Appl Biochem Biotechnol (2010) 162:1785–1803
DOI 10.1007/s12010-010-8959-3

Effect of Extruder Parameters and Moisture Content of Switchgrass, Prairie Cord Grass on Sugar Recovery from Enzymatic Hydrolysis

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Received: 29 December 2009 / Accepted: 29 March 2010 /
Published online: 27 April 2010

Abstract Research on biomass pretreatment to enhance enzymatic digestibility has been done for more than decades, but a viable continuous pretreatment method needs to be developed. Extrusion has the potential to be a viable continuous pretreatment method. This study investigated the effect of compression ratio (2:1 and 3:1), screw speed (50, 100, and 150 rpm), and barrel temperature (50, 100, and 150 °C) on the sugar recovery from switchgrass (SG) and prairie cord grass (PCG) over a range of moisture contents (15, 25, 35, and 45% wb).The pretreated samples were subjected to enzymatic hydrolysis for sugar recovery measurement. Statistical analyses revealed that a 3:1 screw compression ratio (compared to 2:1) increased glucose recovery by 12% and 8% and combined sugar recovery by 37% and 40% for SG and PCG, respectively. For SG, the highest sugar recovery (45.2%) was obtained at the lowest screw speed (50 rpm) and the highest temperature (150 °C) with moisture content of 15%. The highest glucose, xylose, and combined sugar recovery of 61.4%, 84.3%, and 65.8% were recorded for PCG extruded at a screw speed of 50 rpm and a temperature of 50 °C with a moisture content of 25%. Glycerol and acetic acid were byproducts found in low concentration (0.02–0.18 g/L) for both biomass.

Keywords Biomass · Pretreatment · Switchgrass · Prairie cord grass · Screw speed · Temperature · Extruder

Introduction

The search for a renewable fuel resource has become a global priority, due to declining fossil fuel resources, and economic and environmental concerns. In the US, land use patterns, grain starch supply limitations, and corn productivity have also led to the search for alternate resources for biofuel production. Lignocellulosic biomass appears to be an attractive feedstock

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due to its renewability, positive environmental impacts resulting in no net release of carbon dioxide (CO_2) and very low sulfur content [1]. While storage carbohydrates from grain materials are readily available for enzymatic hydrolysis, structural carbohydrates in lignocellulosic materials are not readily accessible for bioconversion. Lignocelluloses are composed of cellulose, hemicellulose, and lignin, wherein the protective layer provided by lignin hinders biochemical conversion more than any other component. Hence, pretreatment is necessary to open up the structure and make it accessible for enzymes. Pretreatment is the primary, most expensive and inevitable step in converting biomass into biofuels [2].

Several pretreatment strategies, such as dilute acid, alkali, steam explosion, hot water, wet oxidation, and ammonia fiber expansion (AFEX) methods are under investigation. The elevated temperature used in hydrothermal pretreatment usually leads to sugar degradation. Acid or alkali pretreatments promote hydrolysis and improve the yield of glucose recovery from cellulose by removing hemicellulose or lignin. Though dilute sulfuric acid achieves up to 90% hemicellulose yield and an enzymatic hydrolysis yield of glucose over 90% [2], it requires costly construction material, high pressures, neutralization and conditioning of hydrolyzate prior to biological steps, it slows down cellulose digestion by enzymes, and binds enzymes to lignin in a nonproductive way [3–5]. Formation of degradation products and release of natural biomass fermentation inhibitors are other characteristics of acid pretreatment [2]. Sodium hydroxide and other bases are expensive and too difficult to recover and recycle to make them viable for producing fuels and chemicals [3]. AFEX pretreatment simultaneously reduces lignin content and removes some hemicellulose while decrystallizing cellulose. The cost of ammonia and ammonia recovery are critical to AFEX [6]. Literature survey revealed that no perfect technology exists for large-scale biofuel production from biomass. Still there is a need to develop a viable continuous pretreatment method to be used within the bioethanol industry.

Extrusion is a process in which ingredients are subjected to simultaneous heating, mixing, and shearing, resulting in physical and chemical changes as the material is being conveyed along the extruder barrel [7]. The extruder has many advantages such as the ability to provide high shear, rapid heat transfer, and effective and rapid mixing. Because of its adaptability to many different process modifications such as the addition of chemicals or removal of materials, and the application of high pressure and expansion treatment (using steam or other solvents)—all in a continuous process, extrusion has the potential to become a viable biomass pretreatment method. Because extrusion is a continuous pretreatment, it would be practicable and easy to adopt at large-scale production [8]. Moreover, no liquid fraction is produced during extrusion therefore no effluent disposal/treatment problem. A typical extruder consists of three zones: the feed, the transition/compression, and the metering. A feed hopper, screw, barrel, die, and drive are the major components of an extruder as shown in Fig. 1. Screw design strongly influences the optimum temperature profile, work done on the material, and amount of frictional heat generated [9].

Extruder parameters such as compression ratio, screw speed, and barrel temperature are important factors influencing sugar recovery from the biomass. Compression ratio has a direct impact on shear development within the extruder barrel; it affects sugar release from the biomass due to the process of plasticization which occurs in the compression zone. Compression can be achieved in several ways by varying the screw and barrel configuration: by increasing the root diameter, by decreasing the pitch or barrel diameter with constant root diameter, by decreasing the screw pitch in a decreasing barrel diameter, and by introducing restrictions. A gradual decrease of flight depth in the direction of the discharge and a decrease in the pitch in the compression section are the most common ways to achieve compression [10].

Extruder barrel temperature facilitates the melting or softening/plasticizing of the feed. Screw speed is responsible for the rate of shear development and the mean residence time

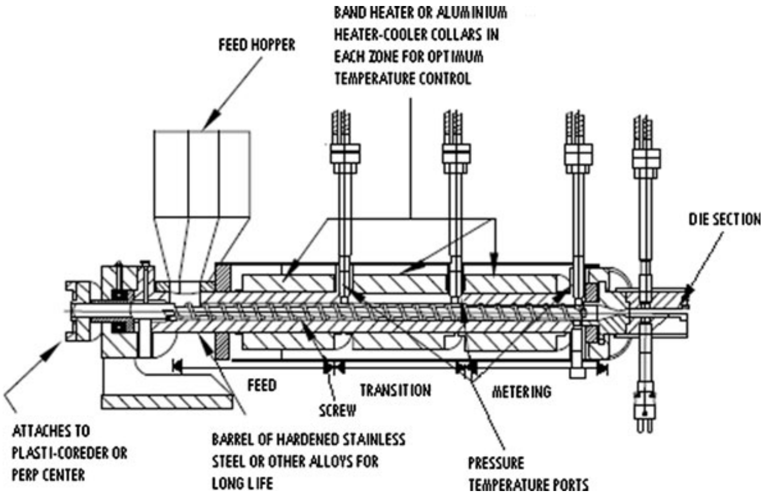

Fig. 1 Sketch showing extruder components

of the feed. Moisture content plays a role in thermal softening by utilizing the barrel temperature and rate of shear development. The high shear developed during extrusion disturbs the biomass structure, thereby increasing access of enzyme to cellulose. Dale et al. [11] reported a 32% increase in enzymatic hydrolysis yield over untreated corn stover when AFEX process was employed using a twin-screw extruder and a 23% increase over material extruded without ammonia. A combination of twin screw extrusion (100 rpm and 100 °C) and alkali pretreatment (NaOH 12% and 70 °C) of miscanthus resulted in 77% delignification, 69% glucose conversion, and 38% of xylose and arabinose conversion [12].

Recently, Lee et al. [8] reported cellulose to glucose conversion of 62.4% from Douglas fir fibrillated using a twin screw extruder at 50 rpm and 40 °C when ethylene glycol was added as a cellulose affinity additive. Because moderate temperature prevents degradation of carbohydrates and oxidation of lignin, formation of potential fermentation inhibitors is avoided [12]. Karunanithy et al. [13] reported that the screw speed and barrel temperature had a significant effect on sugar recovery from different biomasses. These studies showed that there is scope to explore extrusion as one of the feasible pretreatment methods. A literature survey revealed that no comprehensive study combining screw compression ratio, screw speed, temperature, and biomass moisture content has been carried out. Therefore the objectives of this study were: (1) to identify a screw compression ratio for maximum sugar recovery, (2) to investigate the effect of extruder parameters such as screw speed and barrel temperature on sugar recovery, and (3) to understand the influence of the moisture content of switchgrass and prairie cord grass on sugar recovery.

