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Liquid Hot Water Pretreatment of Lignocellulosic Biomass at Lab and Pilot Scale

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Abstract: Liquid hot water pretreatment is considered to be a promising method for increasing biomass digestibility due to the moderate operational conditions without chemical additions. A necessary step towards the scalability of this pretreatment process is performing pilot plant trials. Upscaling was evaluated with a scaling factor of 500, by using 50 mL in the laboratory and 25 L in a pilot plant batch reactor. Pretreatment times were varied from 30 to 240 min, and temperatures used were 180–188 °C, while applying similar heating profiles at both scales. The initial mass fraction of poplar wood chips ranged from 10% to 16%. Liquid hot water pretreatment at laboratory and pilot scale led to analogous results. The acetic acid analysis of the liquid and solid fractions obtained after pretreatment indicated that complete deacetylation of poplar biomass can be achieved.

Keywords: biorefinery; pilot plant; lignocellulosic biomass; liquid hot water; pretreatment; autohydrolysis

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1. Introduction

The continuous increase in greenhouse gas emissions and the depletion of fossil carbon sources reinforce a transition from oil-based towards bio-based production processes. This motivates extensive research on development of production of bio-chemicals, bio-energy and bio-materials from renewable sources [1]. The use of lignocellulosic biomass as main source for these biorefinery processes is widely accepted, especially because second generation feedstock does not compete with the food supply chain and in many cases even increases sustainability of food production [2].

Pretreatment of biomass is one of the key technologies in bio-based industry, and it has been extensively studied due to the complexity of the internal structure of the lignocellulosic material [3,4]. Different methods have been applied, which usually focus on enhancement of the availability of carbohydrates for subsequent enzymatic hydrolysis and fermentation to bio-fuels and bio-based chemicals [5].

Various compounds are released during the deconstruction of lignocellulosic biomass from pretreatment. Pentoses as well as hemicellulosic hexoses are obtained from hemicellulose hydrolysis. Depending on the pretreatment, cellulose- and lignin-derived monomers and degradation products are also found in significant concentrations after the pretreatment step [6]. Furfural and 5-hydroxymethylfurfural (HMF) are produced from degradation reactions of the C-5 and C-6 monosaccharides, respectively, while acetic acid is generated from the cleavage of acetyl groups, present in the hemicellulose structure up to 5% dry weight [7]. Whereas HMF can further degrade, to formic and levulinic acid, for example, furfural might also decompose to formic acid and other molecules [8]. The severity of the pretreatment increases the extent of carbohydrate release but also the extent of these degradation reactions [9,10].

Most of these side-products are considered inhibitors for a wide range of microorganisms during fermentation, although they are interesting building blocks for bio-based chemicals themselves [11]. Therefore, knowing the presence and proportion of the most relevant side-products after pretreatment might help the design of an efficient bioprocess, as further strategic steps could be taken into account accordingly [12].

Among the physical, chemical, physicochemical and biological pretreatment methods of biomass, liquid hot water (LHW) pretreatment is one of the most attractive procedures, because it involves no addition of chemicals and has relative moderate operational conditions [13]. In contrast, large amounts of water and energy might be required, leading to diluted products and large pieces of equipment [14].

This hydrothermal treatment, also known as autohydrolysis of biomass, involves the addition of water and increase of temperature. This causes a decrease of pH in the system due to the ionization of water and the release of carboxylic acids (mainly acetic acid), in combination with the effect of the high temperature and pressure applied [15,16]. This autohydrolysis leads to the disruption of the lignocellulosic biomass structure, improving the accessible surface area, and therefore enhancing the subsequent enzymatic hydrolysis [17–20] and fermentation steps [21].

Hydrothermal Pretreatment at Pilot Scale

Most previous studies have focused on LHW pretreatment at laboratory scale, and generated a substantial amount of laboratory data [22–24]. Implementation of industrial biorefinery processes entails an appropriate scaling-up procedure to achieve a successfully bioprocess design at industrial level, where a pilot plant is still a "must-have" step in the scalability of the process.

According to Harmsen et al. [25], a pilot plant scale process might be performed for many different reasons, such as provision of reliable stream compositions and insight into realistic process conditions. Hydrothermal pretreatment methods have been previously studied and modeled at pilot scale for a wide range of conditions and settings, including continuous tubular reactors [26–28]. Recently, Ruiz et al. [29] reviewed hydrothermal pretreatment strategies at pilot scale, including biomass structure changes, reactor technologies and engineering from batch to continuous.

