THERMO-MECHANICAL PULPING AS A PRETREATMENT FOR AGRICULTURAL BIOMASS FOR BIOCHEMICAL CONVERSION

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The use of thermo-mechanical pulping (TMP), an existing and well known technology in the pulp and paper industry, is proposed as a potential pretreatment pathway of agriculture biomass for monomeric sugar production in preparation for further fermentation into alcohol species. Three agricultural biomass types, corn stover, wheat straw, and sweet sorghum bagasse, were pretreated in a TMP unit under two temperature conditions, 160 ºC and 170 ºC, and hydrolyzed using cellulase at 5, 10, and 20 FPU/g OD biomass. Wheat straw biomass was further pretreated at different conditions including: i) soaking with acetic acid, ii) longer steaming residence time (15 and 30 min), and iii) refined at lower disk gap (0.0508 and 0.1524 mm). Preliminary results showed that carbohydrate conversion increased from 25% to 40% when the TMP temperature was increased from 160 to 170 ºC. Carbohydrate conversion was relatively similar for the three biomasses under the same pretreatment conditions and enzyme loading. Acetic acid soaking and refining at a reduce disk gap increases carbohydrate conversion. Further studies within this technological field to identify optimum process and TMP conditions for pretreatment are suggested.

Keywords: Corn stover; Sweet sorghum bagasse; Wheat straw; Glucan, Enzyme; Hydrolysis; Ethanol; Thermo-mechanical pulping; Refining

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INTRODUCTION

Background

In order to meet future domestic energy demand many countries are developing strategic plans to reduce consumption through more efficient use of their resources, while developing alternative sources of energy such as solar, geo-thermal (electricity and heating), wind, biomass, hydroelectric, and ocean (Turner 1999). Of all the energy sources mentioned, biomass is the only one that can be used to produce liquid fuels (to replace internal combustion engine fuels) in addition to heat and electricity (Gonzalez et al. 2011a; Jackson et al. 2010; Wu et al. 2010; Gonzalez et al. 2011b). To date, conversion of biomass to bioethanol has been widely studied, incorporating many different feed-stocks and process technologies. The U.S. and Brazil are the major producers of fuel ethanol worldwide (RFA 2010), relying mainly on corn and sugar cane, respectively (Goldemberg 2007; Pimentel and Patzek 2008). The use of food-based raw materials for fuel production has instigated the fuel versus food debate (Runge et al.

2007). Besides the ethical issues surrounding the use of grains (i.e. corn) for ethanol production, the fact that there will not be enough corn to meet future ethanol demand in the U.S. is an additional concern (Bohlmann 2006; Yacobucci and Schnepf 2007). As an alternative, cellulosic biomass (especially from agriculture residues and forest operations) has been identified as a promising source for cellulosic ethanol production due to several features reported elsewhere (Galbe and Zacchi 2002; Perlack, Wright et al. 2005; Zhu and Pan 2009). Agricultural biomass and its residues (those left after harvesting or obtained as a co-product) represent an important source of raw material for bioenergy. Studies have reported that the U.S. alone can sustainably produce ca. 998 million dry tons of cellulosic biomass from agriculture per year (Perlack et al. 2005). Availability of various feedstocks across the year brings the possibility of processing multiple feedstocks at one facility and increases flexibility of the biomass supply chain; this might be used as a leverage point to negotiate the cost (price) of raw materials; also it might be an alternative to reduce storage costs of biomass crops having short harvesting windows.

On the other hand, the ease of biomass hydrolysis (i.e. in an enzymatic process) has been reported to depend on such factors as physical barriers due to the presence of lignin (Fan et al. 1982; Mooney et al. 1998), the crystallinity of cellulose, and the low surface area exposed to enzymes (Overend et al. 1987; Zhu et al. 2008). There are specific features intrinsic to the biomass that influence enzymatic digestibly and overall conversion cost. These features are mainly related to the amount and type of carbohydrates, lignin, as well as lignin-carbohydrate complex. Moreover, lignin type, composition, and structure are different between softwoods, hardwoods, and agricultural feedstocks (Mooney et al. 1998; Chandra et al. 2007). Generally speaking, agricultural biomass tends to contain lower lignin (%) and similar carbohydrate content to that found in most softwoods and hardwoods (McKendry 2002; Lee et al. 2007; Zhu and Pan 2009). Because of the chemical composition (structure and types), agricultural biomass may require less severe pretreatments to reach the same level of enzymatic digestibility when compared with recalcitrant woody biomass such as many softwood species (Chandra et al. 2007).

In the mean time, more efficient and low-cost pretreatments for cellulosic biomass have been widely researched for years, resulting in the development of novel technologies and transformation of existing ones. Some technologies are accompanied by uncertainty for scale-up feasibility. The use of existing and well-known technologies for cellulosic ethanol production brings several potential advantages, including the availability of equipment manufacturers, experienced operators, and lower risks when estimating capital investments. Recent studies have demonstrated the potential use of existing technologies in the pulp and paper industry as pretreatment pathways for producing cellulosic ethanol (Walton 2009; Wang et al. 2009; Zhu et al. 2009; Jin et al. 2010; Wu et al. 2010; Zhu et al. 2010a).

Thermomechanical pulping (TMP), an existing and practiced technology in the pulp and paper industry, can be potentially used as pretreatment of lignocellulosic biomass for ethanol production. Chemical pretreatment such as oxygen delignification and alkaline hydrolysis can be combined with TMP to increase accessible surface area by partially removing hemicelluloses and lignin (Mosier et al. 2005). These chemical pretreatment processes coupled with steam and mechanical pretreatments (Chemithermo-mechanical pulping, CTMP) can be used to boost pulp characteristics that result in more efficient enzymatic hydrolysis (Wang et al. 2009; Zhu et al. 2009). The use of TMP and CTMP as pretreatments for cellulosic biomass can be the basis for the repurposing of old and closed TMP mills as the demand for newsprint paper continues to decline. TMP yield (pulp yield) is generally higher compared to other pulping pathways (Smook 2003).