Materials and Methods

Biomass and Composition

This study investigated switchgrass (SG) which is a native perennial grass designated as an energy crop by the US Department of Energy because of its high biomass production capability for producing renewable sources of fuel and electricity [14, 15]. Switchgrass has an average yield of 14.6 t/ha [14] and its bulk density is 67–82 kg/m^3 [16]. Prairie cord grass (PCG), which has a reasonable yield of 10 tons of dry matter/ha [17] and higher bulk density (170–197 kg/m^3), was also selected for this study. SG (Sunburst) and PCG (Red river) obtained from a local farm were ground in a hammer mill (Speedy King, Winona Attrition Mill Co, MN, USA) using a 4-mm sieve for further pretreatment. The ground biomass was stored in a cylindrical container at room temperature before extrusion pretreatment, whereas the pretreated samples were stored in a freezer. The moisture content of the biomass samples was determined as described by Sluiter et al. [18] in NREL/TP-510-42621. The moisture content of the ground biomasses was adjusted to 15, 25, 35, and 45% (wb) by adding water and equilibrating overnight to determine the effect of moisture content on sugar recovery. The compositional analysis of SG and PCG such as glucose, xylose, arabinose, lignin, and ash content were carried out in triplicate as outlined by Sluiter et al. [19, 20] using a muffle furnace and high-performance liquid chromatography (HPLC).

Extrusion Pretreatment

Extrusion was performed in a single screw extruder (Brabender Plasti-corder Extruder Model PL2000, Hackensack, NJ, USA), with a barrel length to screw diameter ratio (*l/d*) of 20:1. In order to understand the influence of screw compression ratios, screws with 3:1 and 2:1 compression ratio were used in this study. The screws were identical except for the depth of the discharge flight at the screw end. There was a constant, continuous taper (decrease of flight depth) from the feed to the discharge, 3.81–1.27 and 3.81–1.90 mm for the screws with compression ratios of 3:1 and 2:1, respectively. The single screw extruder was fitted with a 7.5 hp motor, which had a provision for adjusting the screw speed from 0 to 210 rpm. The extruder screw speed was maintained at 50, 100, and 150 rpm during various pretreatments. The extruder barrel had provisions to control the temperature of the feed and transition zones in both the barrel and die sections. The transition zone and die section temperature was maintained at 50, 100, and 150 °C during the extrusion of the biomass samples. The extruder barrel temperature and screw speed were controlled by a computer connected to the extruder. Extruder feeding was done manually. About 500 g of biomass was extruded under each pretreatment condition, divided into two batches, and considered replicates.

Enzymatic Hydrolysis

Enzymatic hydrolysis of pretreated samples was carried out using a 0.1 M, pH 4.8 sodium citrate buffer for 72 h at 50 °C and 150 rpm as described by Selig et al. [21] in NREL/TP-510-42629. Based on the literature survey and a previous study [22], the amount of cellulase (NS50013, activity 70 FPU/g) enzyme was chosen as 15 FPU/g of dry matter. The ratio of cellulase to β-glucosidase (NS50010, activity 250 CBU/g) was maintained at 1:4 based on an earlier study [22]. All these enzymes were provided by Novozymes. After hydrolysis, the samples were kept in boiling water for 10 min to inactivate the enzyme activity. The supernatant was centrifuged and then frozen twice before injection into the HPLC to remove the impurities, which increased the pressure in the HPLC. Soluble sugars and byproducts were quantified using the HPLC (Agilent Technologies, Santa Clara, CA; Bio-Rad Aminex 87H column, Hercules, CA, USA) with a mobile phase of 0.005 M H_2SO_4 at a flow rate of 0.6 mL/min at 65 °C and a sample volume of 20 μL, as mentioned by Sluiter et al. [23] in NREL/TP-510-42623. Ground SG and PCG were also subjected to enzymatic hydrolysis and analyzed as the control. The sugar concentration (g/L) obtained from the chromatogram was divided by the dry weight of the biomass taken for enzymatic hydrolysis in order to determine the percentage of different sugars. The sugar recovery reported in this paper using Eqs. 1 and 2 was after the enzymatic hydrolysis of the pretreated samples. Instead of separately reporting the amount of arabinose, the arabinose value was added to glucose and xylose for the respective treatment combination and reported as combined sugar. Acetic acid and glycerol were the byproducts found in the pretreated samples, and their concentration was reported in gram per liter.

$$Y_i = \frac{S_{ip}}{S_{ir}} * 100 \quad (1)$$

$$Y_c = \frac{\sum S_{ip}}{\sum S_{ir}} * 100 \quad (2)$$

Y_{i} individual sugar recovery, %
Y_c combined sugar recovery, %
S_{ip} individual sugar obtained in hydrolysate of pretreated samples after enzymatic hydrolysis through HPLC
S_{ir} individual sugar from raw material

Statistical Analysis

The moisture balanced SG and PCG were extruded using two different screw compression ratios (3:1 and 2:1), three different screw speeds (50, 100, and 150 rpm), three barrel temperatures (50, 100, and 150 °C), and four moisture content levels of the biomass (15, 25, 35, and 45% wb). This resulted in a full factorial design of 72 treatment combinations per biomass (2×3×3×4=72). Each treatment run was divided into two batches, and the samples collected were considered replicates, which resulted in 144 observations/biomass. The data were analyzed with Proc GLM procedure to determine the main, interaction, and treatment combination effects in SAS 9.1 (SAS Institute, Cary, NC, USA) using a type I error (α) of 0.05.

Results and Discussion

Composition of Switchgrass and Prairie Cord Grass

The average glucose, xylose, arabinose, lignin, and ash content (with standard deviation) on dry matter basis (%) of SG and PCG were presented in Table 1. In general, glucose is referred to as cellulose, and xylose, arabinose, galactose, and mannose are combined together and termed hemicellulose. The glucose content of switchgrass used in this study was lower than the values reported in the various studies listed in Table 1. The hemicellulose content of SG was comparable to the values reported by Kurakake et al. [24], Suryawati et al. [25] Hu and Wen [26], and Hu et al. [27], and lower than those of DOE [28], Alizadeh et al. [29], and Dale et al. [30]. The lignin content of the switchgrass used in this study was higher than the values reported in the literature. The lower glucose

Table 1 Chemical composition of switchgrass and prairie cord grass.

Biomass	Glucose, %	Xylose, %	Arabinose, %	Lignin, %	Ash, %	Reference
Switchgrass	25.5±5.8	17.4±2.1	4.9±1.2	24.7±2.1	2.9±0.06	Present study
Switchgrass	32.0–33.5	26.0–27.0		17.4–17.8		[28]
Switchgrass	34.1	22.1	3.1[b]			[29]
Switchgrass	41.4	20.9[a]		17.3		[24]
Switchgrass	40.7–44.9	31.4–35.1[a]		5–12	4.6–5.8	[30]
Switchgrass	36.6±0.2	21.0±0.3	2.8±0.1	18.5±0.1	5.0±0.2	[25]
Switchgrass	33.6±1.0	19.3±0.6		21.4±0.8	3.9±0.3	[26, 27]
Prairie cord grass	33.1±0.4	13.5±2.0	1.6±0.6	21.0±0.5	5.6±0.04	Present study
Prairie cord grass	41	33[a]			6.0	[17]

[a] Hemicellulose: sum of xylose, arabinose, galactose, and mannose

[b] Combination of arabinan and galactan

and higher lignin content offer more resistance to any pretreatment method. Boe and Lee [17] reported a higher cellulose and hemicellulose content for PCG than had the PCG used in this study. As observed in the table, PCG had higher glucose and lower lignin than SG. In general, the chemical composition of any biomass varies by geography depending upon the agronomic practices, varieties, and storage period.

