Green Chemistry

Dynamic Article Links

Cite this: *Green Chem.*, 2012, **14**, 1982

www.rsc.org/greenchem

PAPER

Low liquid–solid ratio (LSR) hot water pretreatment of sugarcane bagasse

María Evangelina Vallejos,[a,b] Marcia Dib Zambon,[c] María Cristina Area[a,b] and Antonio Aprigio da Silva Curvelo*[c,d]

Received 18th March 2012, Accepted 25th April 2012
DOI: 10.1039/c2gc35397k

Low liquid–solid ratio (LSR) can be used to obtain high-content xylo-oligosaccharide (XOS) spend liquor by hot water pretreatment. Developing a technology based on low LSR results in more efficient water usage in the system and thus in lower capital and operating costs. Xylans from xylan rich agro-industrial waste are abundant hemicellulosic polymers with enormous potential for industrial applications. Currently, freeze-dried xylo-oligosaccharides are used as bio-based polymers and hydrolysates containing high xylose contents are converted to several chemical products. In this study, sugarcane bagasse was treated with water at low LSRs and mild temperatures in order to assess the effects of varying the pretreatment conditions on the xylo-oligosaccharide and xylose concentrations, and use a central composite experimental design to optimize the process parameters. The pretreatments were performed in the ranges temperature: 143.3–176.7 °C, time: 20–70 min and LSR: 1 : 1 to 11 : 1 (g g^{-1}). The maximum concentrations of xylose and xylan were 13.76 and 36.18 g L^{-1} (equivalent to 48.29 g L^{-1} of xylan), respectively, which were achieved by treating bagasse at 170 °C for 60 min, with LSR of 3 g g^{-1}. The amount of xylan removed under these conditions was almost 57%. The soluble xylan consisted mainly of xylo-oligosaccharides (74 wt% of the identified compound in the spent liquor).

1. Introduction

The growing demand for food, materials and energy and the need to limit the emission of greenhouses gases has motivated research on renewable, bio-based resource and the development of technologies to convert these resources to usable materials and fuels. Lignocellulosic biomass has been the main renewable resource used throughout the 20th century as a raw material for energy, building materials, pulp and paper. In recent years, lignocellulosic materials have been studied intensively as alternative raw materials for new bio-based fuels, chemical, pharmaceutical and material industries.[1] Lignocellulosic waste generated by agriculture and forestry is a renewable low-cost source of carbohydrates and energy, available in large quantities. Sugarcane bagasse is an important agro-industrial waste product in Brazil, India, China and Thailand. These four countries account for more than 50% of the world's sugarcane production.[2] Brazil, the world's largest producer of sugarcane, generated about 93.6–156 million t of bagasse in the 2010/2011 harvest (150–250 kg bagasse t^{-1} sugarcane).[3] It is usually burned in the sugar and ethanol mills to produce energy. Bagasse is composed of 40–45% cellulose, 20–30% lignin, 30–35% hemicelluloses and minor amounts of extractives and inorganic compounds.[4–6] Sugarcane bagasse hemicelluloses are mainly composed of two pentosan polymers (xylans and arabinans).

Cellulose and lignin from lignocellulosic materials are highly valued as a commodity and fuel, respectively. Although hemicelluloses are more easily extracted from the biomass than cellulose and lignin, at the moment, their potential for conversion into bio-based products has not yet been developed commercially. Further studies and technical advances will be needed for such exploitation to be technically and economically feasible. Xylans are the most abundant hemicellulosic polymers with enormous potential for industrial applications. Xylo-oligosaccharides from xylan rich agro-industrial waste can be used in a wide range applications, including their conversion to xylose, xylitol, furfural, succinic acid, additives in papermaking, ethanol, hydrogels, films and bio-based polymers.[7,8]

Hemicelluloses extraction, prior to the transformation of lignocellulosic materials to high-value bio-based chemicals or materials, could improve the economic efficiency of such processes.[9–11] In pulping processes, such as the kraft process, the hemicelluloses are dissolved in the black liquor and later they are burned in the recovery boiler, even though they have lower calorific power than to lignin. The removal of hemicelluloses before pulping would reduce the consumption of alkali in neutralizing of the acetic acid generated by deacetylation of

[a]*Programa de Celulosa y Papel, FCEQyN, Universidad Nacional de Misiones, C.P.3300, PosadasMisiones, Argentina*
[b]*Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), C.P.1033 Capital Federal, Argentina*
[c]*Grupo de Físico-química orgânica, Departamento de Físico-química, Instituto de Química de São Carlos, Universidade de São Paulo, Av. Trabalhador São Carlense 400, Caixa Postal 780, 13560-970 São Carlos, São Paulo, Brazil. E-mail: aprigio@iqsc.usp.br; Fax: +55 16 3373 9952; Tel: +55 16 3373 9938*
[d]*Laboratório Nacional de Ciência e Tecnologia do Bioetanol (CTBE), Centro de Pesquisa em Energia e Materiais (CNPEM), Caixa Postal 6179, 13083-970 Campinas, São Paulo, Brazil*