Few studies regarding the use of TMP or CTMP as pretreatments on agriculture biomass for cellulosic ethanol production are available. Most of the available information utilizing these technologies aims to produce pulp and paper, using wood as a raw material, with a clear concern paid to the high energy consumption by the process (Zhu and Pan 2009; Zhu et al. 2010b). Previous studies suggest that thermomechanical pulping is a potential pretreatment for cellulosic ethanol production because the two stages (steaming and refining) separate raw materials into individual fibers and bundles such that "the accessibility to enzyme attack is already much enhanced" (Overend et al. 1987), and also the fact that the addition of chemicals will develop more efficient fractionations. Previous studies have shown mechanical pulp from softwood to be resistant to enzymatic hydrolysis (Mooney et al. 1998). The use of TMP and CTMP in less recalcitrant feedstock (agriculture biomass) may lead to new biochemical conversion pathways, while the economics of the biorefinery can benefit from the use of existing assets in closed facilities, experienced labor in the region, as well as operating with a proven technology in place.

Objective

This paper considers the potential use of thermomechanical pulping, an existing technology in the pulp and paper industry, as a pre-treatment for agricultural biomass for cellulosic ethanol production. The main objective of this paper is to identify process conditions in thermomechanical pulping affecting carbohydrate conversion during enzymatic hydrolysis of agricultural biomass.

MATERIALS AND METHODS

Biomass

Three types of agricultural cellulosic biomass were studied: corn stover (CS), wheat straw (WS), and sweet sorghum bagasse (SS). Corn stover (obtained from Indianola, Iowa) and wheat straw (obtained from Ontario, Canada) have been kept under enclosed storage conditions for two years. Sweet sorghum bagasse was obtained from a local farming operation in Garland, North Carolina. Sweet sorghum bagasse was collected after the squeezing operation (in farm, Fall 2009) and was air dried using fans for about 3 weeks (at NC State University).

Biomass was cut at 2.5 to 3.8 cm length, and then soaked (Fig. 1). After soaking, the biomass was centrifuged to a solids content of ca. 31%. The biomass was then fed into the thermo-mechanical pulping unit with the conditions described in the thermo-mechanical pulping section. The pulp obtained was then sent directly "as is" to enzymatic hydrolysis with the conditions described in the enzymatic hydrolysis section. Filtrate from enzymatic hydrolysis was collected and analyzed to measure monomeric sugars (see sugar quantification section). The overall process is illustrated in Fig. 1.

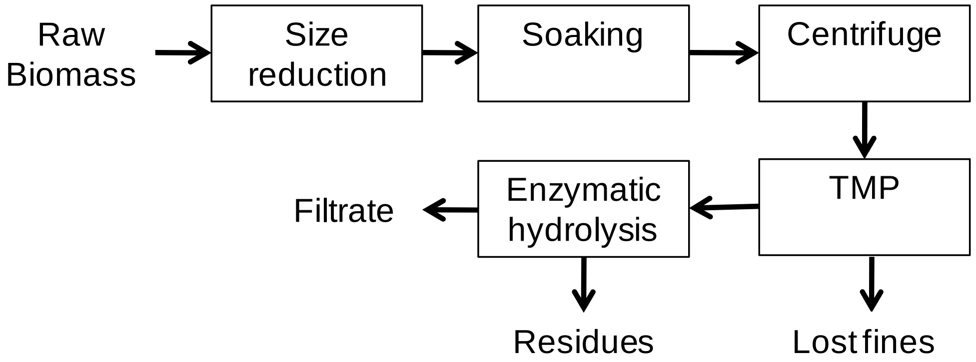

Figure 1. Process flow of thermo-mechanical pulping pretreatment

Soaking, Thermomechanical Pulping and Yield

All three biomass types were soaked in water (at room temperature) the night before at a liquid-to-biomass ratio of 10. After 12 hours soaking, the biomass was centrifuged for 15 min (to a solids content of ca. 31%). Other exploratory pre-treatments were investigated included soaking in acetic acid (9% charge based on dry mass after centrifugation). Pretreatments were carried out in a TMP unit at the Department of Forest Biomaterials, NC State University. For the three raw materials, experiments were carried out at 160 ºC and 170 ºC for 15 minutes (steaming – residence time). When the residence time was reached, biomass was passed through a refiner with a disk gap of 0.1524 millimeters (only one refining pass). Other treatments (for wheat straw only) included comparison of different residence times (15 and 30 min) and refining at lower refiner disk gap (0.0508 millimeters). TMP yield was estimated by dividing the total output (pulp and material stuck in the steam vessel, oven dry equivalents) by the total input of biomass (oven dry weight equivalent).

Chemical Composition

Contents of Klason lignin and acid-soluble lignin of the pretreated substrates were determined according to the procedures of NREL chemical analysis and testing standard procedures (NREL 2005). Structural sugars in the pretreated substrates were determined using the filtrate of Klason lignin measurements. The supernatant obtained after Klason lignin determination was filtered using a 0.2 μm nylon filter before sugar analysis. Monomeric sugars from the Klason lignin hydrolyzate were analyzed using an ion chromategraphy (IC) system (ICS 3000, Dionex Corp., CA), equipped with an auto sampler, a GP40 gradient pump, a high-pH anion exchange column (CarboPac TM Pal, Dionex Corp., Billerica, MA), as described by Lee et al. (2010). The column was eluted with deionized water at a flow of 1.0 mL/min. The column was re-conditioned using 0.2 M NaOH after each analysis. Baseline stability and detector performance were optimized by adding 0.4M NaOH to a post-column at 0.1 mL/min. Sugar content was quantified by comparison with standards and all standards and samples contained an internal sugar standard of fucose(L-(-) Fucose (F2252, Sigma, Saint Louis, MO). The cellulose and hemicellulose content are reported on a polymeric basis after correcting sugar analysis data, using hydration correction coefficients (Iyer and Lee 1999; Lee et al. 2010).