Main Effect of Compression Ratio on Glucose Recovery

As evident from the compositional analysis, the major portion of sugar present in the biomass is glucose which is fermentable but not readily available for hydrolysis, thus pretreatment becomes inevitable. Hence, the efficiency of pretreatment can be assessed in terms of glucose recovery. The main effect analysis confirmed that the compression ratio, screw speed, barrel temperature, and moisture content had strong influence (statistically significant) on glucose recovery for both grasses, as is shown in Figs. 2a–d and 3a–d. When screw compression ratio was increased from 2:1 to 3:1, glucose recovery increased ($p=0.0001$) correspondingly by 12% and 37% for switchgrass and prairie cord grass, respectively. The cause for the increase is attributed to the more work put (shearing force and residence time) on the material by the 3:1 screw compression ratio than by the 2:1 screw. Irrespective of the screw compression ratios, prairie cord grass had a higher glucose recovery than did switchgrass, possibly due to the differences in lignin to glucose ratio of prairie cord grass (0.63) and switchgrass (0.97).

Main Effect of Screw Speed on Glucose Recovery

The influence of screw speed on sugar recovery between switchgrass and prairie cord grass differs. The higher screw speed (150 rpm) resulted in a higher glucose recovery for switchgrass, but the lower screw speed (50 rpm) yielded a higher glucose recovery for prairie cord grass. As the screw speed was increased from 50 to 150 rpm, glucose recovery increased by 7.5% (significant) for switchgrass while it decreased by 4% (not significant) for prairie cord grass (Figs. 2c and 3c). As observed in the figures, regardless of screw speed, prairie cord grass had a higher glucose recovery than switchgrass. The screw speed was important for both switchgrass and prairie cord grass; however, the levels of recovery

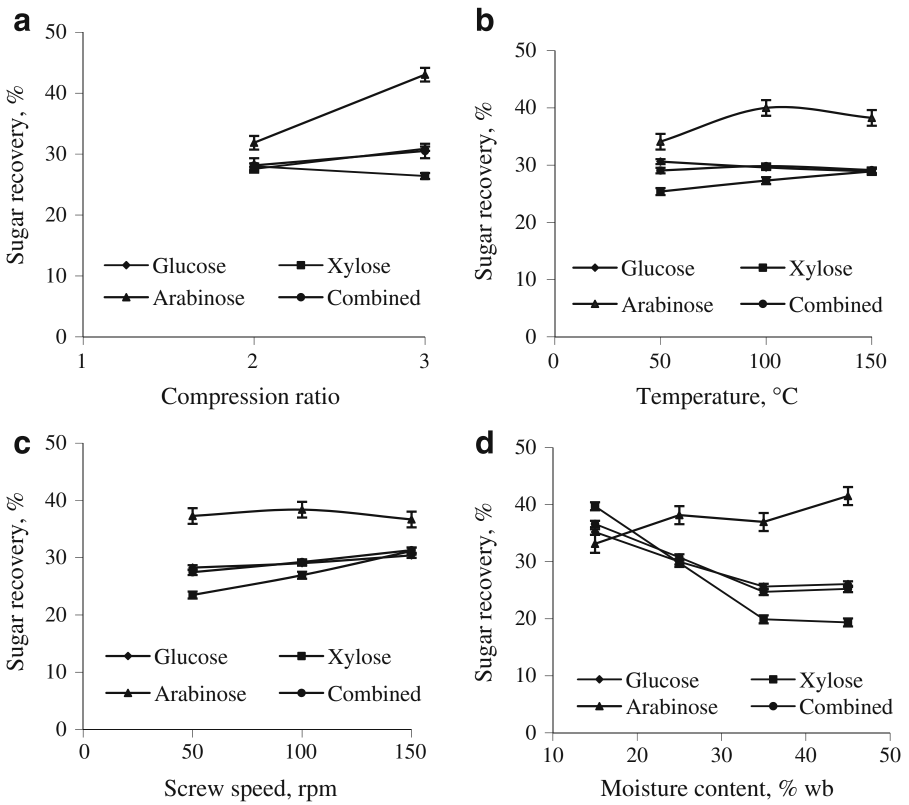

Fig. 2 Main effect analyses of screw compression ratio, temperature, screw speed, and moisture content on sugar recovery from switchgrass

differ. The rate of shear development (high speed) was important for switchgrass, but for prairie cord grass the residence time (low speed) was crucial to obtain high glucose recovery. Inherent characteristics of the grasses also play a role in glucose recovery. The screw speed of 50 and 100 rpm for switchgrass, 100 and 150 rpm for prairie cord grass produced a similar glucose recovery. In general, a higher screw speed resulted in a higher glucose recovery from switchgrass, whereas lower screw speed yielded higher sugar recovery from PCG.

Main Effect of Barrel Temperature on Glucose Recovery

As evident from the main effect analysis (Figs. 2b and 3b), as the barrel temperature was increased from 50 to 150 °C, glucose recovery decreased correspondingly by 6% and 25% for switchgrass and prairie cord grass, respectively. An increase in barrel temperature up to 100 °C had a positive contribution to glucose recovery for both switchgrass and prairie cord grass. The higher temperature contributed to thermal softening of these grasses during their passage through the extruder barrel where a high shear force developed [31]; hence, a low glucose recovery resulted. Moreover, increase in barrel temperature attributed to decrease in viscosity, which resulted in a more flowable material and decrease in residence time

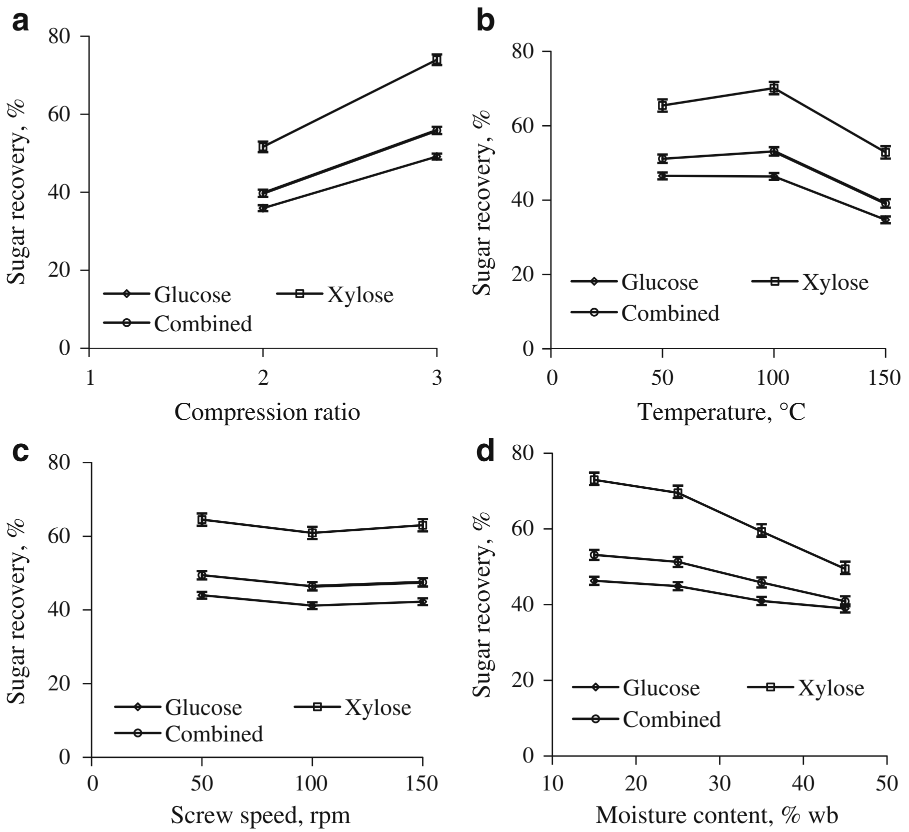

Fig. 3 Main effect analyses of screw compression ratio, temperature, screw speed, and moisture content on sugar recovery from prairie cord grass

ultimately made less disturbance to the biomass cell wall. No significant differences in the glucose recovery between the barrel temperatures of 50 and 100 °C were observed.