hemicelluloses and would improve the energy efficiency. In the case of the "cellulosic ethanol" or "second generation ethanol" processes, a pretreatment step is required to disrupt the carbohydrate–lignin complex and thus allow the hydrolytic enzymes to gain access to the carbohydrates.[12–14] The pretreatment process defines the cost at which the cellulose (and hemicelluloses) can be converted to cellulosic pulp or ethanol, since it represents an important part of the total project cost.[14] In order for pretreatments to be effective and economically feasible, low energy and water consumption, low investment costs, inexpensive and easily recoverable reagents and applicability to various types of raw material are required. At present, a lot of effort is going into research and technological development, to determine which pretreatment is more efficient for lignocellulosic materials. Although these pretreatments have been widely studied, it is as yet unclear which is the best technological process. The development of technologies based on the biorefinery concept could improve the utilization of lignocellulosic materials through the separation of its components in a sequential process. Recent articles provide an overview of the several pretreatments used in these processes.[7,12–14]

Hot water pretreatment uses pressurized hot water at moderate temperatures for the selective removal and degradation of the xylans present in the lignocellulosic material. Compared to dilute acid pretreatment, hot water pretreatment is an environmentally friendly process, with low corrosion of equipment and less xylose degradation, hence smaller amount of inhibitors in the spent liquor, and it is simpler and cheaper.[12]

Hot water pretreatment has potential commercial application in the biorefinery industry and it has proved to be efficient in the extraction of hemicelluloses from various lignocellulosic materials.[15–21] These processes have the potential to recover from 55 to 84% of the hemicelluloses and to produce hydrolysate almost without inhibitors of the fermentation of glucose to ethanol.[22] This pretreatment use pressurized hot water at temperatures of 150–230 °C. The catalysts are the hydronium ions (H_3O^+) generated *in situ* by auto-ionization of water and the acetic acid generated by hydrolysis of the hemicelluloses.[22,23] At temperatures above 220 °C, cellulose is partially degraded to 5-hydroxymethylfurfural (HMF), whereas below 100 °C the hydrolysis of lignocellulosic material is not significant.[7] Generally, the time of pretreatment depends on temperature. The liquid–solid ratios (LSR) are usually around 10 but these ratios can range from 2 to 100 g g^{-1}.[7,24] Low LSRs raise the acetic acid (catalyst) concentration in the reaction medium promoting the release of acetyl groups, which favours autohydrolysis of the lignocellulosic material. Hence, an interesting way of reducing the energy costs of the process would be to use a low liquid–solid ratio and mild temperatures in the pretreatment, thus economizing both steam and electricity.

Thus, if a low LSR were used to obtain high-content xylo-oligosaccharides syrups from lignocellulosic materials by hot water pretreatment, the amount of water in the system would be reduced and operating costs would be lower. This would result in less water to remove downstream, lower usage of fresh water and steam usage and a smaller storage tank and pumping system for the same quantities of feedstock. Low LSRs offer economic advantages but result in a partial diffusion and transfer of the dissolved hemicelluloses. The conventional hot water processes, with high LSRs, produce low xylo-oligosaccharides and monosaccharides concentrations, so that the water must be removed by evaporation, with high energy requirements. Moreover, autohydrolysis at mild temperatures produces high molecular weight xylo-oligosaccharides, without substantially modifying the cellulose and lignin, enabling their recovery for further processing. Some recent applications for xylose and high and low molecular weight xylo-oligosaccharides and required concentrations are shown in Table 1.

Table 1 Some applications for xylose and high and low molecular weight xylo-oligosaccharides (d.s.: dried solid)

Raw material	Applications	Concentration	Reference
Xylose	Xylitol	48–155 g L^{-1}	25–28
	Furfural	d.s.–100 g L^{-1}	29–31
	Ethanol	7.5–70 g L^{-1}	32–34
	Succinic acid	52 g L^{-1}	35
High and low molecular weight xylo-oligosaccharides	Films	d.s.	36–39
	Gels	d.s.	40, 41
	Additives	d.s.	42
	Nanocomposites	d.s.	43–46
	Bio-based polymers	d.s.	47, 48

The effect of varying the LSRs on the hemicelluloses concentration range has been studied in relation to the dilute acid pretreatment of biomass, but there are few references about its effect on the autohydrolytic pretreatment, where the influence of LSR is unclear.[12,49–51]

The LSR did not affect the XOS recovery from brewery's spent grain autohydrolysis.[52] The concentration of xylose and lignin content did not depend on the values of the LSR for the *H. funifera* hydrothermal pretreatment.[53] Limited information is available on the dependence of the hemicelluloses concentration range in the hydrolysate as a function of temperature, time and LSR. Up to now, most studies of hot water hydrolysis have focused on the investigation of the kinetic models and the fermentability of the hydrolysates. LSR should also be taken into consideration and optimized because, it provides a simple means of energy saving and waste water reduction. In this study, low-LSR hot water pretreatment of sugarcane bagasse was tested at mild temperatures, to assess the influence of the pretreatment on the xylo-oligosaccharide and xylose concentrations achieved, while using a central composite design (CCD) of experiment to optimize the process parameters.