Rocha et al. [30] compared pilot scale steam explosion (200 L) of sugarcane bagasse with dilute acid pretreatment, using 1% (w/v) H_2SO_4 in a 350-L batch reactor at 120 °C and 1:10 solid-liquid ratio during 10 min, the former method being more efficient at the tested conditions, with higher hemicellulose solubilization and further cellulose conversion. The same 350 L reactor was used for pilot alkaline pretreatment [31] and also to obtain Enzymatic Hydrolysis Residue Lignin by hydrothermal pretreatment of sugarcane bagasse, processed at 190 °C for 10 min with 1:10 solid-liquid ratio [32].

For the conversion of lignocellulosic biomass into biofuels and a potential scale-up at pilot plant, Rossner and Parra [33] evaluated different pretreatment methods for eucalyptus wood in the laboratory. Autohydrolysis or hydrothermal pretreatment performed better than organosolv and steam explosion. They pretreated *Eucalyptus globulus* wood chips in an 88 L packed bed reactor at 190 °C with 20% solids, obtaining low concentrations of degradation products such as formic acid, levulinic acid, furfural and HMF (values not reported).

LHW pretreatment at pilot scale has been tested also by applying different temperatures (185, 190 and 195 °C) during 10 min to pretreat sugarcane bagasse in a 20 L batch reactor, leading to a high extent of solubilization of hemicellulose and providing a reduction in the recalcitrance of the lignocellulosic material [34].

Krátký et al. [35] processed wheat straw, among other lignocellulosic materials, in an 8 L batch pressure vessel by applying LHW pretreatment with expansion for testing pilot biogas production. They obtained promising results in biogas production by increasing temperature and residence time during pretreatment, although there was no analysis of degradation compounds and their possible effect on the anaerobic digestion.

Nascimento et al. [36] performed hydrothermal pretreatment at laboratory and pilot scale. They pretreated sugarcane bagasse at 190 °C for 10 min, but performed steam explosion at pilot and LHW pretreatment at laboratory, including significant differences during the heating phases. Mosier et al. [37] compared continuous LHW pretreatment of corn fibers at 160 °C for 20 min at lab and pilot scales, using direct injection of steam as heating source at the laboratory, while using indirect heat exchanger instead at larger scale, to avoid dilution and reduce energy consumption. Aguilar et al. [38] also evaluated the hydrothermal pretreatment of agave bagasse for bioethanol production, using the severity factor as scalability factor from 125 mL to 750 mL.

Thus, studies on LHW pretreatment process for lignocellulosic biomass have been performed at larger scale, but systematic comparison to lab scale, at similar composition and heating profile, has not been performed yet.

In order to improve the comparability between scales, as mass and heat transfer depend on scale and stirring conditions, it is important to have similar temperature profiles.

In this work, we try to perform the LHW pretreatment process of poplar biomass at pilot plant scale and laboratory at equivalent conditions, with special interest in using the same temperature profile, in order to confirm the scalability of the process by a factor 500 and the possible optimization at laboratory scale. Poplar wood was selected as biomass source, as in a preceding study [39]. Typical conditions for LHW pretreatment include temperatures around 200 °C with a residence time of a few minutes [17]. To obtain sufficient hemicellulose degradation, the selected pretreatment conditions in this work were in the temperature range of 180–200 °C and residence (contact) time range of 30–240 min.

2. Materials and Methods

2.1. Materials

Acetic acid (≥99.7%), furfural (≥99%), 5-hydroxymethylfurfural (HMF) (≥98%) and xylose (≥99%) were from Sigma-Aldrich. D-Glucose (≥99.5%) was from Merck. Formic acid (≥98%) was from Honeywell Fluka. The lignocellulosic material (poplar wood with stems and bark) was provided by Woodoo GmbH & Co., KG, Grunow-Dammendorf, Germany, through the Bioprocess Pilot Facility B.V, Delft, the Netherlands. Samples were pre-milled in a bench mill and sieved to a mesh size of ~1 cm (moisture mass fraction of 10.8%).