Main Effect of Moisture Content on Glucose Recovery

Low moisture content resulted in higher glucose recovery from both biomass, as seen in Figs. 2d and 3d. When moisture content of the biomass was increased from 15% to 45%, glucose recovery decreased (p=0.0001) correspondingly by 26% and 16% for switchgrass and prairie cord grass, respectively. In a single screw extruder, the material is mainly conveyed by friction [32]. When the moisture content was low, friction development was high due to resistance offered by the biomass, and as the moisture content went up, the material became soft and was conveyed without much disturbance to the cell wall. An increase in moisture content resulted in decreasing the friction between the material, screw shaft, and extruder barrel [33]; moreover, high moisture acts as lubricant [34] contributes to slippage.

The glucose recovery from switchgrass was in agreement with the values (55.2% and 43.7%) reported by Kurakake et al. [24] using ammonia water pretreatment (2 mL ammonia water/g, 120 °C) for two varieties of switchgrass. However, glucose recovery from the present study was lower than the glucan conversion (93%) from the ammonia fiber expansion pretreatment of switchgrass at 120 °C for 5 min with 1:1 switchgrass (80% moisture) to ammonia [29] and the dilute acid pretreatment (91.4%) with 1.2% H_2SO_4, 180 °C for 30 s [35]. Similarly, Dien et al. [36] obtained 73–86% glucose from another dilute acid pretreatment of switchgrass. It is well known that alkali removes lignin and that acid solubilizes hemicellulose, thereby increasing the accessibility of the enzymes and resulting in higher glucose recovery. In the case of extrusion pretreatment, hemicellulose was intact after extrusion. The present results (45.3%) were higher than the results of Muthukumarappan and Julson [37] for switchgrass extruded in a twin screw extruder with a moisture content of 20% and a screw speed of 200 rpm. Lee et al. [8] reported a cellulose to glucose conversion of 62.4% from Douglas fir fibrillated using a twin screw extruder at 50 rpm and 40 °C when ethylene glycol was added as a cellulose affinity additive.

Keshwani et al. [38] obtained 32 g/l of reducing sugar per gram of switchgrass pretreated in 3% NaOH using microwave heating (250 W) for 10 min. Xu et al. [39] reported 424 mg of reducing sugar per gram of switchgrass pretreated with lime (0.10 g/g, 121 °C, 15 min), while the present study resulted in 175 mg/g. Hu and Wen [26] reported a glucose recovery of 87% from switchgrass presoaked in an alkali solution (0.1 g alkali/g biomass) and treated by microwave heating (190 °C, 50 g/L solid content, 30 min). Glucose recovery of 68% from an alkali loading of 0.20–0.25 g NaOH/g biomass, RF heating temperature of 90 °C, and a solid content of 20% was reported for switchgrass by Hu et al. [27]. Alkali (NaOH) pretreatment targets intermolecular bonds between lignin and hemicelluloses, and improves the porosity of the biomass [40]. Since the mechanism of alkali pretreatment is the removal of lignin the difference in glucose recovery might be due to the facilitation of enzyme access caused by lignin removal.

Table 2 Interaction effects of different parameters (*p* value) for sugar recovery from switchgrass and prairie cord grass.

Biomass	Switchgrass				Prairie cord grass		
Source	Glucose	Xylose	Arabinose	Combined	Glucose	Xylose	Combined
CR	<0.0001	0.0272	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
MC	<0.0001	<0.0001	0.0044	<0.0001	<0.0001	<0.0001	<0.0001
SS	0.0028	<0.0001	0.6743	<0.0001	0.1036	0.3119	0.1791
Temperature	<0.0001	0.0005	0.0102	0.4663	<0.0001	<0.0001	<0.0001
CR*MC	0.0008	0.9553	0.9598	0.3508	0.1379	0.0003	0.0200
CR*SS	0.9875	0.1207	0.1841	0.6180	0.2424	0.5342	0.2936
CR*Temp	0.1352	0.2912	0.6720	0.4314	0.0303	0.0770	0.0571
MC*SS	0.0016	<0.0001	0.2586	0.0001	0.0035	0.0205	0.0144
MC*Temp	0.0439	0.0016	0.0151	0.0207	0.4182	0.7248	0.5218
SS*Temp	0.0025	0.0047	0.1835	0.0544	0.3753	0.2210	0.2093
CR*MC*SS	0.9716	0.0309	0.1260	0.7285	0.0056	0.0425	0.0116
CR*MC*Temp	0.5254	0.6928	0.1079	0.4163	0.7662	0.4639	0.6270
CR*SS*Temp	0.8194	0.8893	0.7446	0.9295	0.1379	0.1934	0.1035
MC*SS*Temp	<0.0001	<0.0001	0.5419	<0.0001	0.0168	0.0589	0.0346
CR*MC*SS*Temp	0.3391	0.2157	0.8776	0.3662	0.0617	0.1916	0.0480

CR screw compression ratio, *MC* moisture content, *SS* screw speed

Interactions and Treatment Combination Effect on Glucose Recovery

All the parameters considered (compression ratio, screw speed, temperature, and moisture content) in the present study had significant statistical differences in glucose recovery. Most of the interaction effects between independent variables on glucose recovery of switchgrass and prairie cord grass were significant, as is shown in Table 2. The screw speed was insignificant as an independent variable in glucose recovery for prairie cord grass, but the interaction of the screw speed with moisture content was significant. Furthermore, among second order interactions, the interaction of the screw speed and moisture content with either the compression ratio or the temperature was significant. However, the highest order interaction of these factors was insignificant.

Statistical analyses across the treatment combinations showed the existence of statistical differences (α=0.05) between treatment combinations for switchgrass and prairie cord grass, as presented in Tables 3, 4, 5, and 6. The highest glucose recovery of 45.3% was recorded for switchgrass with a pretreatment combination of 15% moisture content at a screw speed of 50 rpm and a barrel temperature of 150 °C with a screw compression ratio of 3:1 (Table 3). For prairie cord grass, the highest glucose (61.4%) was recovered at a screw speed of 50 rpm and a barrel temperature of 50 °C with a

Table 3 Effect of treatment combination on sugar recovery (%) from switchgrass using 3:1 screw compression ratio.

