysate. WAC resins can be used as ion exchangers because of their high selectivity towards di- and trivalent cations.

2.3 Chromatographic Separation

Chromatographic separation experiments were conducted at laboratory scale (V_{bed} = 120 mL) and pilot scale (V_{bed} = 11 L) at 50 °C (temperature limit of the columns) with purified water as the eluent. A high temperature was used in the experiments to minimize the viscosity of the highly concentrated feed solu-

Table 1. Compositions (after pH adjustment) of the hydrolysates used in this work.

	Hydrolysate 1		Hydrolysate 2 (pH 6)		Hydrolysate 2 (pH 10)[c)]	
Component	C [g L^{-1}]	x [wt %][b)]	C [g L^{-1}]	x [wt %][b)]	C [g L^{-1}]	x [wt %][b)]
Lignin	18.2	3.9	31.6	8.1	n.a.	–
Monosaccharides[a)]	438.1	93.9	343.6	87.8	361.2	–
Formic acid	1.6	0.3	1.1	0.3	1.7	–
Acetic acid	7.8	1.7	13.6	3.5	15.0	–
Furfural	0.2	0.0	0.2	0.1	0.04	–
HMF	0.7	0.2	1.4	0.4	0.04	–
Ca^{2+}	0.4	–	0.9	–	n.a.	–
Mg^{2+}	0.1	–	0.23	–	n.a.	–
Cu^{2+}	2.3×10^{-4}	–	5.0×10^{-4}	–	n.a.	–
Fe^{3+}	1.0×10^{-2}	–	2.6×10^{-2}	–	n.a.	–

[a)] Treated as one pseudo component; [b)]based on the concentrations of the analyzed organic components; [c)]after a few days in storage, solution pH was ~ 8.

tions. A decrease in viscosity results in higher mass transfer rates and a reduction in dispersion.

The laboratory-scale batch chromatography unit consisted of two two-piston pumps (for eluent and feed streams) attached to a glass column (d_{bed} = 1.5 cm, h_{bed} = 70 cm) with a water heating jacket via a fluid degasser and an injection valve. The column outlet stream was monitored with an online conductivity detector, a refractive index (RI) detector, and an ultraviolet (UV) detector. A fraction collector was also connected to the column outlet. Valve control and data collection were performed with LabVIEW software (National Instruments).

The pilot-scale unit consisted of two magnetic drive gear pumps connected through three mass flow meters and an injection manifold to a chromatographic column with a water heating jacket. The column outlet stream was monitored with an online conductivity detector, an RI detector, a UV detector, and a pH detector. Fractions were also collected from the column outlet. Unit control and data collection were performed with the LabVIEW software.

Duplicate experiments were conducted to address the reproducibility of the experiments. However, because of the similarities among the online signals measured at the column outlet, the fractions that were analyzed in detail were collected from just one experiment.

2.4 Bed Porosity

The resin bed porosities were determined from the retention times of the small Blue Dextran 2000 (1.5 g L^{-1}) and NaCl pulses (1 mol L^{-1}). With the laboratory scale unit, the feed volume V^{F} [1)] was 0.5 mL, and the flow rate Q was 1.0 mL min^{-1}. With the pilot scale unit, V^{F} was 0.004 BV, and Q was 0.9 BV h^{-1}. The bed porosities for the CA10GC resin were approximately 0.32–0.33 and for the CA16GC approximately 0.35–0.36.

2.5 Analyses

The lignin concentrations were determined from the UV absorbance in accordance with a standardized method described in TAPPI Useful Methods (UM) 250 (acid-soluble lignin in wood and pulp) [40]. A wavelength of 205 nm was used for quantification [40, 41]. Turbid samples were filtered with a syringe filter (0.45 µm polypropene filter; VWR), and the UV absorbance was measured with an Agilent 8453 spectrophotometer. The lignin concentration was calculated from the absorbance value of a sample by using the extinction coefficient 0.11 L $mg^{-1}cm^{-1}$ (average value for samples containing different wood species) [42].

The concentrations of monosaccharides, formic acid, acetic acid, HMF, and furfural were analyzed with an off-line high-performance liquid chromatography (HPLC) (HP/Agilent 1100) instrument equipped with a refractive index detector and a UV detector (variable wavelength detector; wavelength 280 nm). An organic acid analysis column, MetaCarb 87H (Varian/Agilent), was employed. The column temperature was 65 °C, V^{F} was 5 µL, and 0.005 mol L^{-1} sulfuric acid served as an eluent with flow rate gradients of 0.6–1.0 mLmin^{-1}.

The concentrations of calcium and other metal ions in the hydrolysates were determined with inductively coupled plasma–optical emission spectrometry (ICP-OES) (Thermo Scientific iCAP Duo 7600 Simultaneous ICP-OES analyzer). The samples were prepared according to a standardized method (ISO 11885).

3 Calculations

The separation efficiency was evaluated with the recovery yield and the productivity and water consumption. Recovery yields Y_i (%) of the monosaccharides, lignin, HMF, and organic acids were calculated from [43]:

$$Y_i = \frac{m_i^{out}}{m_i^{feed}} \times 100\% \quad (1)$$

where m_i is the mass of component i and superscripts out and feed stand for target fraction and feed, respectively.

The efficiency of the separation process with respect to cycle time and volume for the resin bed was given as monosaccharide productivity Pr_{ms} (kg m^{-3}(bed) h^{-1}) calculated as [43]:

$$Pr_{ms} = \frac{m_{ms}^{out}}{V_{bed}\, t_{cycle}} \quad (2)$$

where V_{bed} is the resin bed volume and t_{cycle} is the cycle time. The amount of water needed for the production of 1 kg monosaccharides EC_{ms} (m^3kg^{-1}) was calculated by Eq. (3) [43]:

$$EC_{ms} = \frac{Qt_{cycle} - V^{F}}{m_{ms}^{out}} \quad (3)$$

where Q is the flow rate and V^{F} is the feed volume.