2.2. Pilot Plant Pretreatment

The pilot scale experiments were carried out at the Bioprocess Pilot Facility (BPF). The equipment consisted of a 50 L batch autoclave (Büchi AG, Uster, Switzerland) with approximate inner dimensions Ø 38 cm diameter and 45 cm height. It was provided with a thermic oil-filled jacket connected to an automatic temperature controller, an agitation system with anchor blade stirrer and an inlet line of compressed nitrogen gas. The temperature sensor was positioned parallel to the stirrer axis at 9 cm from the reactor wall and 15 cm from the reactor bottom.

The autoclave vessel was filled with 2.5 kg of poplar chips (including 10.8% moisture content) and then 22.5 kg of water were added (except for the experiment at higher biomass/water ratio, in which 2 kg of biomass were filled together with 10.5 kg of water). The mixture was stirred manually for few minutes and then the vessel was lifted and screwed to the autoclave base. Automatic stirring with the anchor impeller was then switched on at 130 rpm. The oil jacket was switched on to start the heating after flushing the reactor head space with N_2 gas three times (between 0.2–0.4 MPa), with the purpose of avoiding other hydrothermal mechanisms, such as wet oxidation [40].

Temperatures of oil bath and the inside of the reactor were monitored and used by the automatic control system to follow the sequence described below. First, the oil bath was heated to its maximum (200 °C), to achieve the shortest heating phase possible (generally around 80–90 min). Once the inside of the vessel reached 180 ± 2 °C (except for the experiments performed at a pretreatment temperature of 188 °C), the pretreatment phase

was started by switching the control system to oil bath to keep the temperature inside the reactor at this setpoint. At the end of the pretreatment phase (after 30, 120, 180 or 240 min), the pressure was recorded and the oil bath temperature was automatically decreased, allowing the reactor to cool down. When room temperature was reached inside the reactor, the autoclave was opened. Then, the suspension was filtered in a pilot sieve, and the wet solids and liquid fractions were weighed. Samples of both fractions were stored at −20 °C until further analysis.

2.3. Laboratory Pretreatment

At lab scale, a pressurized reactor was used as described in Jimenez-Gutierrez et al. [39], which consists of a stainless steel 100 mL vessel with an electrical heating and an agitation system. The reactor was connected to an inlet line for N_2, and to two outlet lines for gas, one of them with a safety relief valve set at 2.2 MPa.

The reactor vessel was filled with 5 g of biomass, including its 10.8% moisture content, and 45 g of water (except for the experiment at higher biomass/water ratio, in which 5.25 g of biomass was used with 29.75 g of water). Insulation fabric was then placed around the vessel. Before starting the experiments, all pipes and valves were shortly flushed with N_2, assuming negligible evaporation of water. Then, the agitation and the heating system were switched on, increasing the temperature inside the vessel up to 180 °C (except for the experiment performed at pretreatment temperature of 188 °C), which increased the pressure up to the "pretreatment pressure" value (see Supplementary Material). The reactor was kept at the set temperature by the controller during the pretreatment time (30, 120, 180 and 240 min). Subsequently, the heating system was switched off and the insulating fabric was removed from the reactor, allowing a fan to speed up the cooling of the vessel to room temperature. When ambient conditions were reached, the reaction suspension was filtered (Whatman 0.2 µm), and the pH of the liquid fraction was measured. Wet solid and liquid fractions were weighed to calculate the percentage of total mass loss due to handling (see Supplementary Material), as the difference from the total mass initially added to the reactor (biomass + water) and the total amount obtained after LHW pretreatment (solid + liquid fractions). Several liquid samples were then taken and stored at −20 °C until further analysis by HPLC. For the laboratory experiments, the solids were washed twice with 100 mL of demi-water, dried overnight in an oven at 105 °C and weighed. For the pilot scale, not all solids were used for the analytical procedure but only samples of the solids. Samples of the washing liquid, as well as the washed and dried solids were stored for further analysis.

2.4. Analysis of Samples

Solid samples were analyzed following a slightly modified methodology based on the NREL (National Renewable Energy Laboratory) laboratory analytical procedure (LAP) for determination of structural carbohydrates in biomass [41]. A two-step acid hydrolysis process was carried out. Firstly, 1.5 g of the dried, milled and sieved solids (180 µm mesh size) were incubated with a 72% w/w solution of H_2SO_4 during 60 min at 30 °C and then, after dilution to 4% w/w, the mixture was autoclaved for 1 h at 121 °C. Once room temperature was reached, calcium carbonate was added to an aliquot of the filtered liquid (Whatman 0.2 µm) to neutralize the acid until pH reached 6–7. The neutralized liquid samples were filtered again and analyzed by HPLC as described below. Lignin and ash content were not determined in this work.