2. Materials and methods

2.1. Raw materials

Whole sugarcane bagasse was supplied by the Ipiranga sugar mill and ethanol plant (Descalvado, State of São Paulo, Brazil).

The bagasse was washed with constant stirring (70 °C, 1 h), to remove residual sugar, after which it was air-dried under room conditions (final moisture content <10% of dry weight), homogenized and stored in plastic containers.

2.2. Analysis of bagasse

One sample of bagasse was used for ash determination at 525 °C (TAPPI T211 om-02 "Ash in wood, pulp, paper and paperboard"). Another sample was milled and sieved (<1 mm sieve) for quantitative analysis of extractives by Soxhlet extraction in cyclohexane–ethanol (1 : 1 v/v) for 48 h. Bagasse was also hydrolyzed by sulfuric acid (72 wt% H_2SO_4, at room temperature for 2 h). The acid-insoluble lignin was then filtered, dried to constant weight and weighed (TAPPI T222 om-02, "Acid-insoluble lignin in wood and pulp"). Monosaccharides (glucose, xylose, arabinose), organic acids (acetic acid, formic acid) and products of saccharide degradation (HMF, furfural) present in the hydrolyzate were analyzed by HPLC (high performance liquid chromatography) to determine glucan, xylan, arabinan and acetyl groups (NREL – Laboratory analytical procedure: LAP-002 "Determination of Carbohydrates in Biomass by High Performance Liquid Chromatography" and LAP-004 "Determination of Acid-Soluble Lignin in Biomass").

The monosaccharides, organic acids and degradation products were determined by HPLC in a Shimadzu CR 7A chromatograph. Cellobiose, glucose, xylose, arabinose, acetic acid and formic acid were separated on an Aminex HPX 87H column (300 × 7.8 mm, BIORAD), with Shimadzu R10-6A IR detector. The eluent was 5 mM H_2SO_4, flowing at 0.6 mL min^{-1}, at 35 °C. The furfural and HMF were separated on a Hewlett–Packard C18 column, model RP 18, with a Shimadzu SPD-10A UV-Vis detector set to 274 nm (eluent: phase acetonitrile–water 1 : 8 v/v solution and 1% acetic acid v/v, flow: 0.8 mL min^{-1}, at 25 °C). The weights of cellobiose and glucose were converted to glucan equivalents, the xylose and arabinose to xylan and arabinan, respectively, and the acetic acid to acetyl groups, by multiplying them by the hydrolysis factors: 0.95, 0.90, 0.88 and 0.717, respectively. HMF and furfural were converted to amounts of glucan and xylan by multiplying by 1.286 and 1.375, respectively.

2.3. Hot water pretreatment

The thermal hydrolysis of bagasse was performed in a stainless steel cylindrical reactor (capacity 195 cm^3, OD 5.0 cm × ID 4.0 cm, length 15.5 cm) heated in a thermostated glycerol bath (Marconi, São Paulo, Brazil). The water content of the bagasse was accounted for explicitly in the material balances. The pretreatment conditions were varied in the ranges 143.3–176.7 °C, 20–70 min and 1–11 (w/w) LSR. The reactors were immediately chilled in an ice-water bath at the end of the reaction time. Solid residues and liquid fractions were recovered by pressing to a moisture content of approximately 50% using a hydraulic press (20 ton). The hydraulic press was adapted by incorporation of a steel cylinder (5.2 cm ID × 11.5 cm height) mounted on a perforated steel plate (9 cm diameter × 1.1 cm thickness, gap: 0.5 mm diameter). The pretreated solids were pressed into the cylinder and the residual liquors were collected in the bottom of the perforated plate. Finally, the solid residue was washed with water and the washings were analyzed by HPLC.

Unwashed residual solids were air dried and the yields of hydrolysis products were determined. Samples of spent liquor were filtered and diluted (1 : 10) in water, to determine monosaccharides, organic acids and degradation products, by HPLC, employing standard procedures for the determination of sugars, organic acids and degradation products in liquid samples (NREL LAP-015 "HPLC analysis of liquid fractions of process samples for byproducts and degradation products"). The oligomers present in the spent liquors were subjected to post-hydrolysis (1 : 10 spent liquor/water, 4 wt% H_2SO_4, 121 °C, for 1 h) and then the products of post-hydrolysis were determined by HPLC. The oligomer contents were calculated from the increase in the concentration of monomers before and after post-hydrolysis.