4 Results and Discussion

4.1 Selection of Separation Material

Both WAC resins were found to be suitable for this separation task. The resins were compared by varying Q and V^{F}. The typical elution profiles are presented in Fig. 1.

The elution order of the main compounds in the hydrolysates was lignin, Na-salts of formic acid and acetic acid, furans, and monosaccharides (Fig. 1). Lignin is a group of large strongly hydrophobic three-dimensional heterogenic aromatic polymers; thus, lignin elutes first. The furans are also hydrophobic molecules; consequently, they co-elute with lignin. At pH 6, acetic acid and formic acid are mostly in the form of Na-salts and are thus dissociated to a great extent. They co-elute with lignin because of (partial) electrostatic exclusion from the resin. Monosaccharides are highly hydrophilic compounds. In addition, they have some level of interaction with the Na^+ ions

1) List of symbols at the end of the paper.

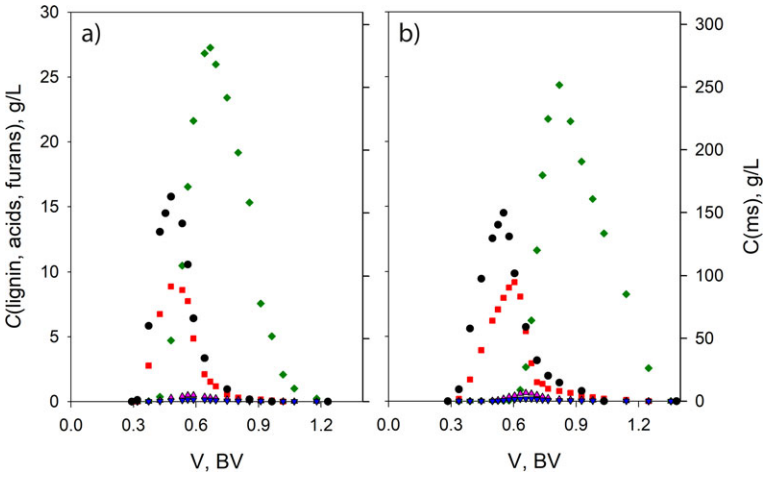

Figure 1. Typical elution profiles obtained in chromatographic recovery of lignocellulose-based monosaccharides with WAC resins in Na^+ form with a laboratory-scale column (V_{bed} ≈ 120 mL). (a) CA16GC (bed porosity = 0.35); (b) CA10GC (bed porosity = 0.32). Experimental conditions: feed = hydrolysate 1; top-down flow; Q = 2.0 BV h^{-1}; V^F = 0.3 BV; T = 50 °C. Symbols: ◇ = monosaccharides; ○ = lignin; □ = carboxylic acids (formic acid and acetic acid); △ = HMF; ▽ = furfural.

of the WAC resins (ligand exchange [44]) and are therefore most strongly sorbed.

Strong tailing of the lignin and organic acids was observed (Fig. 1) which was a result of the high-viscosity region in which the monosaccharides eluted. High viscosity slows down mass transfer and causes tailing of the profiles. The furan profiles were strongly dispersed with both resins and eluted under the lignin and the monosaccharide profiles. This was also a result of the slow mass transfer caused by the high viscosity. Based on the elution profiles obtained, the hydrolysate can be fractionated into two product fractions: the first containing lignin, carboxylic acids, and furans, and the second containing the monosaccharides.

The degree of resin cross-linking had a clear effect on the elution profiles (Fig. 1). With the less cross-linked CA10GC resin (Fig. 1 b), the elution profiles were slightly more dispersed than with the CA16GC (Fig. 1 a). This was attributed to the stronger shrinking of the CA10GC resin because of the high monosaccharide concentration in the feed. From the examples presented in Fig. 1, during the experiment the CA16GC resin shrank approximately 2.6 % and the CA10GC approximately 4.3 %. The shrinking increases dispersion because a void space is formed on the top of the resin bed between the bed and the upper adapter. Although shrinking occurs, it should be noted that it is reversible, and the void space disappears when no hydrolysate is inside the column.

Although the dispersion resulting from the resin shrinkage was slightly stronger with the CA10GC than with the CA16GC (Fig. 1), the separation efficiency was better with the CA10GC (Tab. 2). This also came from the lower degree of cross-linking. The CA10GC swelled more than the CA16GC; thus, the pore size of the resin was larger which affected size exclusion. With a resin that is less cross-linked, components can enter the pores of the resin more freely. Although this influences the sorption of all components to some extent, it has the most profound effect on the monosaccharides, thus enhancing the separation.

To compare the WAC resins, a number of separation runs with varying operating parameters (V^F = 0.05–0.20 BV; Q = 1–3 BV h^{-1}) were performed with the laboratory-scale equipment (Tab. 2). The rather low flow rate range used was justified by the high viscosity because of the high feed concentration. The large viscosity differences between the feed and eluent, combined with the high flow rates, lead most likely to viscous fingering, which has a detrimental effect on separation. The column loading range was chosen on the basis of the preliminary experiments. The CA10GC resin yielded higher recovery yields than the CA16GC because of the better separation. However, the Pr_{ms} values obtained with the CA10GC resin were lower than those obtained with the CA16GC because of the longer cycle time resulting from the more dispersed component profiles and the larger differences in the sorption strength of the main components (Fig. 1).