Screw speed, rpm										
Temperature, °C		50			100			150		
		50	100	150	50	100	150	50	100	150
						Glucose				
Moisture content, % wb	15	$35.09^{\text{d-i}}$	$28.56^{\text{h-s}}$	45.25^{a}	$35.96^{\text{c-g}}$	42.78^{ab}	$32.07^{\text{f-o}}$	43.22^{ab}	$42.57^{\text{a-c}}$	$39.38^{\text{a-e}}$
	25	$32.94^{\text{e-n}}$	$30.20^{\text{f-p}}$	$27.27^{\text{k-t}}$	$34.73^{\text{d-j}}$	$29.24^{\text{g-q}}$	$34.07^{\text{e-k}}$	$36.53^{\text{c-g}}$	$31.89^{\text{f-p}}$	$33.52^{\text{e-m}}$
	35	$30.39^{\text{f-p}}$	$26.33^{\text{l-u}}$	$21.38^{\text{s-u}}$	$27.50^{\text{j-t}}$	$27.66^{\text{ j-t}}$	$21.74^{\text{r-u}}$	$25.58^{\text{n-u}}$	$29.73^{\text{g-q}}$	$24.9^{\text{o-u}}$
	45	$25.91^{\text{n-u}}$	$27.25^{\text{k-t}}$	$28.07^{\text{h-t}}$	$29.82^{\text{f-g}}$	$28.59^{\text{h-s}}$	$24.96^{\text{o-u}}$	$25.77^{\text{n-u}}$	$25.71^{\text{n-u}}$	$25.98^{\text{n-u}}$
						Xylose				
Moisture content, % wb	15	$22.68^{\text{l-q}}$	$24.36^{\text{j-p}}$	$43.86^{\text{a-d}}$	$32.71^{\text{f-l}}$	50.26^{ab}	$33.73^{\text{e-j}}$	$42.90^{\text{a-e}}$	50.02^{ab}	$47.49^{\text{a-c}}$
	25	$23.13^{\text{k-g}}$	$27.01^{\text{g-n}}$	$24.35^{\text{j-p}}$	$25.89^{\text{g-p}}$	$25.49^{\text{h-p}}$	$35.22^{\text{d-h}}$	$34.82^{\text{d-i}}$	$29.30^{\text{g-m}}$	$34.84^{\text{d-i}}$
	35	$20.61^{\text{m-q}}$	$18.83^{\text{m-q}}$	$16.85^{\text{n-q}}$	$19.72^{\text{m-q}}$	$20.52^{\text{m-q}}$	$18.45^{\text{n-q}}$	16.05^{pq}	$23.17^{\text{k-q}}$	$19.38^{\text{m-q}}$
	45	13.46^{q}	$19.77^{\text{m-q}}$	$22.52^{\text{l-q}}$	$19.84^{\text{m-q}}$	$20.40^{\text{m-q}}$	18.91^{mq}	$16.20^{\text{o-q}}$	$17.96^{\text{n-q}}$	$20.28^{\text{m-q}}$
						Arabinose				
Moisture content, % wb	15	$37.59^{\text{b-m}}$	$31.9^{\text{c-m}}$	$42.53^{\text{b-m}}$	$40.73^{\text{b-m}}$	$25.66^{\text{h-m}}$	$48.30^{\text{a-h}}$	$35.72^{\text{b-m}}$	$42.23^{\text{b-m}}$	$40.53^{\text{b-m}}$
	25	$40.84^{\text{b-m}}$	$51.08^{\text{a-f}}$	$48.39^{\text{a-h}}$	$35.24^{\text{b-m}}$	$42.49^{\text{b-m}}$	$39.14^{\text{b-m}}$	$45.14^{\text{a-k}}$	$42.87^{\text{b-m}}$	$53.62^{\text{a-d}}$
	35	$33.88^{\text{b-m}}$	56.10^{ab}	$21.91^{\text{k-m}}$	$46.82^{\text{a-i}}$	55.88^{ab}	$38.95^{\text{b-m}}$	$31.06^{\text{c-m}}$	$53.93^{\text{a-c}}$	$40.38^{\text{b-m}}$
	45	$33.78^{\text{b-m}}$	$44.27^{\text{a-l}}$	$47.48^{\text{a-i}}$	$50.22^{\text{a-g}}$	$52.73^{\text{a-e}}$	66.48^{a}	$35.64^{\text{b-m}}$	$46.55^{\text{a-j}}$	$49.46^{\text{a-h}}$
						Combined				
Moisture content, % wb	15	$30.83^{\text{g-q}}$	$27.37^{\text{k-t}}$	44.47^{ab}	$35.27^{\text{d-j}}$	$43.74^{\text{a-c}}$	$34.35^{\text{e-m}}$	$42.33^{\text{a-d}}$	45.24^{a}	$42.44^{\text{a-d}}$
	25	$30.19^{\text{g-s}}$	$31.19^{\text{f-p}}$	$28.39^{\text{k-s}}$	$31.57^{\text{f-p}}$	$29.24^{\text{h-s}}$	$35.01^{\text{d-k}}$	$36.80^{\text{b-h}}$	$32.08^{\text{f-o}}$	$36.07^{\text{c-i}}$
	35	$27.19^{\text{k-t}}$	$26.67^{\text{l-t}}$	19.78^{ut}	$26.66^{\text{l-t}}$	$27.97^{\text{k-t}}$	$22.32^{\text{s-u}}$	$22.68^{\text{q-u}}$	$29.83^{\text{h-s}}$	$24.49^{\text{o-u}}$
	45	$22.19^{\text{s-u}}$	$26.29^{\text{m-t}}$	$28.05^{\text{k-t}}$	$28.29^{\text{k-s}}$	$28.10^{\text{k-t}}$	$27.03^{\text{j-t}}$	$23.31^{\text{p-u}}$	$24.98^{\text{o-u}}$	$26.33^{\text{m-t}}$

Control, 19.35% glucose, 16.39% xylose, and 20.57% combined sugar recovery. Values with the same superscripts are not significantly different at p=0.05 for each sugar

Table 4 Effect of treatment combination on sugar recovery (%) from switchgrass using 2:1 screw compression ratio.

Screw speed, rpm										
Temperature, °C		50			100			150		
		50	100	150	50	100	150	50	100	150
		Glucose								
Moisture content, % wb	15	27.44^{j-t}	27.66^{j-t}	33.55^{e-l}	31.15^{f-p}	36.81^{b-f}	27.26^{k-t}	41.53^{a-d}	35.20^{d-h}	28.43^{h-s}
	25	26.94^{k-u}	25.35^{o-u}	24.76^{o-u}	28.19^{h-s}	28.41^{h-s}	25.18^{o-u}	30.87^{f-p}	29.20^{g-q}	30.67^{f-p}
	35	28.20^{h-s}	25.26^{o-u}	22.72^{q-u}	29.74^{f-q}	24.67^{o-u}	19.76^{u}	25.77^{n-u}	29.07^{g-r}	21.32^{s-u}
	45	27.68^{j-t}	25.63^{n-u}	24.57^{p-u}	27.80^{i-t}	26.24^{i-u}	22.75^{q-u}	26.07^{n-u}	26.13^{m-u}	20.69^{tu}
		Xylose								
Moisture content, % wb	15	24.17^{k-g}	34.34^{d-j}	48.79^{a-c}	34.22^{d-j}	50.67^{ab}	35.71^{d-g}	53.19^{a}	45.79^{a-c}	40.21^{c-f}
	25	19.75^{m-q}	18.34^{n-q}	27.36^{g-n}	24.36^{j-p}	26.61^{g-o}	33.17^{e-k}	41.48^{b-f}	39.70^{c-f}	45.44^{a-c}
	35	20.15^{m-q}	17.70^{n-q}	17.7^{n-q}	24.92^{i-p}	17.76^{n-q}	17.67^{n-q}	20.75^{m-q}	22.06^{m-q}	26.05^{g-p}
	45	18.44^{n-q}	18.62^{n-q}	20.87^{m-q}	20.99^{m-q}	17.44^{n-q}	22.06^{m-q}	18.97^{m-q}	19.21^{m-q}	22.97^{k-q}
		Arabinose								
Moisture content, % wb	15	29.51^{e-m}	34.56^{b-m}	23.70^{i-m}	23.69^{i-m}	21.01^{lm}	35.63^{b-m}	26.38^{g-m}	34.47^{b-m}	22.62^{k-m}
	25	39.91^{b-m}	34.41^{b-m}	28.85^{f-m}	29.78^{d-m}	39.46^{b-m}	33.73^{b-m}	23.87^{i-m}	38.27^{b-m}	19.93^{m}
	35	36.25^{b-m}	40.91^{b-m}	37.67^{b-m}	28.52^{f-m}	31.06^{c-m}	20.60^{lm}	22.72^{j-m}	32.55^{b-m}	36.29^{b-m}
	45	33.37^{b-m}	35.39^{b-m}	30.74^{c-m}	29.47^{e-m}	35.56^{b-m}	50.13^{a-g}	28.37^{f-m}	36.68^{b-m}	41.13^{b-m}
		Combined								
Moisture content, % wb	15	26.46^{m-t}	30.80^{g-r}	38.08^{a-g}	31.50$_{f-p}$	40.22^{a-e}	31.19^{f-p}	43.85^{a-c}	38.98^{a-f}	32.11^{f-o}
	25	25.66^{n-t}	23.73^{o-u}	26.12^{m-u}	26.96^{j-u}	28.89^{h-s}	28.97^{h-s}	34.01^{e-n}	33.95^{e-n}	34.93^{d-l}
	35	26.10^{m-t}	24.11^{o-u}	22.44^{r-u}	27.86^{j-t}	22.75^{q-u}	19.09^{u}	23.63^{p-u}	26.87^{k-u}	24.58^{o-u}
	45	24.91^{o-u}	24.08^{o-u}	23.86^{o-u}	25.49^{o-u}	24.00^{o-u}	25.31^{o-u}	23.73^{o-u}	24.70^{o-u}	23.62^{p-u}

Control, 19.35% glucose, 16.39% xylose, and 20.57% combined sugar recovery. Values with the same superscripts are not significantly different at p=0.05 for each sugar

moisture content of 25% when a screw compression ratio of 3:1 was used. As noted in Tables 4 and 6, the lowest glucose recovery from switchgrass and prairie cord grass resulted from the samples extruded at 150 °C and a screw speed of 100 or 150 rpm using a screw compression ratio of 2:1 with a moisture content of 35% or 45%. The highest glucose recovery from switchgrass and prairie cord grass was 1.34 and 1.94 times higher than their respective control samples.