Enzymatic Hydrolysis and Sugar Quantification

Enzymatic hydrolysis was performed using a cocktail of three enzymes at a ratio of 1:0.3:0.3 (v/v), as suggested by the enzyme provider. The components of the mixture were, respectively: cellulase from *Tricoderma reesei* (NS-50013), β-glucosidase from Aspergillus Niger (NS-50010), and xylanase (NS-50014) provided by Novozymes (Franklinton, NC). Cellulase dosages were 5, 10, and 20 FPU/g biomass. Incubation was performed in 100 mL of 100 mM citrate buffer (pH 4.8) at a 5% (w/v) solids loading (total solids content). The pulp and enzymes were incubated in a hot air shaker at 50 °C for 48 h, 180 rpm. Enzymatic conversion was determined in duplicate by measuring the monomeric sugars in the filtrate using high-performance liquid chromatography (HPLC).

The filtrate obtained from the enzymatic hydrolysis was boiled for 15 min and then centrifuged for 30 min to denature the enzymes, prevent further hydrolysis, and separate solids from the liquid. The solution was filtered using a 0.2 μm filter and analyzed by HPLC (Dionex, Sunnyvale, California). Sugars were separated through a Shodex SP0810 column at the temperature of 80 °C. Water was used as the eluent with the flow rate of 0.4 mL/min. A refractive index detector was used to quantify arabinose, galactose, glucose, xylose, and mannose in the samples.

RESULTS AND DISCUSSION

Biomass Composition

Cellulose, hemicelluloses (both in polymeric form), lignin, and ash contents for each of the feedstock types are presented in Table 2. Higher carbohydrate and lignin contents were found in wheat straw, followed by corn stover, and then sweet sorghum. Table 1 also shows the pretreated pulp composition for each of the materials and pretreatment conditions. The balance for each of the feedstocks and pretreated pulps does not include extractives, uronic acid, acetyl groups, protein or sugar degradation products (furfural and hydroxymethylfurfural (HMF)). This explains the lower material balance closure, especially for the raw material that was analyzed prior to removing extractives (sweet sorghum bagasse = SS raw, corn stover = CS raw and wheat straw = WS raw). Lignin content is higher in wheat straw, followed by corn stover and sweet sorghum (original raw material).

Thermomechanical Pulping Yield

The yields of TMP pretreated sweet sorghum (SS), corn stover (CS), and wheat straw (WS) at two temperature conditions, 160 and 170 ºC, with 15 min steaming (residence time) are presented in Fig. 2. In general, pulp yield decreased when biomass was pretreated at the higher temperature (170 ºC). For a condition of 160 ºC, TMP yields for the three biomasses ranged from ca. 90.0 % to 95.4%. At 170 ºC, TMP yields ranged from 81.1% to 90%. Those feedstocks with higher lignin content (wheat straw and corn stover) had higher TMP yield.

Table 1. Sugars in Oligomeric Form, Lignin and Ash Content for each Feedstock and Pretreated Pulp

Raw material and Pulp	**Sugars**							**Lignin, Ash, Balance**				
	Glucan	Arabinan	Galactan	Xylan	Mannan	Total Hemicellulose	Total carbohydrate	Klason lignin	Acid soluble	Total Lignin	Ash	Balance
SS raw	37.5	1.3	0.1	15.0	0.5	16.9	54.4	11.9	1.5	13.3	3.5	71.2
CS raw	32.7	2.8	1.0	19.1	0.4	23.3	56.0	15.2	1.5	16.7	5.2	77.8
WS raw	33.3	2.7	0.6	20.9	0.5	24.6	57.9	16.0	2.4	18.4	9.8	86.0
SS EF	40.3	2.0	0.4	22.1	0.3	24.8	65.1	15.4	2.3	17.7	3.1	85.9
CS EF	36.1	3.5	1.4	22.9	0.4	28.2	64.3	13.9	1.6	15.5	5.2	84.9
WS EF	35.4	2.8	0.5	22.4	0.4	26.2	61.6	16.6	2.0	18.6	7.2	87.4
SS160 15 min	38.3	1.5	0.5	21.5	0.3	23.8	62.1	21.2	1.7	22.9	1.5	86.4
SS170 15 min	38.7	1.4	0.6	21.5	0.5	24.1	62.8	22.2	2.0	24.2	1.0	87.9
CS160 15 min	36.2	2.7	1.0	22.1	0.4	26.1	62.4	21.2	1.9	23.0	4.3	89.7
CS170 15 mn	32.1	3.1	1.3	21.9	0.4	26.8	58.8	18.6	1.7	20.3	4.1	83.2
WS160 15 min	36.3	2.6	0.6	22.7	0.4	26.2	62.4	19.0	2.5	21.6	7.7	91.7
WS170 15 min	36.5	2.4	0.9	22.4	0.2	26.0	62.5	20.7	2.8	23.5	6.3	92.3
WS 160 30 min	37.0	2.6	1.0	22.5	0.0	26.1	63.1	19.8	1.6	21.4	6.1	90.7
WS 170 30 min	39.5	2.1	0.9	21.8	0.0	24.8	64.4	19.7	3.1	22.7	6.4	93.5
WS 170C-9% Acetic acid-15m	39.3	1.6	0.8	19.1	0.0	21.5	60.8	21.0	2.8	23.9	6.2	90.8
WS 170C-9% Acetic acid-30min	42.1	0.9	0.6	16.1	0.0	17.6	59.7	20.7	1.5	22.2	6.0	87.9

All values in % w/w. Legend: SS = sweet sorghum bagasse, CS = corn stover, WS = wheat straw, raw = original biomass, EF = extractive free, 160 = 160 ºC, 170 = 170 ºC), 15 min = 15 min steaming residence time, 9% Acetic acid based on oven dry biomass.

Figure 3 shows TMP yield of pretreated wheat straw with the residence time base (15 min) and at 30 min. Other data include treatments with wheat straw soaked with acetic acid and refined at a lower refiner gap (0.0508 millimeters) compared to the refiner disk gap base (0.1524 millimeters). Refining at lower gap space (0.0508 millimeters) did not considerably affect TMP yield. However, wheat straw soaked with acetic acid resulted in the lowest TMP yield (ca. 70 to 75%).