The resins were also compared on pilot scale (Fig. 2). Although the laboratory-scale experiments suggested that the CA10GC resin was more efficient for the separation task in question, the situation was opposite with the pilot-scale unit. With the CA10GC, the elution profiles were considerably more dispersed than those obtained at laboratory scale (Fig. 1 b vs. Fig. 2 b). It is noteworthy that the bed heights, i.e., 70 cm and 72 cm at the laboratory and pilot scales, respectively, and the linear flow rates were approximately the same in both columns. Thus, the column efficiency, i.e., number of theoretical plates

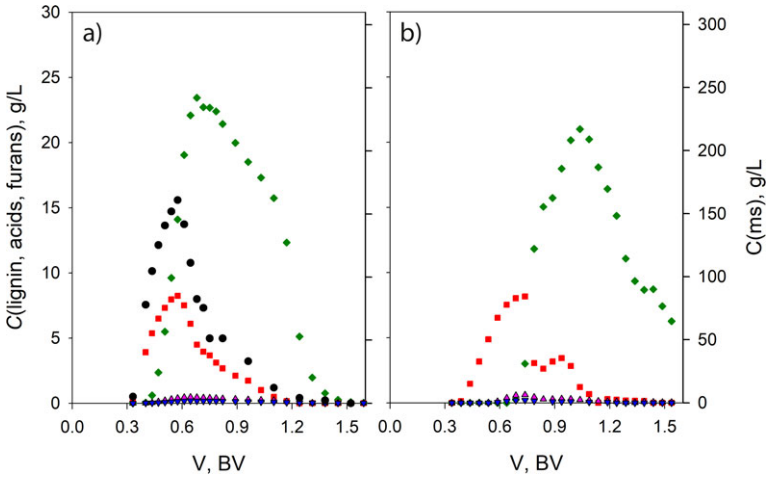

Figure 2. Typical elution profiles obtained in the chromatographic recovery of lignocellulose-based monosaccharides with WAC resins in Na^+ form with a pilot-scale column (V_{bed} ≈ 11 L). (a) CA16GC; (b) CA10GC. Experimental conditions: feed = hydrolysate 1; top-down flow; Q = 2.0 BV h^{-1}; V^F = 0.3 BV; T = 50 °C. Symbols: see caption for Fig. 1. Lignin profile for CA10GC resin is not shown.

Table 2. Separation efficiency of CA16GC and CA10GC resins in the recovery of monosaccharides from lignocellulosic hydrolysate (hydrolysate 1) at the laboratory scale. Fractionation done with single cut with target Y_{ms} = 95 %.

Q [BV h^{-1}]	V^F [BV]	Y_{ms} [%]	Y_{lignin} [%]	Y_{acids} [%][a)]	Y_{HMF} [%]	$Y_{furfural}$ [%]	Pr_{ms} [kg m^{-3}(bed) h^{-1}]	EC_{ms} [L kg^{-1}]
CA16GC								
1.0	0.05	95.1	94.6	104.3	40.2	30.1	38.3	23.9
1.0	0.10	95.1	90.4	97.4	41.1	31.3	64.5	13.2
1.0	0.20	95.0	77.6	80.1	33.5	26.1	113.6	6.5
2.0	0.05	95.0	87.1	91.2	29.5	21.0	61.0	30.8
2.0	0.10	95.0	80.6	88.9	27.5	19.3	108.9	16.3
2.0	0.20	95.1	55.5	57.6	21.0	16.5	193.5	8.1
3.0	0.05	95.0	80.5	86.5	23.8	15.3	91.2	30.8
3.0	0.10	95.1	70.5	73.9	21.7	16.9	157.2	16.8
3.0	0.20	95.1	57.0	58.8	19.7	16.4	264.9	9.0
CA10GC								
1.0	0.05	95.1	95.7	110.2	88.9	89.7	30.0	31.4
1.0	0.10	95.0	98.7	107.0	82.8	83.8	53.9	16.4
1.0	0.20	95.0	92.9	105.4	72.0	73.4	88.4	9.2
2.0	0.05	95.0	100.3	102.4	70.1	69.4	54.5	35.2
2.0	0.10	95.0	94.3	94.1	63.3	64.6	80.9	22.9
2.0	0.20	95.1	87.2	97.4	57.3	57.7	149.4	11.0
3.0	0.05	95.1	91.1	107.9	62.6	64.7	73.0	39.0
3.0	0.10	95.0	89.2	100.5	58.0	58.6	137.3	19.7
3.0	0.20	95.0	84.1	86.4	47.1	47.4	236.1	10.6

[a)] Values larger than 100 % are the result of analytical inaccuracies.

(NTP), should have been approximately the same. The differences can be explained by the diameters of the columns being 1.5 vs. 14 cm, respectively.

With negligible wall support in the larger column for the packing, a considerably larger void space was formed between the CA10GC resin bed and the upper column adapter because of the resin volume changes during the separation run. This outcome, combined with the large viscosity and density differences, resulted in the dispersed elution profiles (Fig. 2 b). The occurrence of this phenomenon renders the CA10GC resin unsuitable for the separation, at least with the industrially relevant feed concentrations used in this study.

The effects of resin shrinkage and differences in viscosity and density on the separation efficiency were considerably smaller with the CA16GC than with the CA10GC (Fig. 2). Nevertheless, the profiles were more strongly dispersed than they were at the laboratory scale (Fig. 1 a).

The separation efficiency for the CA10GC resin was not evaluated in the example case shown in Fig. 2 b because of the incomplete elution of the monosaccharides in a reasonable time. It is clear from the pilot-scale experiments that the CA10GC resin was less efficient than the CA16GC.