Liquid samples were analyzed on a Waters HPLC system with a Bio-Rad HPX-87H column (300 × 7.8 mm) at 60 °C. Phosphoric acid (1.5 mmol/L at 0.6 mL/min) was used as eluent. Glucose, xylose, acetic acid, furfural and HMF were detected by refraction index (Waters 2414), while formic acid was detected using UV (Waters 2489) at 210 nm. Quantification was performed using external standards. Volumes of injection of standards and samples were 10 µL at 15 °C. All samples were analyzed in quadruplicate. After statistical analysis of these data, in most of the cases, the values of each analysis showed less than 5%

of relative standard deviation, and in many cases lower than 1% (see Supplementary Data). The small error bars have not been displayed in the graphical representation of the results to facilitate readability.

3. Results and Discussion

3.1. Heating Profiles

As previously indicated [42], the LHW pretreatment method follows a semi-isothermal profile. The temperature profile is divided in three parts: heating phase, pretreatment phase and cooling phase. The temperature profiles that we obtained at lab and pilot scale were equivalent during the pretreatment and cooling phase. Only small differences were observed during the heating phase (see Figure 1 and Supplementary Material). Hydrolysis of hemicellulose typically occurs at 170–190 °C, whereas cellulose hydrolysis typically occurs above 200 °C [43,44]. Therefore, the major part of the hydrolysis is assumed to happen during the (isothermal) pretreatment phase. However, some hemicellulose hydrolysis may occur during the last part of the heating phase, named the "active heating", while in the cooling phase the temperature drops quickly, such that hydrolysis during this phase is considered negligible compared to the other two phases. Assuming an active heating from 160 °C up to the pretreatment temperature, the average difference during this period between temperature profiles was 3.8 °C/min. In this case, the duration of this active heating was 30 min, which was four times lower than the total pretreatment time. Consequently, the experiments performed with 30 min pretreatment time may be slightly "overestimated" for the pilot plant. However, for the rest of the pretreatment times used, both profiles are considered analogous, facilitating the comparison of scales.

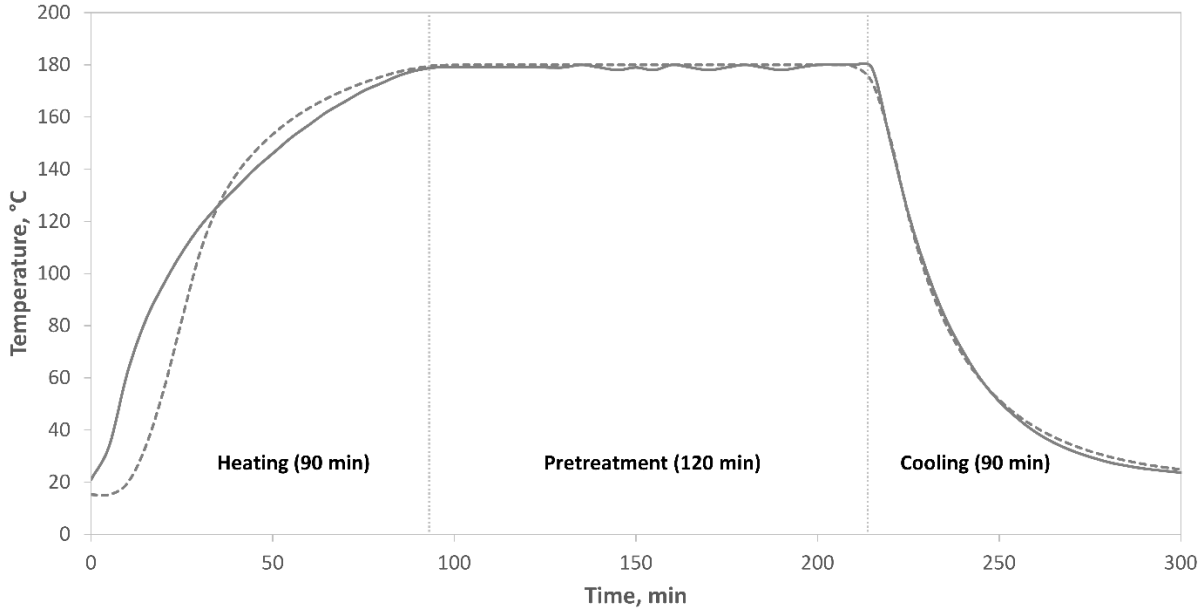

Figure 1. Temperature profiles at laboratory (—) and pilot plant (- - -) scale during liquid hot water pretreatment of poplar at 180 °C and 120 min.