Table 2 Levels of the parameters studied in the CCD statistical experiment

	Coded variable level				
Variable	−1.68	−1	0	+1	+1.68
T (°C)	143	150	160	170	177
t (min)	20	30	45	60	70
LSR (g g^{-1})	1	3	6	9	11

2.4. Data analysis

Since process conditions affect the efficiency of hydrothermal extraction of hemicelluloses from bagasse, these conditions have to be optimized in order to extract the greatest amount of hemicelluloses under the feasible process conditions. The optimization of process conditions was carried out by means of an experimental design, namely a central composite design (CCD). The hot water pretreatment parameters (independent variables) selected for this study were temperature (T, °C), time (t, min) and liquid–solid ratio (LSR, g g^{-1}), as shown in Table 2. A 2^3 full factorial CCD was used for the three independent variables (formed of 8 factorial points, 6 axial points and 2 center points). The axial points are at a distance of $\pm\alpha$ from the center point ($\pm\alpha = \pm 1.68$). The center points allow the experimental error and the reproducibility of the results to be determined. The total numbers of experiments (N) were calculated from the eqn (1), where n is the number of factors and n_c the number of center points.

$$N = 2^n + 2n + n_c = 2^3 + 2 \times 3 + 2 = 16 \quad (1)$$

The experimental results were analyzed with STATISTICA 10.0 (StatSoft, Inc.) routines for regression and graphical analysis.

The following second degree polynomial equation was fitted to the results:

$$Y = \alpha_0 + \alpha_1 T + \alpha_2 t + \alpha_3 \mathrm{LSR} + \alpha_{12} Tt + \alpha_{13} T\mathrm{LSR} + \alpha_{23} t\mathrm{LSR} + \alpha_{11} T^2 + \alpha_{22} t^2 + \alpha_{33} (\mathrm{LSR})^2 \quad (2)$$

where Y is the predicted response (dependent variable) and the coefficients α are adjustable constants.

The dependent variables studied were: weight loss of treated bagasse and the compositions of the spent liquor, treated solid and water washing. The results were assessed by ANOVA and the degree of fit was estimated from R^2, which indicates the variability of Y.

Table 3 Regression coefficients and R^2 measuring the correlation and significance of the models[a]

Regression coefficients	Weight loss (wt%)	X (g L^{-1})	Xyn (g L^{-1})	TXyn (g L^{-1})	AAc (g L^{-1})	X (wt %)	Xyn (wt%)	TXyn (wt%)	Lignin[b] (wt%)
α_0	387.72	262.05	−336.01	−105.41	66.999	130.06	−51.836	80.883	151.24
α_1	−5.4426	−3.4571	2.9581	−0.0841	−0.8901	−1.6561	0.2693	1.4657	−1.6747
α_{11}	0.0189	0.0112	−0.0042	0.0056	0.0031	0.0052	0.0003	0.0058	0.0054
α_2	−0.2825	−1.1160	0.6171	−0.3650	−0.4442	−0.3542	0.3980	−0.0274	−0.3290
α_{22}	0.0009	0.0004	−0.0028	−0.0024	0.0002	−0.0002	−0.0009	−0.0011	0.0002
α_3	2.5829	7.9300	16.681	23.660	2.7251	1.1669	0.9287	3.2744	0.8285
α_{33}	0.0172	0.0563	0.5477	0.5972	0.0677	−0.0002	−0.0251	−0.0300	0.0120
α_{12}	0.0024	0.0084	−0.0006	0.0068	0.0031	0.0027	−0.0015	0.0018	0.0022
α_{13}	−0.0148	−0.0480	−0.1583	−0.2006	−0.0224	−0.0058	−0.0030	−0.0153	−0.0058
α_{23}	−0.0016	−0.0313	−0.0178	−0.0453	−0.0063	−0.0056	−0.0001	−0.0082	−0.0015
R^2	0.9418	0.8806	0.9478	0.9668	0.9492	0.9107	0.8838	0.9221	0.9096

[a] X, Xyn, TXyn and AAc are xylose, xylan, total xylan and acetic acid concentrations (g L^{-1}) and contents (wt%) in the liquor respect to the initial bagasse. [b] Lignin contents in the residual bagasse.

3. Results and discussion

The chemical composition of the sugarcane bagasse used in this study was, on a dry weight basis (wt%) was: 42.6% glucans, 24.9% xylans, 3.4% arabinans, 3.6% acetyl groups, 21.7% lignin, 2.1% extractives and 1.45% ash.

The total carbohydrate content was 74.5 wt% of the bagasse including acetyl groups linked to branched chains. Xylan was the main component of the hemicelluloses (78 wt%). Acid-insoluble and soluble lignin contents were 19.5 and 2.2 wt% of the bagasse, respectively. This sugarcane bagasse chemical composition is in agreement with other reports.[13,52]

Hot water pretreatment generates the partial breakdown of hemicelluloses into soluble lower molecular weight polymers, oligosaccharides and monosaccharides. The proportion of these soluble fragments depends on the operational conditions. These experiments on the hot water pretreatment of sugarcane bagasse were carried out to assess the effectiveness of mild condition processes in hemicelluloses extraction and to obtain high-content xylo-oligosaccharide liquor with low contents of glucose and xylose degradation products. ANOVA was used to test for the significant differences between means.