The performance indicator values for the CA16GC in the case shown in Fig. 2 a were with the monosaccharide recovery yield Y_{ms} target of 95.0 %: Y_{lignin} = 42.6 %, Y_{acids} = 42.5 %, Y_{HMF} = 12.2 %, Pr_{ms} = 190 kg m^{-3}(bed) h^{-1}, and EC_{ms} = 8.3 L kg^{-1}. With Q = 2 BV h^{-1} and V^F = 0.2 BV (target Y_{ms} = 95 %; lignin was not analyzed), Y_{acids} = 66.1 %, Y_{HMF} = 31.2 %, Pr_{ms} = 159 kg m^{-3}(bed) h^{-1}, and EC_{ms} = 10.3 L kg^{-1}. Due to more strongly dispersed elution profiles, the values obtained at pilot scale are lower than those obtained with the laboratory-scale unit under the same conditions (Tab. 2).

WAC resins are used in the sweetener industry in the de-ashing (removal of divalent cations) of sugar solutions [39, 45, 46]. Thus, it is expected that the ionic form of the selected separation materials eventually changes from Na^+ to Ca^{2+} or other divalent cations in the hydrolysates (see Tab. 1). This was not observed in this study because no long cyclic runs were done. Such a change in the ionic form of the resin can be avoided by removing the divalent cations from the hydrolysate beforehand. This can be done in a separate column placed in before the chromatographic column. A WAC resin in Na^+ form can also be used as the separation material in such ion-exchange step [39, 45, 46].

Because of the obtained results, the CA16GC was applied in Na^+ form as the separation material in subsequent studies. It should be noted that if a highly viscous feed needs to be fractionated chromatographically, the results obtained at laboratory scale might be very different from those achieved with a larger (diameter) column even if the bed height is the same. When the feed viscosity is low, such an effect would probably not be observed, and the scale-up could be done easily.

4.2 Effect of Process Variables on Separation Efficiency with CA16GC Resin

The effect of the particle size of the separation material, flow direction, and feed pH on the separation efficiency was investigated at laboratory and pilot scales. The influence of particle size (250 vs. 300 μm) on separation efficiency was studied with hydrolysate 2 (see Tab. 1) as the feed solution. The typical elution profiles are displayed in Fig. 3. No differences in the elution profiles were observed between the 250-μm and 300-μm particles (Fig. 3). The results were similar at both the laboratory scale (Figs. 3 a, b) and the pilot scale (Fig. 3 c).

As can be seen in Figs. 1–3, the monosaccharide profiles were dispersed, and the lignin and organic acid profiles exhibited long tailing because of the large viscosity and density differences between the feed and the eluent resulting in viscous fingering [47, 48]. Minimization of the viscous fingering was attempted by changing the flow direction (Fig. 4).

At laboratory scale, no significant differences could be observed between the flow directions (Fig. 4 a). In contrast, distinct differences were observed at the pilot scale (Fig. 4 b). The profiles obtained by the top-bottom flow were notably distorted. The organic acid profile had a shoulder under the monosaccharide profile, and the monosaccharide profile had two local maxima. Lignin was not analyzed in the pilot-scale experiments, but the lignin profile would have been distorted in a manner similar to that of the organic acid profile.

Changing the flow direction in the pilot-scale column to bottom-top significantly decreased the distortion of the profiles (Fig. 4 b). This could be attributed to the minimization of the viscous fingering. Given that at the pilot scale, the flow direction had a clear effect on the elution profiles, a quantitative evaluation was conducted (Tab. 3 for the example presented in Fig. 4 b). The change in the flow direction to bottom-top clearly increased the separation efficiency.

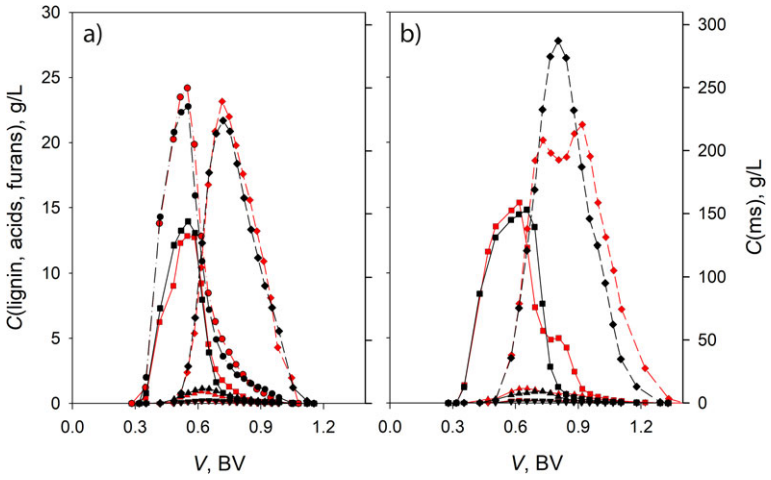

Figure 4. Effect of flow direction on the elution of the main components in lignocellulosic hydrolysates with laboratory-scale and pilot-scale units. (A) Laboratory scale, V^F = 0.2 BV, Q = 2 BV h^{-1}; (B) pilot scale, V^F = 0.2 BV, Q = 2 BVh^{-1}. Experimental conditions: feed solution = hydrolysate 2; T = 50 °C. Symbols: red = top-bottom flow; black = bottom-top flow; for other symbols, see caption for Fig. 1. Lines are presented to guide the eye.

