



A two-stage pretreatment approach to maximise sugar yield and enhance reactive lignin recovery from poplar wood chips



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HIGHLIGHTS

- ▶ A 2-stage steam/organosolv pretreatment resulted in good sugar and lignin recovery.
- ▶ Close to 90% of the original xylan was recovered in a useable form.
- ▶ Close to 90% of the original cellulose could be hydrolysed at low enzyme loadings.
- ▶ The lignin recovered after 2-stage pretreatment was more reactive.

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ABSTRACT

A two-stage pretreatment approach, employing steam followed by organosolv treatment, was assessed for its ability to fractionate and recover most of the hemicellulose, lignin and cellulose components of poplar wood chips. A mild steaming stage was initially used to maximise hemicellulose sugar recovery, with 63% of the original xylan solubilised and recovered after this stage and close to 90% recovered in total. Rather than hindering subsequent organosolv delignification, the prior steam treatment enhanced lignin solubilisation with more than 66% of the original lignin removed after the two-stage pretreatment. The extracted lignin contained at least equal or greater amounts of functional groups as compared to the lignin solubilised after a single-stage organosolv pretreatment. More than 98% of the original cellulose was recovered after the two-stage pretreatment and 88% of the cellulose could be hydrolysed to glucose at enzyme loading of 5 FPU/g cellulose after 72 h.

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1. Introduction

A typical biomass-to-ethanol process usually has the major process steps of pretreatment, fractionation and recovery of the hemicellulose, lignin and cellulose streams, followed by enzymatic hydrolysis of the cellulose rich, water insoluble stream and fermentation of all of the biomass derived sugars to ethanol. The pretreatment step has a “domino effect” on all of the subsequent processing steps, influencing the recovery of the hemicellulose sugars, the ease of lignin extraction and use and the effectiveness of enzymatic hydrolysis of the cellulosic fraction when low enzyme loadings are used (Galbe and Zacchi, 2012). Ideally, any successful pretreatment should be low-cost, provide the clean fractionation and recovery of all of the initial biomass components in a usable form, while producing a cellulosic substrate at high substrate concentrations which can be readily hydrolysed at minimal enzyme loadings over a short period of time. Although there does not, as

yet, seem to be any pretreatment process that meets all of these criteria, the work described below demonstrates the beneficial and complementary characteristics of steam and organosolv pretreatments.

Steam pretreatment is a relatively low-cost pretreatment method which is rapid, requires limited chemical and energy input and is currently being evaluated by a number of companies including Inbicon (Kalundborg, Denmark), Iogen (Ottawa, Canada), Abengoa (Salamanca, Spain) and Mascoma (Rome NY, USA). Unlike other pretreatment technologies such as hydrothermal and dilute acid which typically require additional size reduction steps, steam pretreatment has been used to process a variety of substrates such as chopped straw and conventional pulp chips (Grous et al., 1986; Ballesteros et al., 2006). Pioneering work on steam pretreatment exploited the inherent acetyl groups on the hemicellulose of hardwoods and agricultural feedstocks to “auto-catalyse” the pretreatment resulting in the solubilisation of hemicellulose and an increase in accessibility of the cellulose component (Grous et al., 1986). In this earlier work, so-called optimum steam pretreatment conditions were ones that helped increase the ease of enzymatic hydrolysis of the cellulose component regardless if this was

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accomplished at the expense of recovering most of the hemicellulose or condensing the lignin, making it difficult to extract and to use (Bura et al., 2002). The use of catalysts, such as sulfur dioxide and sulfuric acid, and lower pretreatment severities have been successfully used to solubilise and recover more of the hemicellulose and enhance cellulose hydrolysis, consequently increasing overall sugar recovery (Mackie et al., 1985). Sulfur dioxide's ability to permeate more uniformly throughout most biomass substrates as compared to sulfuric acid, has indicated that is often a more effective catalyst (Mackie et al., 1985; Eklund et al., 1995). Steam pretreatment has also been shown to be particularly effective in processing agricultural residues (corn stover, wheat straw) and hardwoods (poplar), with more than 60% of the hemicellulose recovered and near complete hydrolysis of the water insoluble cellulosic component achieved, albeit at typical cellulase loadings in excess of 15 FPU/g glucan (Excoffier et al., 1991; Ballesteros et al., 2006; Bura et al., 2009). When enzyme loadings are decreased, one of the few ways to obtain as good hydrolysis yields as were obtained previously at the higher enzyme loadings was to pretreat the biomass substrates more severely. However, this is usually achieved at the expense of hemicellulose sugar degradation (Bura et al., 2009), lignin condensation (Li et al., 2007) and the production of increasing amounts of fermentation inhibitors (Palmqvist et al., 1996; Bura et al., 2009). As well inhibiting enzymatic hydrolysis, increases in lignin condensation have also been shown to decrease the potential utility of lignin as a co-product (Shevchenko et al., 1999; Li et al., 2007). Thus, due to this lignin fractions lack of reactivity it is often considered best utilised as a combustion/energy source to power the overall process.