The regression analysis allowed the regression coefficients of the model equations to be determined (Table 3), where X, Xyn, TXyn, AAc are xylose, xylan, total xylan and acetic acid concentrations (g L^{-1}) and contents (wt%) in the spend liquor respect to the initial bagasse. The values of R^2 (0.8806–0.9668) indicate a suitable fitted between the experimental data and model.

The effects of the temperature, time and LSR of hot water pretreatment on xylose and xylan concentrations were investigated. As expected, and according to the analysis of variance (Table 4), the concentration of xylose in the spent liquor depended on the temperature, time and LSR.

The linear effects and interactions had a significant effect ($p < 0.05$), but quadratic effect of time (t^2) was not significant. On the other hand, xylan concentration depended on temperature, LSR and the interaction of temperature with LSR ($p < 0.05$).

The response surfaces in Fig. 1 and 2 show the variation of the xylose concentration (g L^{-1}) with temperature and LSR (time: 45 min), and the dependence contour lines equate to

Table 4 Analysis of variance of the proposed models for xylose and xylan concentrations (g L^{-1})

	SS	DF	MS	*F*-value	*p*-Value
Xylose (g L^{-1}):					
T	57.703	1	57.703	5262.1	0.0088
T^2	11.542	1	11.542	1052.6	0.0197
t	17.686	1	17.686	1612.8	0.0159
t^2	0.0670	1	0.0670	6.0770	0.2453
LSR	28.128	1	28.128	2565.1	0.0126
LSR^2	2.3800	1	2.3800	217.00	0.0432
$T\,t$	12.738	1	12.738	1161.6	0.0187
T LSR	16.564	1	16.564	1510.5	0.0164
t LSR	15.901	1	15.901	1450.0	0.0167
Lack of fit	21.994	5	4.399	401.14	0.0379
Pure error	0.0110	1	0.0110		
Total SS	184.218	15			
Xylan (g L^{-1}):					
T	527.38	1	527.38	557.00	0.0270
T^2	1.6680	1	1.6680	1.7610	0.4111
t	76.065	1	76.065	80.337	0.0707
t^2	3.6170	1	3.6170	3.8200	0.3011
LSR	1020.4	1	1020.4	1077.7	0.0194
LSR^2	225.08	1	225.08	237.72	0.0412
$T\,t$	0.0750	1	0.0750	0.0790	0.8255
T LSR	180.52	1	180.52	190.66	0.0460
t LSR	5.1130	1	5.1130	5.4000	0.2587
Lack of fit	117.61	5	23.521	24.842	0.1511
Pure error	0.9470	1	0.9470		
Total SS	2271.0	15			

different LSR: (b) 1 g g^{-1}, (c) 3 g g^{-1} and (d) 6 g g^{-1}. The xylose and xylan concentrations of the spend liquor for the different temperature, time and LSR are shown in Table 5. The maximum concentrations of xylose and xylan (13.76 and 36.18 g L^{-1}, respectively), were achieved at 170 °C for 60 min, with LSR of 3 g g^{-1}. These values amount to 48.29 g $L^{-1}$of total extracted xylan and correspond to removal of almost 57 wt% of the xylan from untreated bagasse. The soluble xylan consisted mainly of xylo-oligosaccharides (74 wt% of the identified compound in the spent liquor).

According to the ANOVA results, the temperature had the most significant effect on promoting the loss of weight. The higher weight loss, (23.02 wt%) with higher xylose and xylan