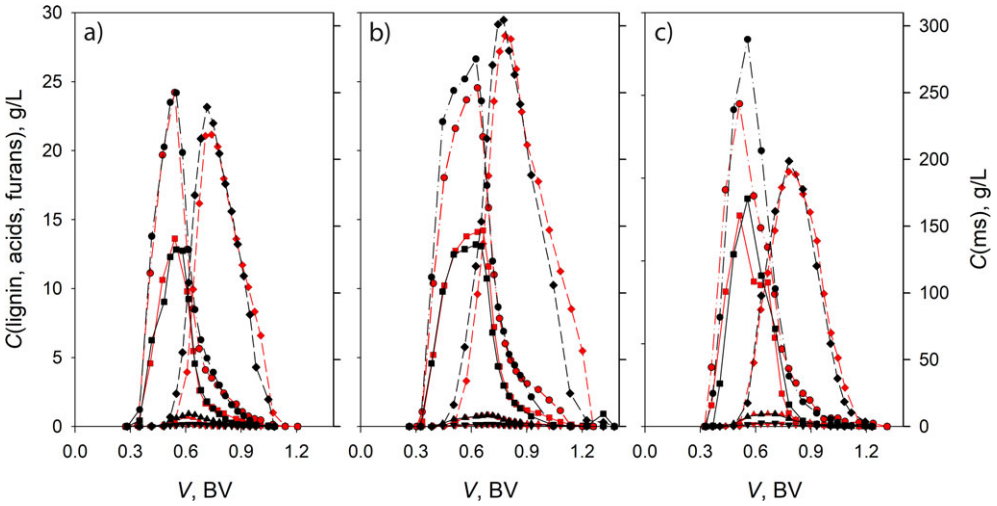

Figure 3. Effect of particle size on the elution profiles with laboratory-scale and pilot-scale units. (A) Laboratory scale, V^F = 0.2 BV, Q = 2 BV h^{-1}; (B) laboratory scale, V^F = 0.3 BV, Q = 3 BV h^{-1}; (C) pilot scale, V^F = 0.2 BV, Q = 3 BV h^{-1}. Experimental conditions: feed = hydrolysate 2; top-down flow; T = 50 °C. Colors: red = 250 μm; black = 300 μm. Symbols: see caption for Fig. 1. Lines are presented to guide the eye.

Table 3. Effect of flow direction on the separation efficiency for the separation experiment shown in Fig. 4 b. Target Y_{ms} = 95 %; Y_{lignin} was not evaluated.

Flow direction	Top-bottom	Bottom-top	$\triangle i$ [%]
Y_{acids} [%]	66.1	67.5	2.1
Y_{HMF} [%]	31.2	33.1	6.0
$Y_{furfural}$ [%]	27.3	31.5	15.3
Pr_{ms} [kg m^{-3}(bed) h)$^{-1}$]	159	171	7.5
EC_{ms} [m^{3}kg^{-1}]	10.3	8.4	−18.4

It can be concluded that the effect of the flow direction on the separation should be investigated if highly viscous feed solutions are treated chromatographically. This investigation should be done with pilot-scale equipment because the viscous fingering might not be visible in separation experiments done with a laboratory-scale column that has a small column diameter.

The pH values of the hydrolysates were adjusted to 6.0 prior to the experiments to minimize the exchange of the Na^+ counterions in the resin with the H^+ ions in the hydrolysate. With the hydrolysate 2, lignin precipitates still formed after the pH adjustment and filtration during storage at 5 °C. To avoid this, a pH value higher than 6.0 should be used. In this study, the effects of pH adjustment to 10 on lignin precipitation and separation efficiency were investigated.

With the increase of the feed pH to 10, the dissociation and solubility of the lignin increased. This resulted in the disappearance of the precipitation problem. As can be seen from the titration curve in Fig. 5 a, the dissociation of the phenolic groups of lignin started at approximately pH 6 and was complete at pH 10. The consumption of NaOH increased by approximately 25 % when compared with the adjustment of the pH to 6. The pH values indicated in Fig. 5 a were taken after a short equilibration period; however, it was observed that the values tended to decrease with time.

The increase of the pH value from 6 to 10 resulted in the effective disappearance of HMF and furfural from the hydrolysate (Tab. 1). The reduction was 97 % for HMF and 83 % for furfural. This could probably be attributed to the condensation of the furans with the phenolic units of lignin under basic conditions. Such reactions are used in the production of phenol-furfural resins and aerogels [49, 50].

The impact of pH adjustment on separation efficiency was also evaluated (Fig. 5 b). In terms of the acid and sugar profiles, similar behaviors were observed for the two systems. The concentrations of formic and acetic acid were approximately 10 % higher in the pH 10 feed. The color profiles measured at 600 nm also matched quite closely, but the color was somewhat more intense at the higher pH values because of the higher level of lignin ionization. It seems, therefore, that a higher pH value does not provide any benefit regarding separation efficiency. However, the higher feed pH had a clear effect on the adsorption of colored substances onto the resin. After two 0.3-BV pulses, no visible change in the bed color was seen at pH 10. In contrast, the top of the bed became distinctly brown with pH 6 feed.

4.3 Recovery of the Main Components in Hydrolysates as Pure Components

In the previous sections, an efficient chromatographic method for the recovery of monosaccharides from hydrolysates was presented. Although the production of pure monosaccharides was facilitated, no separation between lignin, carboxylic acids, or furans could be achieved. However, a review of the literature provides suggestions for some process options for achieving such separations. Because all of these components are valuable platform chemicals, their recovery as pure components or as mixtures of similar components would be beneficial.