From Table 1 it can be observed that wheat straw treated with acetic acid had major degradation of the hemicelluloses. It is apparent that acetic acid treatment caused higher degradation of the pulp. A mild concentration of acetic acid should be considered to understand tradeoffs between pulp yield and total carbohydrate recovery.

Though a good effort was made to collect all material from the pulping process, a portion of fines were lost inside the reactor, screw feeder and refiner. The resulting pulp (slurry) had a solids content ca.15 to 20%. All the pulp was weighed and the solids content was determined to estimate total dry mass and pulp yield.

Effect of Temperature, Steaming-residence Time and Refiner Gap in Carbohydrate Recovery

A series of treatments were performed to understand the effect of temperature, steaming-residence time, and refiner disk gap on the carbohydrate recovery obtained from the enzymatic hydrolysis filtrate.

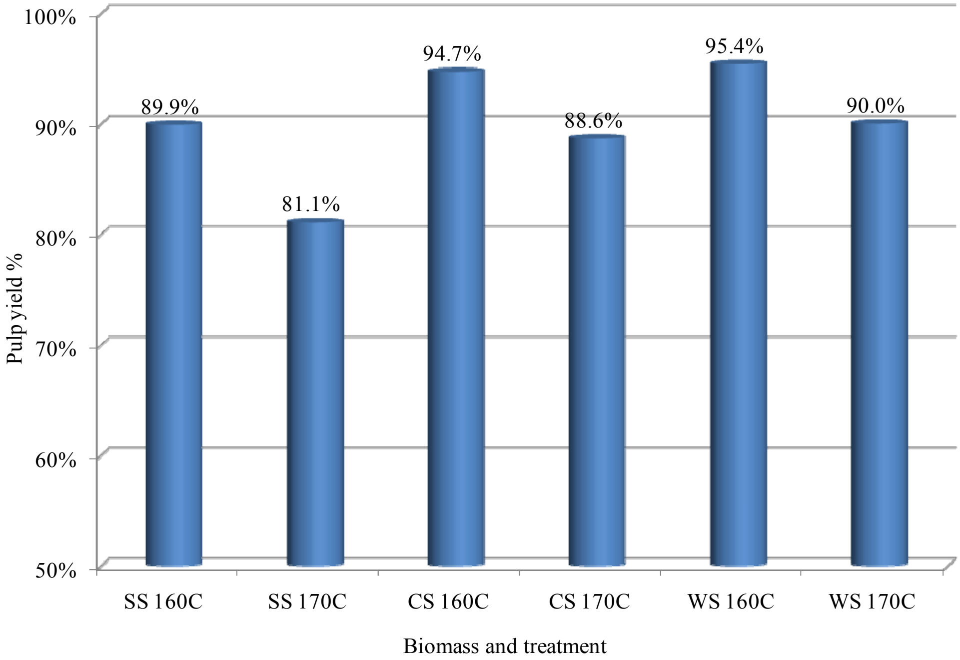

Figure 2. TMP yield for Sweet sorghum bagasse (SS), corn stover (CS) and wheat straw (WS) at 160 and 170 ºC