Unlike steam pretreatment which is primarily focused on solubilisation of the hemicellulose component, organosolv pretreatment acts by extracting the lignin component by scission reactions that facilitate its solubilisation (McDonough, 1993; Pan et al., 2006a). The relatively high purity, reactivity and narrow polydispersity of organosolv lignin have all suggested that it would have considerable potential as a feedstock for the production of higher value products such as adhesives, antioxidants and carbon fibres (Kelley et al., 1989; Kadla et al., 2002; Lora and Glasser, 2002). The effective solubilisation of much of the lignin and hemicellulose during organosolv pretreatment results in a highly accessible cellulosic fraction which is highly amenable to subsequent enzymatic hydrolysis, at low enzyme loadings, regardless of the type of biomass that has been treated (Pan et al., 2006a; Huijgen et al., 2011). However, as noted above, the hemicellulose fraction is typically solubilised along with the lignin, making it difficult to recover and utilise the hemicellulose derived sugars. Thus, it would be highly desirable if the hemicellulose sugars could first be recovered prior to the organosolv pretreatment stage which is primarily aimed at effective lignin removal, recovery and use while further enhancing the accessibility of the cellulose to the cellulase enzyme complex.

The solubilisation of the hemicellulose component of agricultural and hardwood substrates using acid-catalysed pretreatments has been shown to be an important mechanism to ensure the generation of a cellulosic substrate which is more amenable to enzymatic hydrolysis. Therefore, the selective removal of hemicellulose prior to a pretreatment such as organosolv may be an effective approach to aid in the fractionation of the biomass and increasing the accessibility of the cellulose to the cellulolytic enzymes. However, as mentioned earlier, it is possible that the lignin component will undergo condensation during a prior steam pretreatment stage, consequently limiting is extractability in the subsequent organosolv stage (Li et al., 2007). In the work reported here we have assessed if a mild steam treatment step could first solubilise and recover most of the hemicellulose while limiting the influence of the acid-catalysed steam pretreatment on the ease

of extraction of the lignin component. In addition it was hypothesised that, due to the partial deconstruction of the wood chips during the steam pretreatment stage, it might also be possible to solubilise the lignin using milder conditions than those (180 °C, 60 min and 1.25% H₂SO₄) used previously during the organosolv pretreatment of poplar (Pan et al., 2006a). The removal of both hemicellulose (Bura et al., 2009) and lignin (Jin et al., 2010) has been shown to enhance the hydrolysis of pretreated poplar substrates. Thus, to see if we could maximise the beneficial effects of these complementary pretreatment processes a mild steam treatment followed by organosolv pretreatment was used to fractionate the cellulose, hemicellulose, and lignin components of poplar wood chips while generating a cellulosic substrate which could be readily hydrolysed at the relatively low enzyme loading of 5 FPU/g cellulose.

2. Methods

2.1. Biomass substrate

The poplar used in this study was collected in the spring of 2009 by the BC Ministry of Forests, Canada. After air storage the moisture content of the poplar chips was 6.9%. The chips were screened using a plate screen with the fraction between 2.5 × 2.5 cm and 5.0 × 5.0 cm collected as the raw material for pretreatment. The samples were stored and protected from the weather at room temperature. The polysaccharide content of poplar was approximately 71% comprised of glucan (49.3%), xylan (17.6%), mannan (2.8%), arabinan (0.7%) and galactan (0.4%). The total lignin content was 25.4% consisting mainly of acid-insoluble lignin (23.1%).

2.2. Steam pretreatment (SP)

The conditions of steam pretreatment of poplar are shown in Table 1. To facilitate the comparison among the experimental results, the SP conditions were also expressed as the severity factor;

$$R_0 = t \cdot \exp[(T - 100)/14.75]$$

where t is residence time in minutes and T is pretreatment temperature in °C (Overend and Chornet, 1987). Prior to steam pretreatment, 75 g of dry poplar chips were placed in plastic bags, sprayed with water at a solid:liquid ratio of 1:1 and left at room temperature overnight. Sulfur dioxide was subsequently added to the biomass in the plastic bags at the specified loading (Table 1). The impregnated biomass samples were subsequently loaded into a 2-L Stake Tech II steam gun (Stake Tech II batch reactor, SunOpta (formerly Stake Technologies) of Norval, ON, Canada). The resulting slurry, which typically had a solid:liquid ratio of 1:5, was collected and the water-soluble fraction was separated from the solid fraction with vacuum filtration. The water-soluble fraction was centrifuged and the supernatant phase collected and analysed for monomeric and di- and oligomeric sugars. These are referred to in the text as oligomeric sugars. Once the solid fraction was washed with 0.5 L deionized water it was subsequently used as the feedstock for all the organosolv pretreatment experiments.

2.3. Organosolv pretreatment (OS)

The organosolv pretreatment was performed according to the procedure described in detail by Pan et al. (2006a) at conditions shown in Table 2. Briefly, 75 g (on a dry matter basis) of biomass was pretreated in aqueous ethanol with sulfuric acid as a catalyst using a four-vessel (2 L each) rotating digester (Aurora Products, Savona, BC, Canada). At the end of the pretreatment period, the vessels were cooled to room temperature in a water bath. The solid

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