It has been acknowledged that lignin is a strong foulant; thus, it should be recovered from the hydrolysates after the monosaccharides have been recovered. The WAC resin used as the separation material in the monosaccharide recovery step is hydrophilic; consequently, the fouling caused by lignin should be minimal. Therefore, monosaccharide recovery can be performed before lignin recovery. Lignin can be recovered efficiently as a pure product by adsorption. In [25], lignin was found to be strongly sorbed onto acrylate-based XAD-16N adsorbent; however, the other main components of the hydrolysates were only weakly sorbed. Recovery of the adsorbed lignin can be done efficiently with an aqueous ethanol solution (50 wt % ethanol [25]). To avoid the high chemical consumption in the

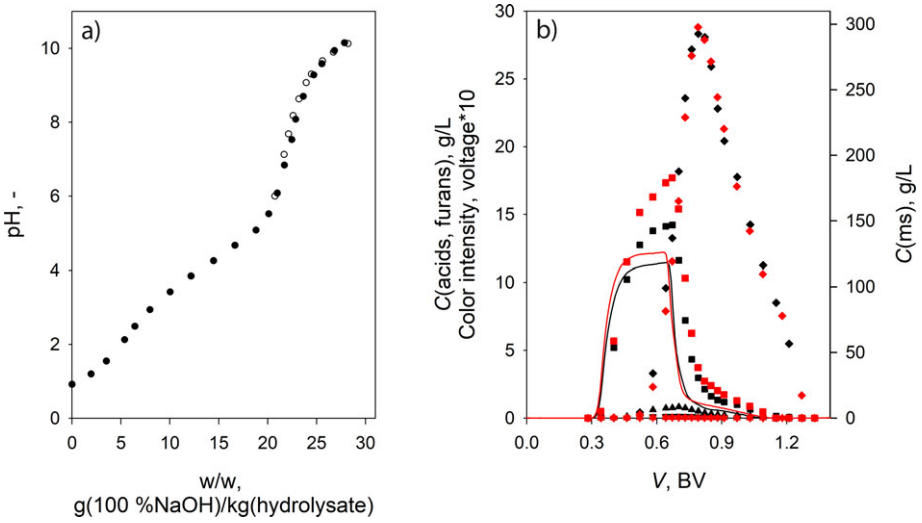

Figure 5. (A) Titration curves of two hydrolysate 2 samples. (B) Effect of hydrolysate pH (hydrolysate 2) on the elution of profiles with a laboratory-scale unit. Experimental details: V^F = 0.3 BV, Q = 3 BV h^{-1}; T = 50 °C. Symbols: black = feed pH 6; red = feed pH 10; solid line = color intensity (λ = 600 nm) as voltage; for other symbols see caption for Fig. 1.

lignin recovery step, the ethanol used in the desorption should be recycled. This could be done by separating the ethanol from lignin by evaporation or membrane filtration [28–30].

After the recovery of monosaccharides and lignin from the hydrolysates, the furans can be separated from the carboxylic acids. This can be done chromatographically with strong cation-exchange resins in H^+ form as the separation material or by adsorption onto a polymeric adsorbent (e.g., [18, 20, 22]). The furans (furfural and 5-HMF) are partly separated from one another by either of these methods and can thus be obtained as pure products [18, 20, 22]. In contrast, the similarities of the carboxylic acids, i.e., formic acid and acetic acid [51], necessitates an additional separation step in their selective separation. The separation of acetic acid from formic acid can be obtained by chromatography [52], reactive distillation (distilled as esters) [53], extraction with aromatic hydrocarbons [54], or extractive distillation [55].

5 Conclusions

An efficient chromatographic method was presented for the recovery of monosaccharides from lignocellulosic hydrolysates. A hydrophilic weak cation-exchange resin in Na^+ form was used as the separation material and water as the eluent. This method enables the production of monosaccharides from hydrolysates with high lignin concentrations in a single step. The column dimensions were observed to have a strong effect on the separation because of the high viscosity differences between feed solution and eluent. These differences caused viscous fingering in the pilot-scale operation, but were absent in the laboratory-scale operation. In a large-scale operation, this can be minimized by using a bottom-top flow direction instead of the more generally used top-bottom flow.

The pH value of the hydrolysate must be adjusted to 6 or higher prior to the start of the separation task to avoid any exchange of the counterions in the resin. A further increase of the pH value to 10 resulted in the disappearance of the furans in the hydrolysate. This can probably be attributed to reactions of the furans with lignin. The pH value of 10 did not affect the separation when compared to the results with pH 6, but the change of the resin color toward brown in the presence of lignin was less evident.

The method presented in this study enabled the effective recovery of monosaccharides as pure products; however, the other main components of the hydrolysates were collected as a mixture. Given that they are also valuable products, their selective recovery should be conducted in additional separation steps that include chromatography and adsorption.

Acknowledgment

The authors are grateful to Ms. Anne Hyrkkänen and Mr. Aleksi Salo for their assistance in conducting the separation experiments. Financial support from the Academy of Finland (grant SA/298548) and the European Union's Horizon 2020 Program (grant 637077) is gratefully acknowledged.

The authors have declared no conflict of interest.

Symbols used

C_i	[g L^{-1}]	concentration of component *i*
d_p	[µm]	average particle diameter
EC_{ms}	[L kg^{-1}]	amount of water needed for the production of 1 kg of monosaccharides
m_i	[kg]	mass of component *i*
Q	[m^{3}h^{-1}]	flow rate
Pr_{ms}	[kg m^{-3}(bed) h^{-1}]	productivity with respect to monosaccharides
t_{cycle}	[h]	cycle time
V_{bed}	[cm^3]	resin bed volume
V^F	[m^3]	feed volume
Y	[%]	recovery yield of component

Subscripts and superscripts

acids	carboxylic acids
feed	in feed pulse
ms	monosaccharides
out	in target fraction

Abbreviations

HMF	hydroxymethylfurfural
WAC	weak cation exchange

References

[1] B. Kamm, M. Kamm, in *White Biotechnology* (Eds: R. Ulber, D. Sell), Springer Science+Business Media, Berlin **2007**, 175–204.