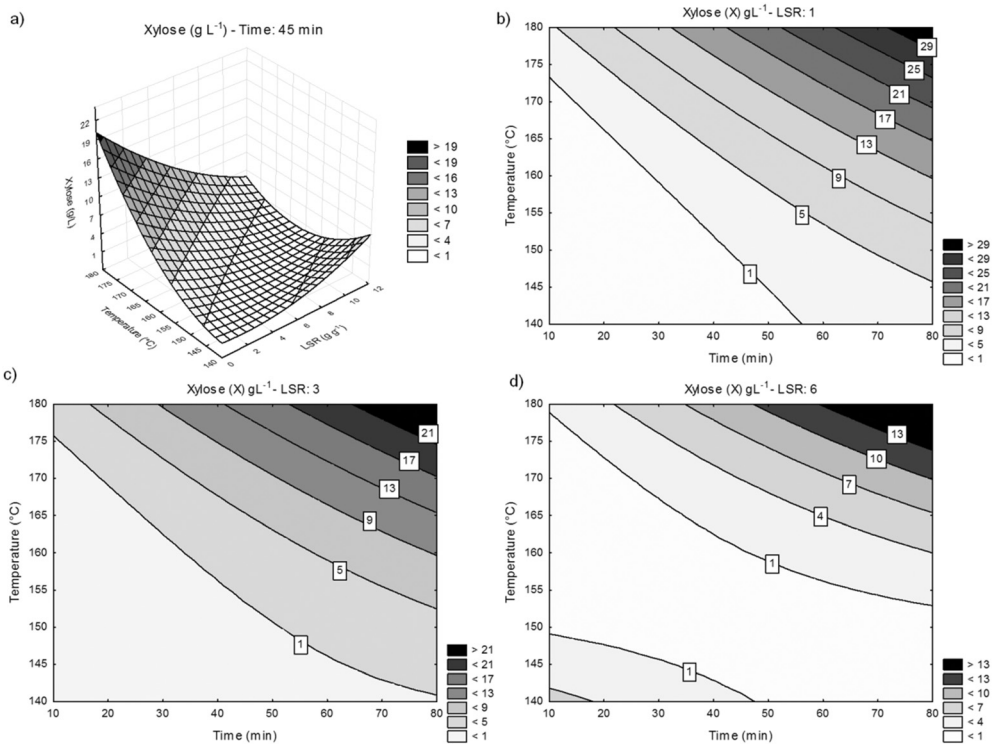

Fig. 1 (a) Fitted response surface of the dependence of xylose concentration with temperature and LSR (time: 45 min); contour lines equate to different LSR: (b) 1 g g^{-1}, (c) 3 g g^{-1} and (d) 6 g g^{-1}.

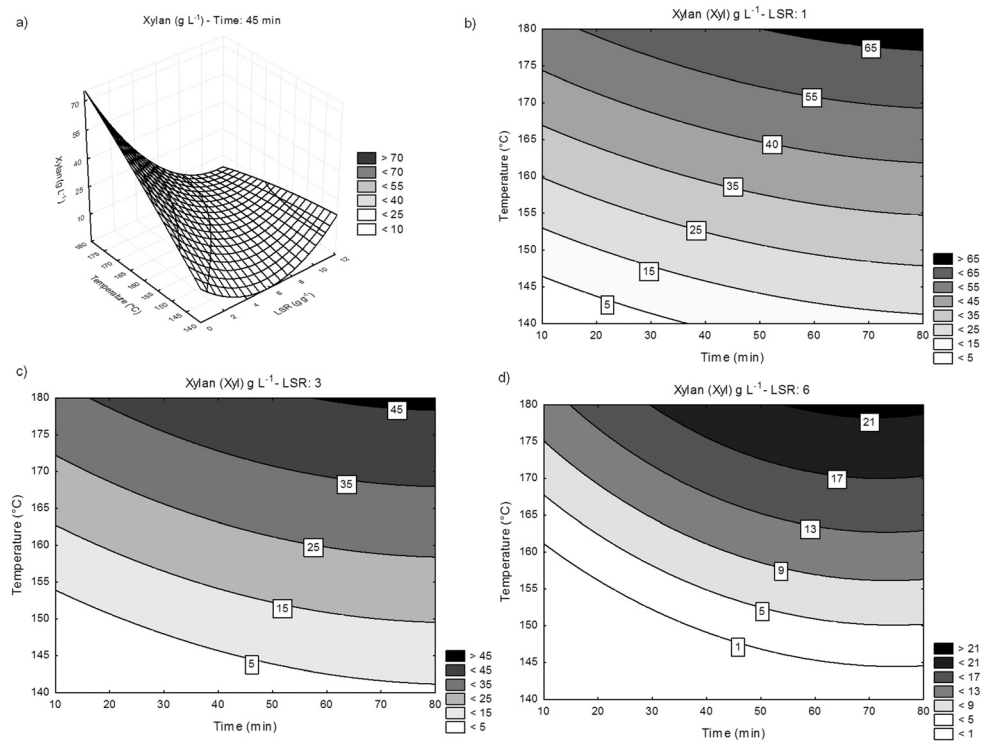

Fig. 2 (a) Fitted response surface of the dependence of xylan concentration with temperature and LSR (time: 45 min); contour lines equate to different LSR: (b) 1 g g^{-1}, (c) 3 g g^{-1} and (d) 6 g g^{-1}.

concentrations, was achieved when the bagasse was pretreated at 170 °C for 30 min, with a LSR of 3 g g^{-1} (Table 6). The yield depended on the temperature of hot water pretreatment ($p < 0.05$). Response surfaces for weight loss represent the regression equation, showing the individual and cumulative effect of the variables and their interactions (Fig. 3).

Table 5 Concentrations of xylose (*X*), xylan (Xyn) and total xylan (TXyn) in the spent liquor and composition (wt%) of spent liquor[a]