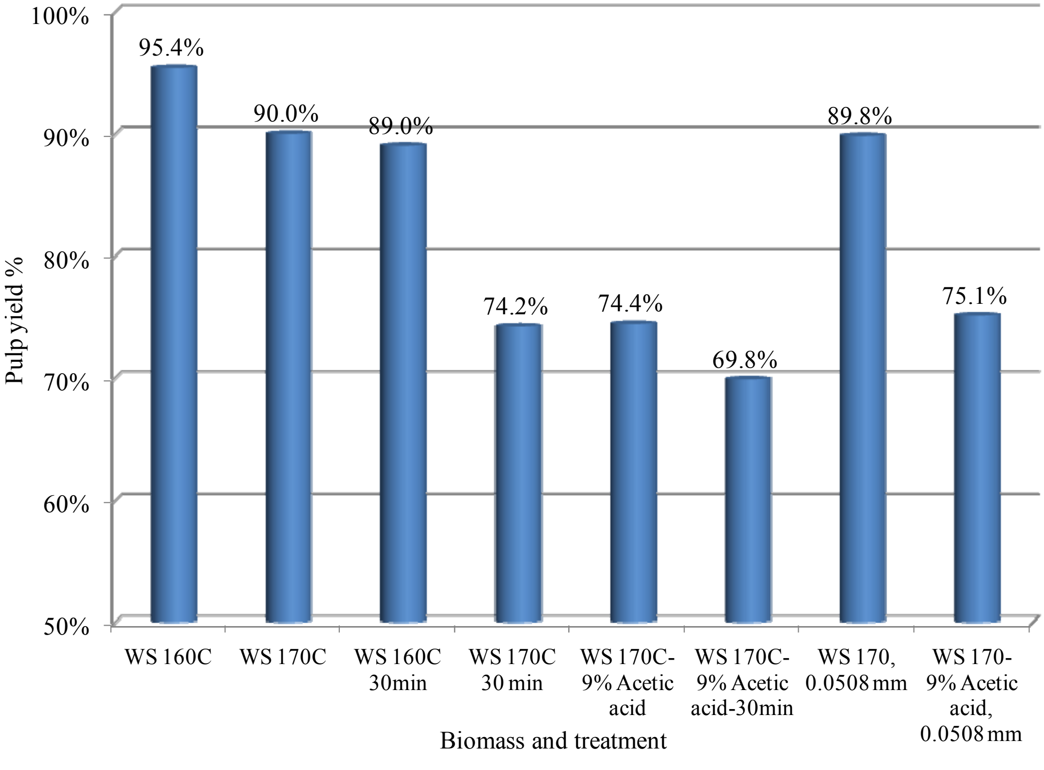

Figure 3. TMP yield of wheat straw for different conditions

Temperature

Figures 4 through 6 show carbohydrate recovery (polymeric form based on total carbohydrate in the enzymatic hydrolysis filtrate (monomeric sugar) compared to the original amount found in the raw biomass), with enzymatic hydrolysis loading of 5, 10, and 20 FPU/g of OD pulp, for each of the three biomass pretreated at 160 ºC and 170 ºC. For all cases, biomass pretreated at 170 ºC showed higher carbohydrate conversion to monomeric sugars (enzymatic hydrolysis yield) when compared to 160 ºC. Carbohydrate recovery was higher at higher enzyme loading. From Fig.4, the data indicate that sweet sorghum bagasse (SS) biomass pretreated at 170 ºC and hydrolyzed at 5 FPU had higher carbohydrate digestibility than SS biomass pretreated at 160 ºC and hydrolyzed at 10 FPU. The same trend can be observed for corn stover (Fig. 5) and wheat straw (Fig. 6), showing the beneficial effect of higher temperature in substrate conditioning to enhance enzymatic digestibility. Wheat straw presented the lowest carbohydrate conversion of the three feedstocks. As reported elsewhere, when temperature increased above 150-180 ºC, components of the lignocellulosic biomass started to solubilize. First the hemicellulose, followed shortly by lignin began to solubilize, resulting in a more accessible lignocellulosic substrate amenable to enzymatic hydrolysis (Bobleter 1994; Garrote, Dominguez et al. 1999; Hendriks and Zeeman 2009).

In order to provide further information on carbohydrate conversion, and to isolate the effect of material loss during the TMP process, Table 2 provides data on glucan and xylan digestibility based on both original raw material and on substrate. Higher digestibility on both glucan and xylan (based on substrate) were found at the higher enzyme loading (20 FPU). Also, higher digestibly was achieve in pulps treated at 170 ºC. For sweet sorghum, no significant difference in xylan conversion was found between 10 and 20 FPU (at both temperature conditions 160 ºC and 170 ºC). The other two feedstocks experienced more significant xylan hydrolysis at higher enzyme loading.

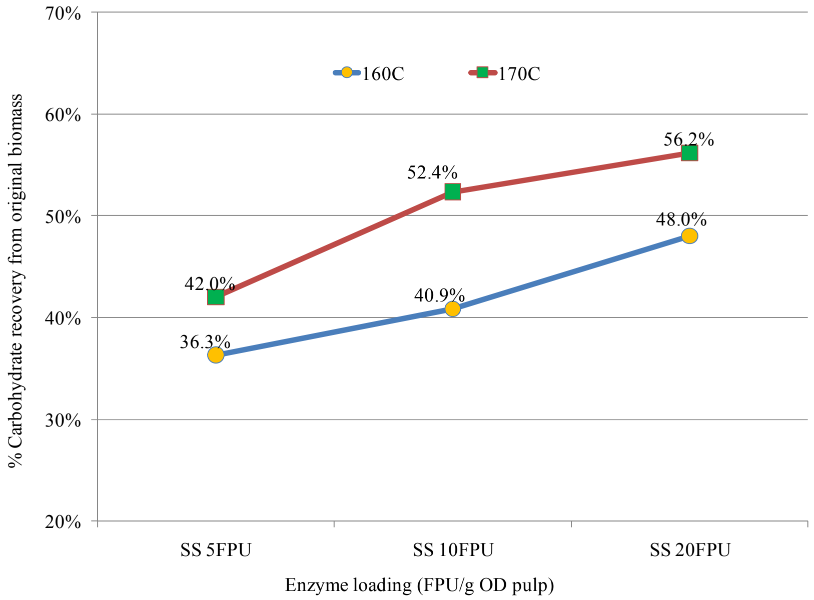

Figure 4. Total carbohydrate conversion from original sweet sorghum biomass, TMP pulp (160 and 170 ºC) at 5, 10 and 20 FPU

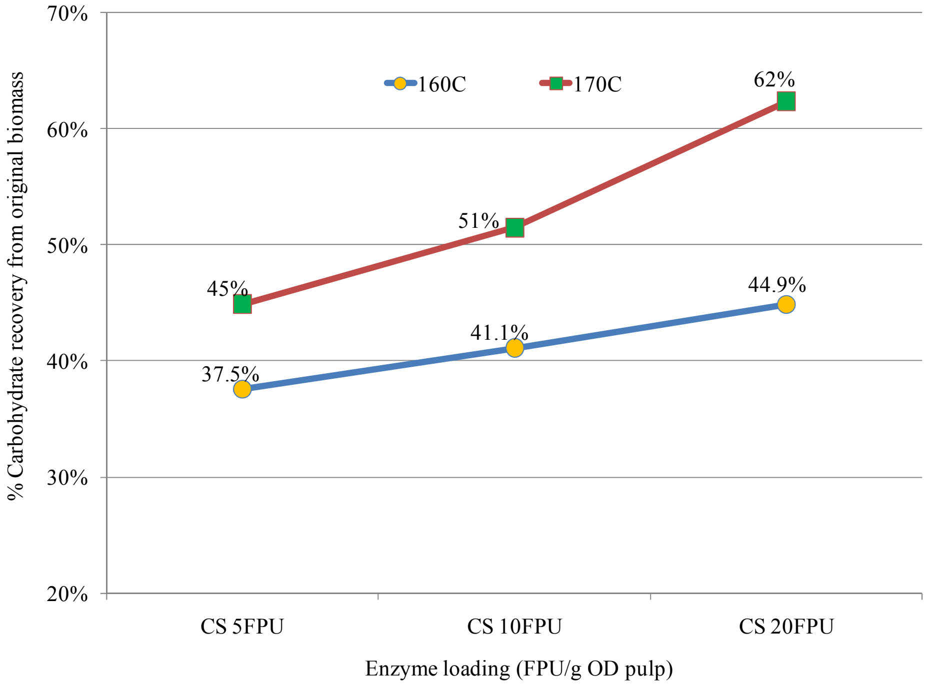

Figure 5. Total carbohydrate conversion from original corn stover biomass, TMP pulp (160 and 170 ºC) at 5, 10 and 20 FPU