[2] E. L. Kunkes, D. A. Simonetti, R. M. West, J. C. Serrano-Ruiz, C. A. Gärtner, J. A. Dumesic, *Science* **2008**, *322*, 417–421. DOI: https://doi.org/10.1126/science.1159210

[3] L. Luo, E. van der Voet, G. Huppes, *Bioresour. Technol.* **2010**, *101*, 5023–5032. DOI: https://doi.org/10.1016/j.biortech.2009.12.109

[4] J. C. Colmenares, A. Magdziarz, A. Bielejewska, *Bioresour. Technol.* **2011**, *102*, 11254–11257. DOI: https://doi.org/10.1016/j.biortech.2011.09.101

[5] B. N. M. Van Leeuwen, A. M. Van Der Wulp, I. Duijnstee, A. J. A. Van Maris, A. J. J. Straathof, *Appl. Microbiol. Biotechnol.* **2012**, *93*, 1377–1387. DOI: https://doi.org/10.1007/s00253-011-3853-7

[6] J. Bozell, J. Holladay, D. Johnson, J. White, *Technical Report*, Pasific Northwest National Laboratory, **2007**. www.pnnl.gov/main/publications/external/technical_reports/PNNL-16983.pdf (Accessed on May 18, 2018)

[7] IEA Bioenergy, *Report*, **2011**. www.ieabioenergy.com/wp-content/uploads/2013/10/Task-42-Biobased-Chemicals-value-added-products-from-biorefineries.pdf (Accessed on May 18, 2018)

[8] M. Galbe, G. Zacchi, *Appl. Microbiol. Biotechnol.* **2002**, *59*, 618–628. DOI: https://doi.org/10.1007/s00253-002-1058-9

[9] Y. Sun, J. Cheng, *Bioresour. Technol.* **2002**, 1–11. DOI: https://doi.org/10.1016/S0960-8524(01)00212-7

[10] M. J. Takerzadeh, K. Karimi, *BioResources* **2007**, *2*, 472–499.