Nevertheless, it was observed that the temperature profiles for the "higher temperature" experiments (188 °C pretreatment temperature) from lab to pilot did not fully match (see Supplementary Material). It took longer to heat the reactor to the set temperature at pilot scale (~120 min) than at lab scale (~60 min). This happened probably because there was a shorter temperature difference between the inside of the reactor and the temperature of the heating source (thermal oil) at the pilot plant, limited to a maximum of 200 °C, while in the lab, the heating source reached higher values (~260 °C). Consequently, despite keeping the same pretreatment times at both scales, the doubled heating time at pilot scale probably increased the extent of reaction during the active heating.

It is important to point out that heating is equipment dependent, and therefore heating profiles depend on scale [45] unless they are controlled at both scales, as in this paper.

3.2. *Effect of Time*

Results from analysis of the liquid fraction samples after LHW pretreatment at 180 °C and different pretreatment times at laboratory and pilot scale are comparable, as shown in Figure 2A,B. Slightly higher concentrations of carbohydrate degradation products (HMF, furfural) are obtained at the pilot scale (from 45–50% more at 30 min pretreatment time, to 20–40% at 240 min, respectively). In addition, formic acid concentrations were generally higher at pilot scale. However, increased carbohydrate degradation is not reflected by lower xylose and glucose concentrations. The concentrations of xylose and glucose reached a maximum point from which their degradation overtook their release by hemicellulose hydrolysis. Such maximum points are generally seen in literature, also when conditions differ somewhat [23,38].

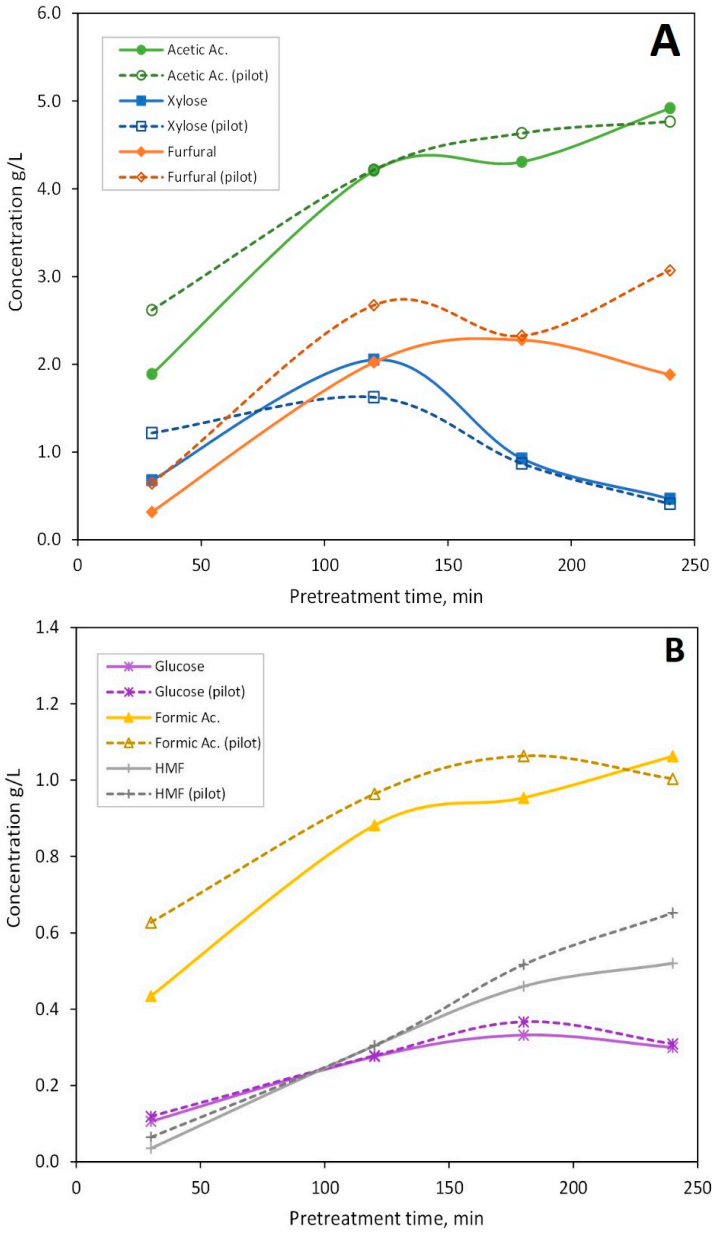

Figure 2. Concentration of compounds released by LHW pretreatment at lab scale (closed markers) and pilot scale (open markers) at 180 °C. The lines describe the trend. (**A**): Acetic acid, xylose and furfural. (**B**): Glucose, formic acid and HMF.