Main Effect of Compression Ratio on Xylose Recovery

All the independent variables considered in this study significantly affected the xylose recovery from switchgrass, as depicted in Fig. 2a–d. Xylose recovery from switchgrass decreased by 6% when the screw compression ratio was increased from 2:1 to 3:1, the opposite of the glucose recovery trend. In the case of prairie cord grass, xylose recovery increased by 43% when the screw compression ratio was increased (p=0.0001) from 2:1 to 3:1, a trend in agreement with glucose recovery. Compression ratio strongly influences the melting process in plastics [41], whereas here it contributes to high shear force and residence time [42]. The differences in pattern might be due to the work put (shear force) on the biomass, the density differences, and their inherent characteristics. Although

Table 5 Effect of treatment combination on sugar recovery (%) from prairie cord grass using 3:1 screw compression ratio.

Screw speed, rpm		50			100			150		
Temperature, °C		50	100	150	50	100	150	50	100	150
						Glucose				
Moisture content, % wb	15	58.88^{ab}	56.23^{a-e}	50.34^{a-j}	54.17^{a-h}	48.50^{a-l}	54.91^{a-g}	43.70^{b-q}	51.97^{a-j}	43.30^{b-r}
	25	61.42^{a}	50.80^{a-j}	44.84^{b-p}	53.53^{a-l}	52.08^{a-j}	37.71^{i-v}	56.60^{a-d}	54.46^{a-h}	48.17^{a-l}
	35	53.27^{a-i}	51.93^{a-j}	48.49^{a-l}	55.04^{a-f}	51.70^{a-j}	26.94^{s-x}	50.36^{a-j}	51.07^{a-j}	40.38^{e-t}
	45	49.68^{a-k}	45.69^{a-o}	51.28^{a-j}	49.13^{a-l}	46.46^{a-n}	36.28^{k-v}	54.28^{a-h}	52.82^{a-i}	33.46^{l-v}
						Xylose				
Moisture content, % wb	15	82.77^{a}	82.22^{a}	79.42^{a-d}	81.00^{ab}	77.02^{a-f}	82.85^{a}	72.97^{a-g}	80.50^{ab}	72.55^{a-g}
	25	84.25^{a}	78.80^{a-d}	75.35^{a-g}	78.77^{a-d}	79.12^{a-d}	59.35^{a-m}	81.78^{ab}	83.28^{a}	78.70^{a-d}
	35	77.09^{a-f}	80.43^{ab}	74.51^{a-g}	83.17^{a}	79.16^{a-d}	38.19^{j-r}	77.29^{a-e}	78.14^{a-e}	62.61^{a-l}
	45	63.81^{a-k}	70.13^{a-h}	77.14^{a-f}	68.29^{a-i}	71.72^{a-g}	53.20^{b-o}	76.79^{a-f}	79.39^{a-d}	41.88^{k-q}
						Combined				
Moisture content, % wb	15	65.11^{ab}	62.45^{a-e}	58.73^{a-g}	62.12^{a-e}	56.23^{a-j}	63.42^{a-d}	50.54^{a-o}	60.71^{a-f}	50.08^{a-o}
	25	65.79^{a}	58.58^{a-h}	51.92^{a-n}	61.05^{a-f}	59.73^{a-f}	43.57^{d-s}	61.91^{a-f}	63.40^{a-d}	55.15^{a-k}
	35	63.32^{a-d}	59.85^{a-f}	55.52^{a-k}	62.06^{a-f}	59.48^{a-g}	29.21^{p-x}	57.44^{a-i}	59.05^{a-g}	45.28^{b-s}
	45	53.04^{a-m}	52.95^{a-m}	60.37^{a-f}	54.85^{a-l}	54.28^{a-l}	39.83^{g-v}	61.21^{a-f}	60.78^{a-f}	31.78^{o-x}

Control, 20.91% glucose, 33.75% xylose, and 23.83% combined sugar recovery. Values with the same superscripts are not significantly different at $p=0.05$ for each sugar

switchgrass had a higher xylose than did prairie cord grass, the higher lignin content of switchgrass might have played a vital role in the reduction of xylose recovery. As observed from the Fig. 3a–d, prairie cord grass always had a higher xylose recovery than did switchgrass. The higher value might be due to the lower lignin content of prairie cord grass as compared to switchgrass.

Main Effect of Screw Speed on Xylose Recovery

Screw speed as an independent variable had a statistically significant influence on xylose recovery from switchgrass but not on xylose recovery from prairie cord grass, as evident from the main effect analysis (Figs. 2c and 3c). Evidently, the rate of shear development at 50 rpm was enough to disturb the prairie cord grass cell wall due to low lignin and xylose content and high mean residence time. For switchgrass, the shear development at 50 rpm was insufficient because of the higher lignin and xylose content as compared to prairie cord grass. When screw speed was increased from 50 to 150 rpm, xylose recovery increased correspondingly by 33% for switchgrass. Thus, the rate of shear development (high screw speed) was a more critical factor than the mean residence time (low screw speed).

Main Effect of Barrel Temperature on Xylose Recovery

An increase in barrel temperature (50 to 150 °C) yielded a 14% higher xylose recovery for switchgrass, but xylose recovery for prairie cord grass decreased by 19% (Figs. 2b and 3b).

Table 6 Effect of treatment combination on sugar recovery (%) from prairie cord grass using 2:1 screw compression ratio.

Screw speed, rpm										
Temperature, °C		50			100			150		
		50	100	150	50	100	150	50	100	150
						Glucose				
Moisture content, % wb	15	$48.89^{\text{a-l}}$	$45.68^{\text{a-l}}$	$33.92^{\text{k-v}}$	$41.75^{\text{i-s}}$	$47.42^{\text{a-m}}$	$28.86^{\text{q-x}}$	$42.64^{\text{c-s}}$	$50.43^{\text{a-j}}$	$31.85^{\text{n-v}}$
	25	$44.33^{\text{b-q}}$	$50.16^{\text{a-j}}$	$31.96^{\text{n-w}}$	$25.84^{\text{t-x}}$	$31.15^{\text{n-w}}$	$29.06^{\text{p-x}}$	$58.00^{\text{a-c}}$	$48.58^{\text{a-l}}$	$29.18^{\text{p-x}}$
	35	21.65^{wx}	$47.14^{\text{a-m}}$	$22.37^{\text{v-x}}$	$49.84^{\text{a-k}}$	$39.54^{\text{f-t}}$	$23.16^{\text{u-x}}$	$42.83^{\text{c-r}}$	$38.87^{\text{g-u}}$	$23.11^{\text{u-x}}$
	45	$34.20^{\text{k-v}}$	$31.58^{\text{n-w}}$	21.26^{wx}	$38.79^{\text{h-u}}$	$38.50^{\text{h-u}}$	$27.83^{\text{r-x}}$	$27.83^{\text{r-x}}$	$30.42^{\text{o-w}}$	14.56^{x}
						Xylose				
Moisture content, % wb	15	$76.89^{\text{a-f}}$	$76.37^{\text{a-f}}$	$55.88^{\text{a-n}}$	$66.93^{\text{a-i}}$	$77.74^{\text{a-e}}$	$48.54^{\text{f-p}}$	$66.49^{\text{ a-i}}$	82.50^{a}	$50.89^{\text{d-p}}$
	25	$76.11^{\text{a-f}}$	$78.80^{\text{a-d}}$	$51.47^{\text{c-p}}$	$36.58^{\text{k-r}}$	$49.65^{\text{e-p}}$	$50.73^{\text{d-p}}$	81.78^{ab}	$79.93^{\text{a-c}}$	$47.12^{\text{g-p}}$
	35	18.69^{qr}	$68.25^{\text{a-i}}$	$30.56^{\text{n-r}}$	$66.26^{\text{a-j}}$	$56.22^{\text{a-n}}$	$35.11^{\text{l-r}}$	$55.73^{\text{a-n}}$	$55.53^{\text{a-n}}$	$31.02^{\text{m-r}}$
	45	$30.77^{\text{n-r}}$	$35.12^{\text{l-r}}$	$23.86^{\text{p-r}}$	$40.67^{\text{i-p}}$	$46.90^{\text{g-p}}$	$35.04^{\text{l-r}}$	$26.17^{\text{o-r}}$	$36.72^{\text{k-r}}$	12.61^{r}
						Combined				
Moisture content, % wb	15	$55.13^{\text{a-k}}$	$53.17^{\text{a-m}}$	$38.96^{\text{h-w}}$	$47.44^{\text{a-q}}$	$54.59^{\text{a-l}}$	$34.45^{\text{m-w}}$	$47.92^{\text{a-o}}$	$59.38^{\text{a-g}}$	$36.23^{\text{k-w}}$
	25	$51.78^{\text{a-n}}$	$58.56^{\text{a-h}}$	$36.37^{\text{l-w}}$	$28.00^{\text{q-x}}$	$35.31^{\text{l-w}}$	$36.93^{\text{j-w}}$	$63.62^{\text{a-c}}$	$58.21^{\text{a-h}}$	$33.25^{\text{n-w}}$
	35	20.10^{wx}	$54.32^{\text{a-l}}$	$23.93^{\text{u-x}}$	$52.80^{\text{a-m}}$	$44.20^{\text{c-s}}$	$25.75^{\text{s-x}}$	$45.48^{\text{c-r}}$	$43.05^{\text{i-t}}$	$24.57^{\text{t-x}}$
	45	$32.10^{\text{o-x}}$	$33.11^{\text{n-w}}$	$21.27^{\text{v-x}}$	$38.03^{\text{i-w}}$	$42.20^{\text{f-u}}$	$28.93^{\text{p-x}}$	$26.44^{\text{r-x}}$	$31.31^{\text{o-x}}$	13.53^{x}