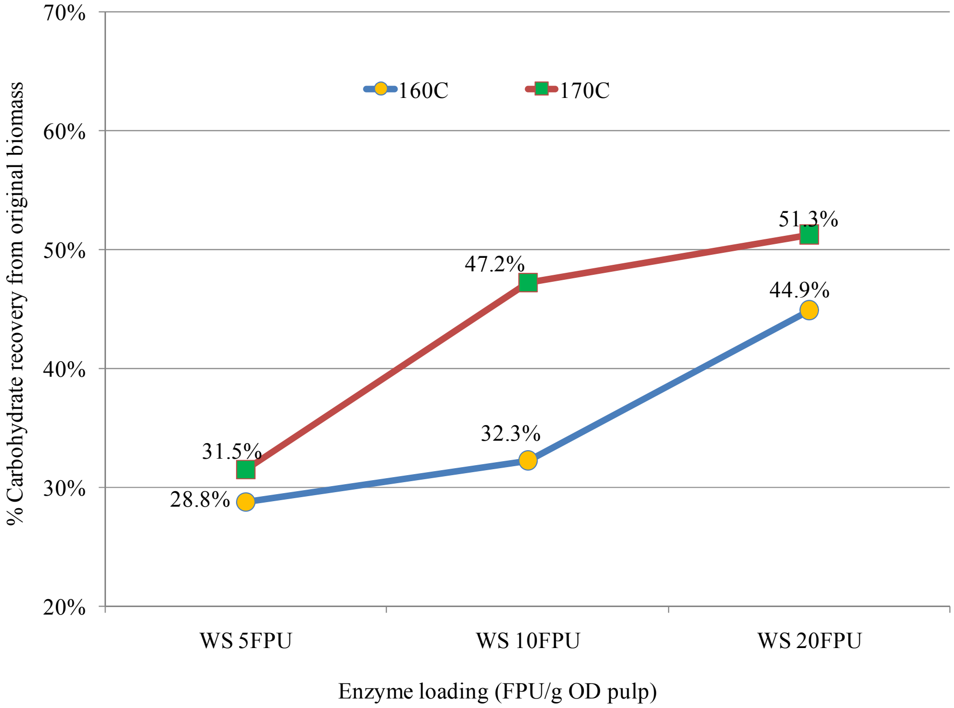

Figure 6. Total carbohydrate conversion from original corn wheat straw biomass, TMP pulp (160 and 170 ºC) at 5, 10 and 20 FPU

Table 2. Average Biomass Composition for Several Cellulosic Feedstocks

Component	Based on original biomass, %w/w						Based on substrate, %w/w					
	Glucan			Xylan			Glucan			Xylan		
Enzyme loading, FPU	5	10	20	5	10	20	5	10	20	5	10	20
Pretreatment												
SS160 15 min	29.4	34.0	42.2	53.6	58.3	61.4	32.0	37.0	45.9	41.5	45.2	47.5
SS170 15 min	37.0	47.5	52.9	54.2	64.9	67.2	44.2	56.8	63.2	46.5	55.7	57.7
CS160 15 min	39.4	43.5	46.5	36.4	40.1	44.9	37.6	41.5	44.3	33.3	36.7	41.0
CS170 15 mn	44.4	50.7	56.2	50.9	58.7	75.4	51.1	58.2	64.6	50.2	57.8	74.4
WS160 15 min	24.5	29.5	44.2	35.5	38.6	47.3	23.5	28.3	42.5	34.2	37.2	45.6
WS170 15 min	28.6	45.2	48.2	37.6	52.8	57.0	29.0	45.8	48.9	38.9	54.7	59.1

Residence Time

In order to understand the effect of residence time on carbohydrate recovery, wheat straw was pretreated at 15 min and 30 min (steaming) residence time, with temperatures of 160 and 170 ºC. Enzymatic hydrolysis took place at enzyme loadings of 5 and 10 FPU. Residence time of 30 min (steaming) considerably increased carbohydrate recovery in wheat straw pretreated at 160 ºC (Fig. 7).

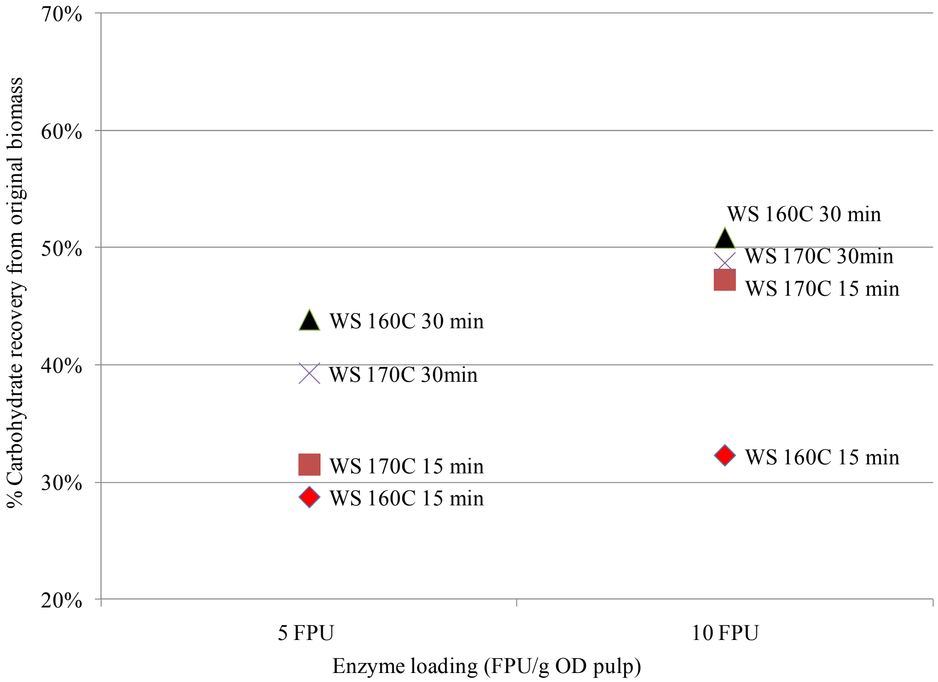

Figure 7. Effect of residence time (15 and 30 min) in carbohydrate recovery in pretreated wheat straw at 160 and 170 ºC

Carbohydrate recovery at 10 FPU was very similar for WS 170 ºC 15 min, WS170 ºC 30 min, and WS160 ºC 30 min. Longer residence times should result in higher solubilization of hemicellulose and lignin, as well as the generation of sugar degraded byproducts (not measured in this study). A combination of longer residence time and conditions within the range of lignin glass transition temperature may result in lignin coating the fiber (Li et al. 2006; Zhu et al. 2009) and thus reducing the accessible surface area for enzymatic hydrolysis (Mosier et al. 2005; Hendriks and Zeeman 2009). Table 3 shows glucan and xylan digestibility, based on both original raw material and on substrate, at the same enzyme loadings. For both glucan and xylan, higher conversions were found in treatments with longer residence time (30 min).