Figure 3A,B represent the glucan and xylan content of the solids remaining from the same experiments, considering glucan as cellulose representative and xylan as hemicellulose representative. Corrected to the anhydrous form of glucose and xylose, respectively, these values are calculated as described in Supplementary Material, taking into accounts the mass loss due to handling/processing and the raw/pretreated biomass ratio in dry basis, with a moisture content of 10.8% for the raw material. The fraction of glucan in the pretreated solids seems to increase slightly, indicating that most of the cellulose remained intact in the solid fraction, whereas the hemicellulose was progressively dissolved, which is in line with literature [22] and with the data obtained from the analysis of the liquid.

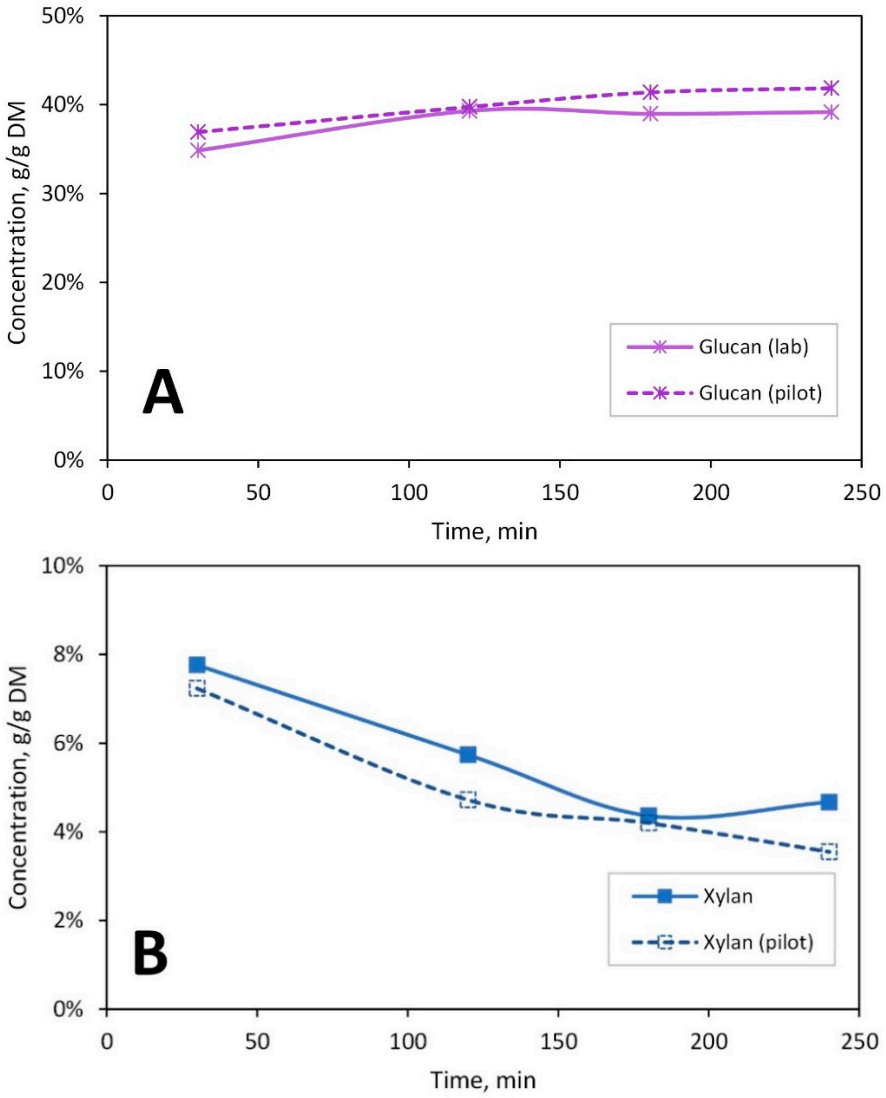

Figure 3. Concentration of glucan (**A**) and xylan (**B**) in the solid fraction analyzed after LHW pretreatment at labs scale (closed markers) and pilot scale (open markers lines) at 180 °C, performed at different pretreatment times.