T (°C)	*t* (min)	LSR (g g^{-1})	X (g L^{-1})	Xyn	TXyn	G (wt%)	X	A	AAc	Gln	Xyn	Ara	HMF	F	Xyn*[b]
150	30	3	0.42	4.97	5.34	0.12	0.10	0.53	0.20	0.59	1.22	0.27	n.d.	n.d.	5.60
170	30	3	2.98	31.13	33.76	0.30	0.75	0.92	0.60	0.58	7.85	0.31	0.01	0.10	37.55
150	60	3	2.22	10.34	12.30	0.24	0.54	0.71	0.19	0.57	2.78	0.37	n.d.	0.02	13.29
170	60	3	13.76	36.18	48.29	0.30	3.44	1.25	1.42	0.91	9.64	0.13	0.10	0.29	56.96
150	30	9	0.20	1.27	1.44	0.43	0.16	0.55	0.21	0.29	1.10	0.28	n.d.	n.d.	4.54
170	30	9	0.93	8.49	9.31	0.60	0.76	1.26	0.52	0.65	7.93	0.34	0.02	0.56	32.73
150	60	9	0.29	3.50	3.76	0.42	0.24	0.83	0.49	0.59	3.20	0.39	n.d.	0.02	11.85
170	60	9	2.14	10.28	12.17	0.36	1.78	1.39	1.10	0.82	8.55	0.32	0.02	0.31	40.81
143	45	6	0.24	2.07	2.29	0.12	0.12	0.50	0.13	0.39	1.01	0.27	n.d.	n.d.	5.09
177	45	6	7.01	13.29	19.46	0.31	3.72	1.36	1.52	0.76	8.01	0.14	n.d.	0.05	43.47
160	20	6	0.22	1.83	2.02	0.24	0.11	0.43	0.20	0.20	1.03	0.29	n.d.	n.d.	4.21
160	70	6	1.20	12.40	13.46	0.26	0.61	1.10	0.66	0.63	6.70	0.39	n.d.	0.08	29.01
160	45	1	3.02	40.36	43.02	0.08	0.14	0.44	0.20	0.42	2.01	0.17	n.d.	n.d.	14.87
160	45	11	0.78	5.29	5.97	0.31	0.76	1.24	0.41	0.80	5.58	0.42	n.d.	n.d.	23.13
160	45	6	1.02	8.54	9.44	0.24	0.52	0.99	0.22	1.05	5.28	0.82	n.d.	0.01	19.98
160	45	6	0.87	9.92	10.68	0.24	0.44	1.04	0.42	0.69	5.21	0.36	n.d.	n.d.	22.42

[a] G: glucose, X: xylose, A: arabinose, AAc: acetic acid, Gln: glucan, Xyn: xylan, Ara: arabinano, HMF: 5-hydroxymethylfurfural, F: furfural and TXyn: total xylan = 0.88 X + Xyn. (n.d: no detected) [b] Xyn*: g equiv. total dissolved xylan/100 g xylan in initial bagasse.

Table 6 Residual bagasse composition (wt%)[a]

T (°C)	*t* (min)	LSR (g g^{-1})	WL	Gln	Xyn	Ara	Ac	Lignin	Xyn*
150	30	3	3.42	40.92	22.58	2.81	3.39	23.03	84.53
170	30	3	17.19	44.04	19.77	2.53	2.67	24.85	63.45
150	60	3	7.65	42.52	23.79	2.73	3.28	23.25	85.13
170	60	3	23.02	48.51	13.06	2.04	1.86	27.46	38.95
150	30	9	4.44	41.31	25.55	2.90	3.72	22.46	94.62
170	30	9	16.56	45.11	18.72	2.78	2.87	24.65	60.54
150	60	9	8.53	40.31	22.79	3.00	3.34	23.49	80.80
170	60	9	21.98	41.40	16.24	2.08	2.43	25.93	49.09
143	45	6	4.27	40.21	24.81	3.23	3.69	22.45	92.04
177	45	6	23.30	45.42	14.48	2.03	2.25	26.59	43.05
160	20	6	3.61	44.31	24.87	3.04	3.27	22.48	92.92
160	70	6	14.44	44.29	18.98	2.43	2.67	23.79	62.94
160	45	1	4.71	41.72	24.81	3.13	3.32	23.03	91.64
160	45	11	13.02	42.76	22.02	2.51	3.14	23.55	74.24
160	45	6	10.80	42.13	22.84	2.57	2.98	23.52	78.79
160	45	6	9.02	43.36	22.23	2.56	3.03	23.41	78.39

[a] WL: weight loss, Gln: glucan, Xyn: xylan, Ara: arabinano, Ac: acetyl group and Xyn*: Xylan remaining in residual bagasse respect to initial content.

Additionally, the amounts of degradation products in the spent liquor were relatively low, 0–0.1 wt% and 0–0.56 wt% for HMF and furfural, respectively. Acetic acid contents were 0.13–1.52 wt% and reached the maximum concentration at 170 °C for 60 min and LSR of 3 g g^{-1}. The glucose and glucan content were both lower than 1.1 wt%, which shows that cellulose was not affected by the hot water pretreatment. Arabinose contents in the spent liquor were in the range 0.44–1.36 wt%, whilst arabinan contents were slightly lower (0.13–0.82 wt%). On the other hand, over 94 wt% of the original amount of lignin remained in residual bagasse. The lignin fraction extracted during hot water pretreatments should generate several lignin degraded products (phenolic derivatives) in the residual liquors. These can act as inhibitors for the further fermentation process.[54]

For hot water pretreatment at temperatures below 150 °C and times lower 60 min, an insignificant amount of xylan was removed. The extraction of xylose starts to be significant at temperatures above 170 °C (>7 wt%). The maximum amount of extracted xylan was almost 57% of the xylan present in the untreated bagasse obtained at 170 °C for 60 min, with LSR of 3 g g^{-1}. The extracted xylan was composed by 3.4 wt% xylose and 9.1 wt% xylan, which was determined by post-hydrolysis of the spent liquor.