Table 3. Average Biomass Composition for Several Cellulosic Feedstocks

	Based on original biomass, %w/w				**Based on substrate, %w/w**			
Component	Glucan		Xylan		Glucan		Xylan	
Enzyme loading, FPU	5	10	5	10	5	10	5	10
Pretreatment								
WS160 15 min	24.5	29.5	35.5	38.6	23.5	28.3	34.2	37.2
WS170 15 min	28.6	45.2	37.6	52.8	29.0	45.8	38.9	54.7
WS 160 30 min	40.7	48.9	50.5	56.1	41.2	49.4	52.7	58.6
WS 170 30 min	34.8	47.4	48.1	53.8	39.5	53.8	62.1	69.4

Acetic Acid Soaking

Wheat straw biomass soaked with acetic acid prior TMP treatment (170 ^{o}C and residence time of 15 and 30 min steaming) showed higher carbohydrate recovery when compared to wheat straw at 170 ^{o}C (15 min steaming) soaked with water. An increase in carbohydrate recovery of almost 50% at 5 FPU and about 30% at 20 FPU (Fig. 8) was observed. The effect of combining acid and thermal treatments on carbohydrate recovery is consistent with reported data in the literature. External acids work as catalytic agents in the solubilization of the hemicellulose, providing a more suitable substrate for enzymatic hydrolysis (Gregg and Saddler 1996; Mosier et al. 2005; Hendriks and Zeeman 2009; Wang et al. 2009; Zhu et al. 2009). It is important to remember that acetic acid impregnation prior TMP process reduced overall TMP yield; mild acetic acid charge treatments should be taken into consideration to understand tradeoffs between carbohydrate recovery and pulp yield. Table 4 shows glucan and xylan digestibility, based on both original raw material and on substrate, at the same enzyme loadings. Acetic acid seems to boost xylan conversion (based on substrate data); major difference in xylan conversion was found at lower enzyme loading (between the three treatments); and lower difference was found in the acetic acid pretreatments at 15 min and 30 min residence time at higher enzyme charge. Despite the higher glucan and xylan conversion in the acetic acid pretreated pulps, total carbohydrate conversion from original biomass was affected by the lower pulping yield.

Table 4. Average Biomass Composition for Several Cellulosic Feedstocks

	Based on original biomass, %w/w						**Based on substrate, %w/w**					
Component	Glucan			Xylan			Glucan			Xylan		
Enzyme loading, FPU	5	10	20	5	10	20	5	10	20	5	10	20
Pretreatment												
WS170 15 min	28.6	45.2	48.2	37.6	52.8	57.0	29.0	45.8	48.9	38.9	54.7	59.1
WS 170C-9% Acetic acid-15min	45.7	57.1	62.8	52.4	55.6	61.8	52.0	65.0	71.5	77.1	81.7	90.8
WS 170C-9% Acetic acid-30min	56.7	73.4	77.3	46.1	53.7	54.6	64.2	83.0	87.5	85.7	87.0	91.3

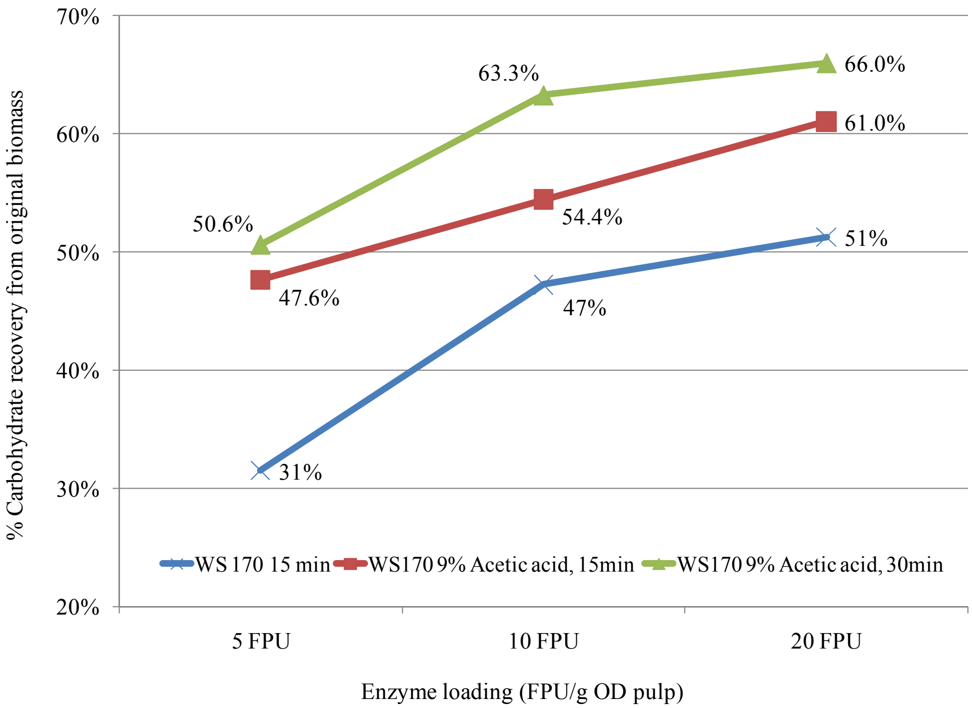

Figure 8. Effect of acetic acid soaking (9% OD charge) in carbohydrate recovery in pretreated wheat straw at 170 °C