Control, 20.91% glucose, 33.75% xylose, and 23.83% combined sugar recovery. Values with the same superscripts are not significantly different at p=0.05 for each sugar

The trend might be due to the difference in utilization of the temperatures for thermal softening of these grasses. As previously discussed, switchgrass required a higher temperature for softening due to the higher lignin content. However, barrel temperatures of 100 and 150 °C produced similar xylose recovery. In the case of prairie cord grass, xylose recovery increased up to 100 °C, a further increase in the temperature reduced xylose recovery. The result might be due to lower lignin and thermal softening at higher temperature.

Main Effect of Moisture Content on Xylose Recovery

When moisture content was varied from 15% to 45% for switchgrass and prairie cord grass, xylose recovery decreased (p=0.0001) by 51% and 32%, respectively, a pattern similar to that of glucose recovery. No statistical difference in xylose recovery from switchgrass at higher moisture content (35% and 45%) was observed. In contrast, no difference in xylose recovery from prairie cord grass was observed at low moisture content (15% and 25%). A possible reason might be the differences in lignin and xylose contents of these biomasses. As mentioned earlier, high moisture decreases the friction [34], more slippage thereby less disturbance to the biomass.

Alizadeh et al. [29] obtained a xylose conversion of 70% from switchgrass pretreated with the AFEX. Dien et al. [36] reported more than 80% of non-glucan sugars from dilute sulfuric pretreatment of switchgrass with an optimal acid concentration of 1.2% (*w*/*v*). Switchgrass pretreated using 30% aqueous ammonium hydroxide with liquid–solid ratios of 5 and 10 ml/g and residence times of 5 and 10 min resulted in 40–50% lignin reduction and

50% hemicelluloses reduction [43]. Hu andWen [26] reported a xylose recovery of 95% from switchgrass presoaked in an alkali solution (0.1 g alkali/g biomass) and treated by microwave heating (190 °C, 50 g/l solid content, 30 min). Hu et al. [27] reported a xylose recovery of 96% for switchgrass pretreated with an alkali loading of 0.20–0.25 g NaOH/g biomass, an RF heating temperature of 90 °C, and a solid content of 20%. As discussed in the section on glucose recovery, lignin removal by alkali might have contributed to the higher xylose recovery.

Interactions and Treatment Combination Effect on Xylose Recovery

Independent variables such as screw compression ratio, barrel temperature, and moisture content included in the present study had a strong influence on xylose recovery, as evident from Table 2. The screw speed alone had no significant influence on xylose recovery from prairie cord grass, but a combination of screw speed with moisture content, and then either with compression ratio or temperature resulted in significant xylose recovery, similar to that of glucose recovery. Most of the interactions were significant for the lower order interactions, but few were significant for higher order interactions both for switchgrass and prairie cord grass (Table 2). Statistical analyses of all independent variable combinations showed a significant statistical difference ($p<0.05$) between the treatment combinations presented in Tables 3 and 6. The highest xylose recovery from switchgrass of 53.2% was recorded at a barrel temperature of 50 °C and a screw speed of 150 rpm using a screw compression ratio of 2:1 with a moisture content of 15%. However, no statistical differences existed between different treatment combinations; thus, for switchgrass pretreatment any of the conditions can be selected from different treatment combinations. The highest and lowest xylose recoveries (53.2% and 13.5%) were recorded with screw compression ratio of 2:1 and 3:1, respectively, a pattern opposite that of glucose recovery from switchgrass. The highest xylose recovery (84.3%) for prairie cord grass was obtained at a screw speed of 50 rpm and a barrel temperature of 50 °C with a moisture content of 25% using a screw compression ratio of 3:1. The highest and lowest xylose recoveries (84.3% and 12.6%) were recorded at the same glucose recovery pretreatment conditions. A possible reason might be that since prairie cord grass becomes soft with high moisture, most of the heat might have been utilized for moisture evaporation instead of cell wall disturbance. The highest xylose recovery from switchgrass and prairie cord grass were 2.25 and 1.50 times higher than their respective control samples.

Main Effect Analysis of Independent Variables on Arabinose Recovery

As observed in Fig. 2a–d, the compression ratio, temperature, and moisture content affected arabinose recovery from switchgrass. Among the independent variables, the compression ratio made a significant contribution towards arabinose recovery as evident from the mean values. The screw compression ratio of 3:1 had a higher (35%) arabinose recovery than that of 2:1. Barrel temperature had a positive impact on arabinose recovery, an increase of 17% when the temperature was increased from 50 to 100 °C. A further increase in temperature did not result in any improvement in arabinose recovery from switchgrass. In contrast to glucose and xylose recovery, arabinose recovery exhibited a positive relation with the moisture content in switchgrass. Arabinose recovery increased from 33.2% to 41.5% when the moisture content was increased from 15% to 45%, the equivalent to a 25% increase in arabinose recovery.

Arabinose recovery from prairie cord grass was recorded for only 66% of the treatment combinations with 3:1 compression ratio and for only 33% with 2:1 compression ratio. The percentage of recovery obtained might be due to the low arabinose content (1.59%) in the raw prairie cord grass.

Interaction Effects and Treatment Combination Effects on Arabinose Recovery

Although the main effect of compression ratio, moisture content, and temperature were significant, their lower and higher order interactions were insignificant for arabinose recovery from switchgrass. Among all first order interactions, only moisture content with temperature was significant (Table 2). Statistical analyses of arabinose recovery across the treatment combinations are presented in Table 3. In general, arabinose recovery increased with an increase in temperature across the screw speeds and compression ratios. A similar trend was observed in the main effect analysis. A maximum arabinose concentration of 3.27 g/L (66.48%) was recorded at a screw speed of 100 rpm and a temperature of 150 °C using a screw compression ratio of 3:1 with a moisture content of 45%. It can be concluded that for higher arabinose recovery, high moisture content was necessary. The possible reason might be the arrangement of arabinose among the sugars in the biomass.