[11] M. J. Takerzadeh, K. Karimi, in *Biofuels Refining and Performance* (Ed: A. Nag), McGraw Hill, New York **2008**, 69–106.
[12] J. Heinonen, A. Tamminen, J. Uusitalo, T. Sainio, *J. Chem. Technol. Biotechnol.* **2012**, *87*, 689–696. DOI: https://doi.org/10.1002/jctb.2766
[13] G. Machado, S. Leon, F. Santos, R. Lourega, J. Dullius, M. E. Mollmann, P. Eichler, *Nat. Resour.* **2016**, *7*, 115–129. DOI: https://doi.org/10.4236/nr.2016.73012
[14] L. Liu, H. M. Chang, H. Jameel, J. Y. Park, S. Park, *Ind. Eng. Chem. Res.* **2017**, *56*, 14447–14453. DOI: https://doi.org/10.1021/acs.iecr.7b03635
[15] J. R. Weil, B. Dien, R. Bothast, R. Hendrickson, N. S. Mosier, M. R. Ladisch, *Ind. Eng. Chem. Res.* **2002**, *41*, 6132–6138. DOI: https://doi.org/10.1021/ie0201056
[16] B. Hahn-Hägerdal, K. Karhumaa, C. Fonseca, I. Spencer-Martins, M. F. Gorwa-Grauslund, *Appl. Microbiol. Biotechnol.* **2007**, *74*, 937–953. DOI: https://doi.org/10.1007/s00253-006-0827-2
[17] R. C. Sun, *BioResources* **2009**, *4*, 452–455. DOI: https://doi.org/10.15376/biores.4.2.452-455
[18] J. Heinonen, T. Sainio, *Ind. Eng. Chem. Res.* **2010**, *49*, 2907–2915. DOI: https://doi.org/10.1021/ie901598z
[19] J. Heinonen, T. Sainio, *Sep. Purif. Technol.* **2014**, *129*, 137–149. DOI: https://doi.org/10.1016/j.seppur.2014.03.031
[20] Y. Xie, D. Phelps, C. H. Lee, M. Sedlak, N. Ho, N. H. L. Wang, *Ind. Eng. Chem. Res.* **2005**, *44*, 6816–6823. DOI: https://doi.org/10.1021/ie049079x
[21] C. Fargues, R. Lewandowski, M. L. Lameloise, *Ind. Eng. Chem. Res.* **2010**, *49*, 9248–9257. DOI: https://doi.org/10.1021/ie100330y
[22] T. Sainio, I. Turku, J. Heinonen, *Bioresour. Technol.* **2011**, *102*, 6048–6057. DOI: https://doi.org/10.1016/j.biortech.2011.02.107
[23] A. C. Ijzer, E. Vriezekolk, E. Rolevink, K. Nijmeijer, *J. Chem. Technol. Biotechnol.* **2015**, *90*, 101–109. DOI: https://doi.org/10.1002/jctb.4294
[24] A. C. Ijzer, E. Vriezekolk, T. Dekic Zivkovic, K. Nijmeijer, *J. Chem. Technol. Biotechnol.* **2016**, *91*, 96–104. DOI: https://doi.org/10.1002/jctb.4631
[25] J. Heinonen, Q. Sanlaville, H. Niskakoski, J. Tamper, T. Sainio, *Sep. Purif. Technol.* **2017**, *186*, 125–134. DOI: https://doi.org/10.1016/j.seppur.2017.06.001
[26] S. S. Mohtar, T. N. Z. Tengku Malim Busu, A. M. Md Noor, N. Shaari, N. A. Yusoff, M. A. Bustam Khalil, M. I. Abdul Mutalib, H. B. Mat, *Bioresour. Technol.* **2015**, *192*, 212–218. DOI: https://doi.org/10.1016/j.biortech.2015.05.029
[27] R. Duan, B. S. Westerlind, M. Norgren, I. Anugwom, P. Virtanen, J. Mikkola, *BioResources* **2016**, *11*, 8570–8588.
[28] A. Toledano, A. García, I. Mondragon, J. Labidi, *Sep. Purif. Technol.* **2010**, *71*, 38–43. DOI: https://doi.org/10.1016/j.seppur.2009.10.024
[29] A. Arkell, J. Olsson, O. Wallberg, *Chem. Eng. Res. Des.* **2014**, *92*, 1792–1800. DOI: https://doi.org/10.1016/j.cherd.2013.12.018
[30] S. Hellstén, J. Lahti, J. Heinonen, M. Kallioinen, M. Mänttäri, T. Sainio, *Chem. Eng. Res. Des.* **2013**, *91*, 2765–2774. DOI: https://doi.org/10.1016/j.cherd.2013.06.001
[31] S. V. Mohan, J. Karthikeyan, *Environ. Pollut.* **1997**, *97*, 183–187. DOI: https://doi.org/10.1016/S0269-7491(97)00025-0
[32] D. Montané, D. Nabarlatz, A. Martorell, V. Torné-Fernández, V. Fierro, *Ind. Eng. Chem. Res.* **2006**, *45*, 2294–2302. DOI: https://doi.org/10.1021/ie051051d
[33] K. I. Andersson, M. Eriksson, M. Norgren, *Ind. Eng. Chem. Res.* **2011**, *50*, 7722–7732. DOI: https://doi.org/10.1021/ie200378s
[34] J. S. Gütsch, H. Sixta, *Ind. Eng. Chem. Res.* **2012**, *51*, 8624–8630. DOI: https://doi.org/10.1021/ie3006116
[35] X. Liu, P. Fatehi, Y. Ni, *Bioresour. Technol.* **2012**, *116*, 492–496. DOI: https://doi.org/10.1016/j.biortech.2012.03.069
[36] J. Shen, I. Kaur, M. M. Baktash, Z. He, Y. Ni, *Bioresour. Technol.* **2013**, *127*, 59–65. DOI: https://doi.org/10.1016/j.biortech.2012.10.031
[37] E. Strand, M. Kallioinen, M. Kleen, M. Mänttäri, K. Spruce, *Nord. Pulp. Pap.* **2015**, *30*, 207–214. DOI: https://doi.org/10.3183/NPPRJ-2015-30-02-p207-214
[38] X. Chen, Q. Yang, C. L. Si, Z. Wang, D. Huo, Y. Hong, Z. Li, *ACS Sustainable Chem. Eng.* **2016**, *4*, 937–943. DOI: https://doi.org/10.1021/acssuschemeng.5b01029
[39] K. Dorfner, in *Ion Exchange* (Ed: K. Dorfner), Walter de Gruyter, Berlin **1991**, 7–189.
[40] G. Brunow, K. Lundquist, G. Gellerstedt, in *Analytical Methods in Wood Chemistry, Pulping, and Papermaking* (Eds: E. Sjöström, R. Alén), Springer-Verlag, Berlin **1999**, 77–124.
[41] E. Sjöström, *Wood Chemistry – Fundamentals and Applications*, 2nd ed., Academic Press, New York **1993**.
[42] TAPPI standard UM 250, in *TAPPI Useful Methods* **1991**, 47–48.
[43] M. Kaspereit, T. Sainio, *Chem. Eng. Sci.* **2011**, *66*, 5428–5438. DOI: https://doi.org/10.1016/j.ces.2011.07.058
[44] F. Helfferich, *Ion Exchange*, Dower Publications Inc., Mineola, NY **1995**.
[45] X. Lancrenon, D. Herve, *Sugar Technol. Rev.* **1988**, *14*, 207–274.
[46] M. Kearney, D. E. Rearick, www.arifractal.com/images/files/Weak-cation-exchange-softening.pdf (Accessed on May 18, 2018)
[47] H. J. Catchpoole, R. A. Shalliker, G. R. Dennis, G. Guiochon, *J. Chromatogr., A* **2006**, *1117*, 137–145. DOI: https://doi.org/10.1016/j.chroma.2006.03.074
[48] R. A. Shalliker, H. J. Catchpoole, G. R. Dennis, G. Guiochon, *J. Chromatogr., A* **2007**, *1142*, 48–55. DOI: https://doi.org/10.1016/j.chroma.2006.09.059
[49] D. Wu, R. Fu, *Microporous Mesoporous Mater.* **2006**, *96*, 115–120. DOI: https://doi.org/10.1016/j.micromeso.2006.06.022
[50] F. B. Oliveira, C. Gardrat, C. Enjalbal, E. Frollini, A. Castellan, *J. Appl. Polym. Sci.* **2008**, *109*, 2291–2303. DOI: https://doi.org/10.1002/app.28312
[51] B. K. Glód, *Neurochem. Res.* **1997**, *22*, 1237–1248. DOI: https://doi.org/10.1023/A:1021933013492
[52] H. G. Nam, G. W. Lim, S. Mun, *J. Chem. Eng. Data* **2012**, *57*, 2102–2108. DOI: https://doi.org/10.1021/je201065u
[53] D. Painer, S. Lux, M. Siebenhofer, *Sep. Sci. Technol.* **2015**, *50*, 2930–2936. DOI: https://doi.org/10.1080/01496395.2015.1085407
[54] L. Alders, G. Baijlé, *U.S. Patent 3 041 373*, **1962**.
[55] L. Berg, *U.S. Patent 4 692 219*, **1987**.