In general, the LHW pretreatment might seem to be slightly faster at pilot scale as compared to lab scale. However, given the analytical error threshold and the relative small difference in the values from both scales, it would be more realistic to claim that the concentration of the compounds analyzed followed similar trends with increasing the pretreatment times, therefore concluding that LHW pretreatment of poplar at the used laboratory and pilot scale behaved in the same way. Besides experimental error, small gradients inside the pilot vessel and small differences between lab and pilot scale procedures may explain the minor differences in concentrations between pilot and lab. In

line with this, the pH values measured from the liquid fraction after pretreatment were also in a similar range for both scales (see Supplementary Material); additionally, no significant differences at the surface of the pretreated material were observed at the microscope (see Supplementary Material), supporting analogous performance at lab and pilot scales. Besides the compounds analyzed, there are numerous other compounds produced, such as humins and pseudo-lignin. We assume that reaction conditions that do not influence the concentrations of analyzed compounds also do not influence the concentrations of non-analyzed compounds, because of constraints imposed by mass balances reaction kinetics. Therefore, concentrations of non-analyzed compounds are assumed to be similar at lab and pilot scale, although deviations cannot be excluded.

The absence of scale effects suggests that mass and heat transfer are not limiting at the conditions and scales used. Hence, lab tests can be used for proper optimization of LHW pretreatment, before switching to the used pilot scale. For even larger scales, mass and heat limitations might still occur and lead to deviations from lab scale results.

3.3. Temperature Effect

When pretreating at higher temperature, in general more hydrolysis of biomass is expected. Concentrations of different compounds of the liquid fraction after LHW pretreatment of poplar at two different temperatures are shown in Table 1. More glucose was obtained at 188 °C than at 180 °C, as more cellulose hydrolysis occurs at higher temperatures [44]. Additionally, a larger quantity of HMF was found, though still in very dilute amounts. Processing at higher temperatures is generally known to lead to a higher HMF concentration [43]. Regarding hemicelluloses, a considerable difference in the dissolved xylose concentration was observed when comparing these two temperatures. The LHW process at 188 °C reduced the xylose concentration, at first sight due to formation of degradation compounds such as furfural. However, the amount of furfural produced is not proportional to the lower xylose concentration (on basis of a 1:1 stoichiometry in the degradation reaction) [46] but lower, in case of both lab and pilot scale. This phenomenon can be explained by thermal degradation of furfural, which is also enhanced at higher temperatures [47], and contributes to the formation of other degradation compounds, such as formic acid. Nevertheless, the concentration of formic acid just slightly increased at higher temperatures, following the trend observed previously [39], suggesting that most furfural degradation products were not analyzed in this work.

Table 1. Concentration of different compounds in the liquid fraction after LHW pretreatment of poplar at two temperatures, both at lab and pilot scale, with 120 min pretreatment time and 10% initial biomass fraction.

Scale	Temperature (°C)	Glucose (g/L)	Xylose (g/L)	Formic Acid (g/L)	Acetic Acid (g/L)	HMF (g/L)	Furfural (g/L)
Lab	180	0.28	2.05	0.88	4.20	0.30	2.02
	188	0.33	0.56	0.97	4.40	0.54	1.80
Pilot	180	0.28	1.62	0.96	4.22	0.31	2.67
	188	0.31	0.39	1.06	4.67	0.65	2.15

The absolute value of the difference between lab and pilot scale is larger for the concentrations in the case of pretreating at 188 °C (19%) than at 180 °C (13%). This may be explained by the larger difference at 188 °C between lab and pilot experiments with respect to the heating profiles, as described in Section 3.1.

The relatively severe heating in the pilot pretreatment at 188 °C led to more degradation (xylose being 30% lower, HMF 21% higher, and furfural 20% higher than in the lab). Others [36] found less degradation at pilot than at lab scale when pretreating at 190 °C. This could be explained by their heating profiles, which were very different from lab to pilot, being 90 min longer at lab scale. Values of the solid fraction composition at 188 °C pretreating temperature are in line with those found in literature [32] when using a 350 L

batch reactor to obtain Enzymatic Hydrolysis Residual Lignin from sugarcane bagasse at 190 °C during a shorter time.