Refiner Disk Gap

The refining of pulp has been shown to produce fibers with a higher degree of fibrillation (Smook 2003; Nakagaito and Yano 2004), resulting in higher exposed surface area for enzymatic attack (Mosier et al. 2005; Hendriks and Zeeman 2009). A reduced gap during refining should further increase surface area (Zhu et al. 2010b). Furthermore, high intensity refining would result in higher energy consumption and more fines produced. In order to understand the effect of refiner gap space on total carbohydrate recovery after enzymatic hydrolysis, one set of treatments was considered to reduce the gap distance between the plates of the refiner from 0.1524 millimeters (base gap) to 0.0508 millimeters. When the refiner plate gap was reduced to 0.0508 millimeters, enzymatic hydrolysis increased by 20 to 25% (at 5 FPU and 10 FPU enzyme loading respectively, treatment WS170 ºC 15min, 0.0508 millimeters) when compared to the base line treatment WS 170 ºC, 15min, 0.1524 mm (Fig. 9). Another set of treatments included a combination of reduced refiner plate's gap (0.0508 millimeters) and soaking with acetic acid. Results showed no difference in enzymatic hydrolysis conversion when compared with the treatment WS170 ºC 15min, 0.0508 millimeters (no acetic acid soaking). This may suggest that, under the conditions tested, a narrower gap between refiner plates achieved substrate digestibility similar to those treatments with acetic acid soaking. Optimum conditions on both refining intensity and acetic acid charge need further research. Table 5 shows glucan and xylan digestibility, based on both original raw material and on substrate, with the same enzyme loadings. Glucan and xylan conversions were considerably higher in the acetic acid treatment. Refiner energy consumption was not evaluated.

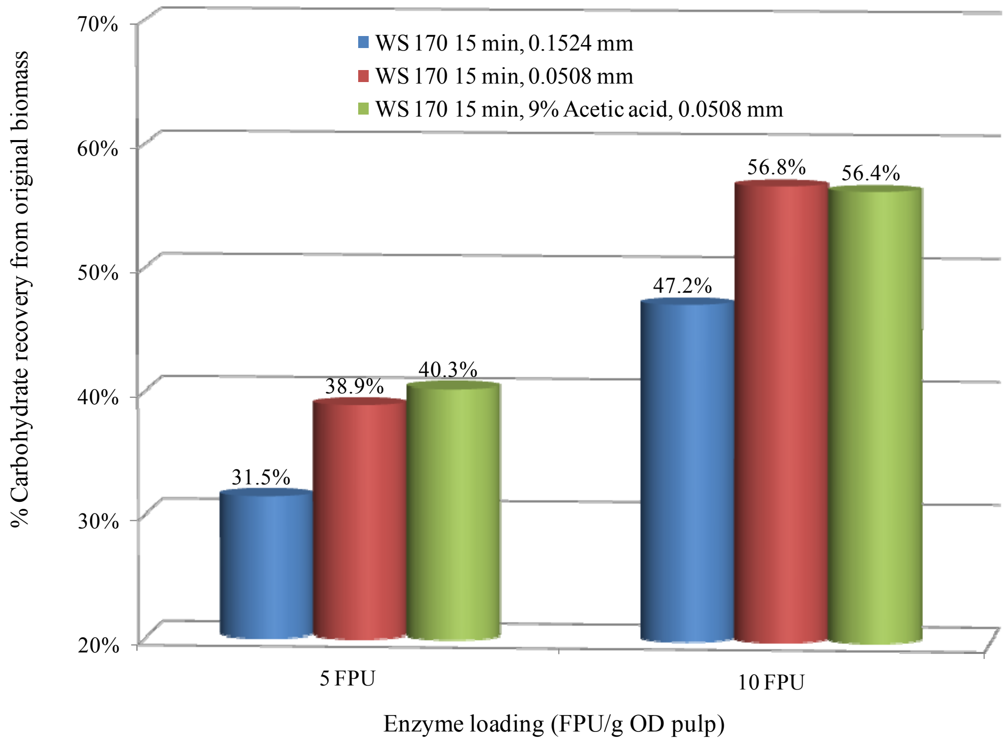

Figure 9. Effect of refiner gap in carbohydrate recovery in pretreated wheat straw at 170C

Table 5. Average Biomass Composition for Several Cellulosic Feedstocks

	Based on original biomass				Based on substrate			
Component	Glucan		Xylan		Glucan		Xylan	
Enzyme loading, FPU	5	10	5	10	5	10	5	10
Pretreatment								
WS 170C-15min-0.1524 mm	28.6	45.2	37.6	52.8	29.0	45.8	38.9	54.7
WS 170C-15min-0.0508 mm	36.0	47.9	44.9	71.0	40.0	53.4	50.0	79.1
WS 170C-9% Acetic acid-15m-0.0508 mm	39.5	52.3	43.7	64.9	52.7	69.7	58.2	86.5

CONCLUSIONS

Results of the study indicate that TMP and CTMP, existing and proven technologies in the pulp and paper industry, can be a potential pretreatment for biomass conditioning for cellulosic ethanol production. Although enzymatic hydrolysis conversions presented in this paper are not as high as other studies looking at different feedstocks and pretreatment technologies (e.g. carbohydrate conversion ca. 80% for other well studied pretreatments (Wu et al. 2010)), it is important to mention that the process is very simple in terms of implementation, while optimal conditions for ethanol production need further research. Higher temperature and residence time can boost substrate susceptibility to enzymatic hydrolysis. Carbohydrate conversion increased from 25% to 40% when the TMP temperature was increased from 160 to 170 ºC. Carbohydrate conversion was relatively similar for the three biomasses under the same pretreatment conditions and enzyme loading. The use of chemicals such as acetic acid in the process and refining at a reduced plate gap increases total carbohydrate recovery from the original biomass, as well as glucan and xylan digestibility (based on substrates). There is room for improvement with the use of the different chemical treatments and TMP conditions. Energy consumption must be taken into consideration to understand the tradeoff between refining energy consumption, the cost of chemicals, and total carbohydrate recovery.

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