Main Effect Analysis of Independent Variables on Combined Sugar Recovery

As mentioned previously, combined sugar recovery was the sum of the glucose, xylose, and arabinose to the total sugar available in the raw biomass. For example, switchgrass pretreated at 150 °C, 50 rpm, and 15% moisture content gave a glucose, xylose, and arabinose of 11.54%, 7.63%, and 2.08%, respectively. The total sugar (47.18% denominator for combined sugar recovery) was calculated from the chemical composition given in Table 1. The individual sugar recovery and combined sugar recoveries calculation are shown below.

Glucose recovery=$(11.54/25.5)\times100=45.25\%$
Xylose recovery=$(7.63/17.4)\times100=43.86\%$
Arabinose recovery=$(2.08/4.9)\times100=42.50\%$
Combined sugar recovery=$(21.25/47.18)\times100=44.47\%$

As observed in Fig. 2a, c, and d, the compression ratio, moisture content, and screw speed had a strong influence on combined sugar recovery for switchgrass. In the case of prairie cord grass, the compression ratio, moisture content, and temperature influenced combined sugar recovery (Table 2) to a lesser extent. When the screw compression ratio was increased from 2:1 to 3:1, the combined sugar recovery increased (p=0.0001) correspondingly by 8.4% and 40% for switchgrass and prairie cord grass, respectively, differences attributed to more work put (shear force, more residence time) on the biomass. The lignin to total sugar ratio of switchgrass and prairie cord grass were 0.52 and 0.44, respectively, showing that more work was required to disturb the switchgrass due to its high lignin content. Irrespective of the screw compression ratios, prairie cord grass had a higher combined sugar recovery than did switchgrass, because of the low lignin and high sugar content.

Screw speed had a positive impact on combined sugar recovery from switchgrass, as is evident from Fig. 2c. As the screw speed was increased (p=0.0001) from 50 to 150 rpm, combined sugar recovery also increased by 14%. This result shows that the rate of shear development (high speed) plays a more critical role in combined sugar recovery than the mean residence time (low speed). No regular trend could be observed for an increase in the barrel temperature from 50 to 100 °C for prairie cord grass, a trend similar to the xylose recovery. Moisture content as an independent variable had a strong influence (p=0.0001) on combined sugar recovery from both biomass (switchgrass and prairie cord grass). Since combined sugar recovery was the addition of individual sugars, it followed the same pattern with respect to moisture content. As the moisture content was increased from 15% to 45%, the combined sugar recovery decreased considerably by 31% and 23% for switchgrass and prairie cord grass, respectively. The differences in percentage can be attributed to the lignin and sugar content of the biomass. When moisture content is low a high friction is developed, which disturbs the biomass structure and results in a high combined sugar recovery. In general, the lower the moisture content, the higher the combined sugar recovery.

Interaction and Treatment Combination Effects on Combined Sugar Recovery

Table 2 shows that some of the lower and higher order interactions were significant on combined sugar recovery for switchgrass and prairie cord grass. Barrel temperature as an independent variable affected individual sugar recovery but was insignificant for combined sugar recovery for switchgrass. Temperature alone was insignificant until combined with moisture and screw speed. Similar to prairie cord grass glucose and xylose recovery, although screw speed as an independent variable was insignificant, it became significant when combined with moisture content, and then compression ratio, temperature. The highest order interaction of independent variables was significant for prairie cord grass, but not for switchgrass, similar to glucose, xylose, and arabinose recovery.

Statistical analyses of all parameters showed a significant difference, as presented in Tables 3 and 6 for switchgrass and prairie cord grass, respectively. The highest combined sugar recovery of 45.2% for switchgrass was recorded at a temperature of 100 °C and 150 rpm using a 3:1 compression ratio with 15% moisture content. This treatment combination coincides with the highest glucose recovery condition. The present result was lower than the value (78.5%) for alkali pretreated switchgrass by RF heating as reported by Hu et al. [27]. Hu and Wen [26] reported 90% combined sugar recovery for switchgrass presoaked in an alkali solution (0.1 g alkali/g biomass) and treated by microwave heating (190 °C, 50 g/L solid content for 30 min). The difference is mostly due to the removal of 82% lignin [26], which resulted in high sugar recovery. For prairie cord grass, the highest and lowest combined sugar recovery coincides with the treatment conditions of the highest and lowest glucose and xylose recovery. The highest combined sugar recovery from switchgrass and prairie cord grass was 1.20 and 1.76 times higher than their respective control samples. In this study the maximum combined sugar concentration obtained was 21.61 and 31.70 g/L for switchgrass and prairie cord grass, respectively, whereas de Vrije et al. [12] reported monomeric sugar as 23.6 g/L for miscanthus. The difference might be due to the type of extruder, the conditions employed (NaOH pretreatment after extrusion), and the inherent nature of biomasses.

Byproducts from Switchgrass and Prairie Cord Grass

Compounds such as furfural, HMF, acetic acid, and formic acid have been described as inhibitors of fermentation to different extents; hence, they must be taken into account. The degradation of sugar is highly dependent on temperature, residence time, and acid concentration/acidic conditions. Acetic acid is released from the hydrolysis of acetyl groups present in hemicellulose, as a result of deacetylzation. Acetic acid was found in most of the treatment combinations in the range of 0.029−0.132 and 0.020−0.122 g/L for switchgrass and prairie cord grass, respectively. The highest acetic acid (0.132 g/L) resulted in switchgrass pretreated at a barrel temperature of 100 °C and screw speed of 100 rpm using

3:1 screw compression ratio with 15% moisture content. For prairie cord grass, the highest concentration of acetic acid (0.122 g/L) was found with a barrel temperature of 150 °C and a screw speed of 150 rpm using a 3:1 compression ratio with 15% moisture content.

Glycerol was found in a few treatment combinations for all biomass. The concentration of glycerol was in the range of 0.02–0.18 g/L for switchgrass and 0.04–0.08 g/L for prairie cord grass. The highest glycerol concentration was found in switchgrass (0.18 g/L) extruded at 50 °C and 150 rpm using a 3:1 screw compression ratio with a moisture content of 25%. In the case of prairie cord grass, the highest glycerol concentration (0.08 g/L) was obtained at high temperature and speed (150 °C and 150 rpm) using a 3:1 screw compression ratio with a 35% moisture content. No furfural and HMF were found in any of the pretreatment conditions studied for switchgrass and prairie cord grass. The finding was in agreement with those for other extrusion pretreatments performed on different biomasses [8, 11, 12]. The possible reasons could be low temperature, less residence time, low moisture content, and no acidic conditions. It was noteworthy that the highest byproducts concentration did not occur with the highest glucose or xylose recovery conditions for switchgrass and prairie cord grass.

Conclusions

This study was conducted in order to investigate the effect of biomass moisture content and extruder parameters such as the screw compression ratio, screw speed, and barrel temperature on various sugar recoveries (glucose, xylose, arabinose, mannose, and combined sugar). The compression ratio, screw speed, temperature, and moisture content as independent variables significantly contributed to glucose, xylose, and combined sugar recovery. Statistical analyses revealed that most of the interactions were significant for both biomasses. In general, 3:1 screw compression ratio resulted in a higher sugar recovery than did one of 2:1. Based on glucose, xylose, and combined sugar recovery (45.3%, 43.9%, and 44.5%), switchgrass can be pretreated at a screw speed of 50 rpm and a barrel temperature of 150 °C with a moisture content of 15%. Prairie cord grass can be extruded at a low temperature and screw speed (50 °C and 50 rpm) with a 25% moisture content for better glucose, xylose, and combined sugar recovery (61.4%, 84.3%, and 65.8%). In addition, these treatment conditions resulted in low concentrations (0.02 and 0.04 g/L) of glycerol and acetic acid for both switchgrass and prairie cord grass. Feedstock size is also an important factor which would affect the efficiency of extrusion pretreatment; accordingly it will be addressed in the future work.

Acknowledgments This research was supported by funding from the Agricultural Experiment Station and North Central Sun Grant Center at South Dakota State University through a grant provided by the US Department of Transportation, Office of the Secretary, Grant No. DTOS59-07-G-00054. Also, enzymes supplied by Novozymes, Inc for conducting this study were greatly appreciated.

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