3.4. Biomass/Water Ratio Effect

Since different amounts of solid loading and water were used, the results were expressed as yield in "milligrams of compound per gram of biomass" (Table 2), estimated by multiplying measured concentrations by the liquid volume initially added and dividing by the biomass loaded. When increasing the biomass fraction up to 15–16%, results indicated again that LHW pretreatment worked correspondingly at both scales. For instance, the yield of acetic acid remained similarly at either scale when adding more solids. A few inconsistencies can be observed, however. For example, the amount of xylose obtained was significantly lower (26%) at pilot scale than at the lab in the case of 10% biomass fraction. This is mirrored by a furfural increase. Yet, at 15–16% biomass no such difference between lab and pilot scale was seen, such that no correlation between scale and conversion remains.

Table 2. Yield of different compounds on initial dry biomass for experiments involving 120 min pretreatment time at 180 °C.

Scale	Initial Biomass Fraction	Glucose mg/g	Xylose mg/g	Formic Acid mg/g	Acetic Acid mg/g	HMF mg/g	Furfural mg/g
Lab	10%	2.48	18.46	7.94	37.84	2.74	18.19
	15%	2.75	9.55	8.83	36.46	2.92	16.17
Pilot	10%	2.50	14.62	8.68	37.96	2.74	24.05
	16%	2.98	9.91	9.71	37.63	3.28	16.85

Irrespective of scale, a minor correlation can be observed between the initial biomass fraction and the yields. Increasing biomass fractions may lead to higher severity, as observed from a decrease in xylose yield and a slight increase in glucose and HMF yield. The higher severity could be explained by the release of more carboxylic acid (from more biomass) in less aqueous liquid. Thus, although the amounts of acetic acid obtained per gram of biomass were almost equal at higher biomass fraction and at both scales, a higher aqueous concentration of acetic acid was observed at larger initial biomass loading (see Supplementary Material).

Consequently, when expressing the results as "concentration in g/L" the about 50% increase in the initial biomass fraction led to proportional increments in the concentrations of most of the compounds analyzed (see Supplementary Material).

3.5. Complete De-Acetylation

During pretreatment of lignocellulosic biomass, acetic acid is released as a result of the hydrolysis of the acetyl groups located in the hemicellulose structure. The selected biomass for this study comprises a high acetyl content. Acetic acid is considered an inhibitor for several microorganisms [48], thus its presence would not be desired for the subsequent enzymatic hydrolysis and fermentation steps after pretreatment [49]. According to results in Figure 2A, acetic acid release showed the expected increase with pretreatment time before reaching a plateau, as observed in previous laboratory experiments [39], which suggested a complete hydrolysis of acetyl groups from hemicelluloses. Similarly, results from the higher biomass/water ratio experiments (Table 2) indicate full acetic acid release regardless the initial biomass fraction loaded. Data obtained from the two-step hydrolysis LAP applied to the solid fraction (Supplementary Data), as well as the analysis of washing water after pretreatment and filtration (Supplementary Material) confirmed the de-acetylation of the poplar biomass by LHW after 120 min at 180 °C at both scales according to the HPLC detection limit (0.01% acetyl content on dry basis as the minimum value for the analytical calibration in HPLC).

4. Conclusions

LHW pretreatment of poplar biomass has been compared at laboratory and pilot scale. At either scale there was a considerable extent of hemicellulose hydrolysis and formation of degradation compounds, such as formic acid and furfural. In general, at similar conditions, including a similar temperature profile, LHW pretreatment behaved in a similar way at laboratory and pilot plant scale, allowing optimization of the process at lab scale before moving to larger scale. Complete de-acetylation of poplar wood was achieved at both scales applying 180 °C for 120 min as a pretreatment condition. At both scales, higher pretreatment temperature led to larger glucose and acetic acid release, but simultaneously to more formation of sugar degradation compounds. Increasing the biomass/water ratio resulted also in similar behavior at both scales, and no significant correlation between scale and conversion per amount of initial biomass loaded was observed.

Supplementary Materials: The following files are available online at https://www.mdpi.com/article/10.3390/pr9091518/s1, Supplementary Material.pdf, containing additional information, and Supplementary Data.xlsx, containing raw data.

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