The total xylan content in the washing water was in the range 5–50 wt% with respect to the extracted total xylan in the hot water pretreatment. The maximum content of total xylan was in the washing water of the pretreatment at 160 °C, for 45 min, with and LSR of 1 g g^{-1}. The washing water can be re-circulated for new treatments, potentially increasing the xylo-oligosaccharide contents in the spent liquor.

4. Conclusions

The hot water pretreatment of sugarcane bagasse performed at 170 °C for 60 min, with LSR of 3 g g^{-1}, produced the

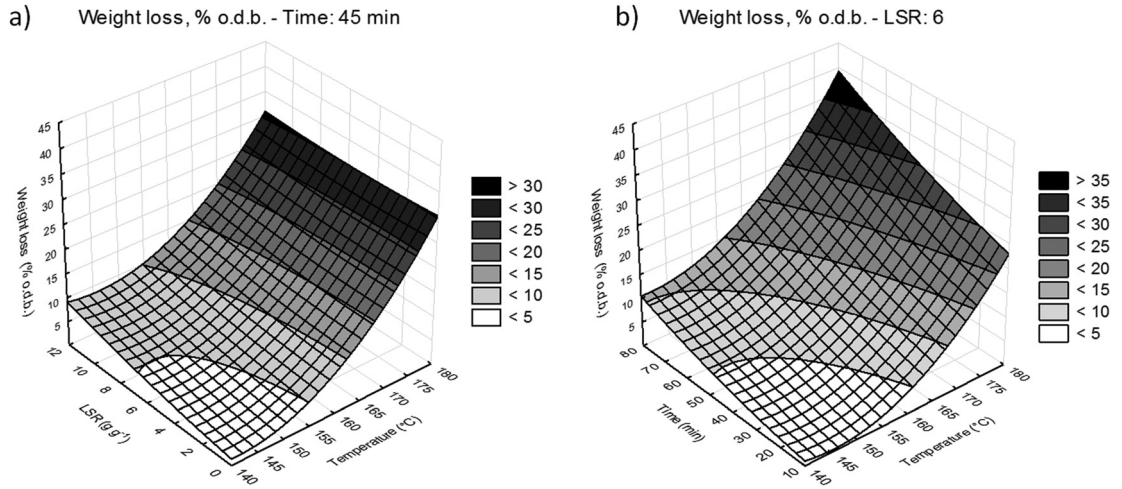

Fig. 3 Response surface 3D plot of CCD for weight loss: (a) time: 45 min and (b) LSR: 6 g g^{-1}. Effect of temperature, time and LSR for weight loss in hot water pretreatment.

maximum concentrations of xylose and xylan (13.76 and 36.18 g L^{-1}, respectively), equivalent to 48.29 g L^{-1} of xylan. The amount of xylan removed under these conditions was almost 57% of the xylan present in the untreated bagasse. The dissolved xylan is mainly composed of xylo-oligosaccharides (74 wt%).

Glucose and glucan contents in the spent liquor were less than 1.1%, which shows that cellulose was not hydrolyzed by the hot water pretreatment. Low liquid–solid ratio (LSR) provided a simple and ambient friendly means to produce high-content xylo-oligosaccharides spend liquor with enormous potential for industrial applications.

Acknowledgements

This research was supported by Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP) and Oxiteno. The authors are grateful to Instituto de Química de São Carlos (IQSC), USP at São Carlos (SP, Brazil) and Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina.

References

1 J. H. Clark, V. Budarin, F. E. I. Deswarte, J. J. E. Hardy, F. M. Kerton, A. J. Hunt, R. Luque, D. J. Macquarrie, K. Milkowski, A. Rodriguez, O. Samuel, S. J. Tavener, R. J. White and A. J. Wilson, *Green Chem.*, 2006, **8**, 853–860.
2 *FAOSTAT – Sugarcane*, Food and Agriculture Organization of the United Nations (FAO). http://www.fao.org (15/02/2012).
3 Acompanhamento da Safra Brasileira de Cana de Açúcar. 05/2011, Ministério da Agricultura, Pecuária e Abastecimento. http://www.conab.gov.br. (10/07/2011).
4 G. J. M. Rocha, A. R. Gonçalves, B. R. Oliveira, E. G. Olivares and C. E. V. Rossell, *Ind. Crops Prod.*, 2012, **35**, 274–279.
5 L. P. Novo, L. V. A. Gurgel, K. Marabezi and A. A. da S. Curvelo, *Bioresour. Technol.*, 2011, **102**, 10040–10046.
6 C. M. Area, F. E. Felissia and M. E. Vallejos, *Cellul. Chem. Technol.*, 2009, **43**, 271–279.
7 F. Carvalheiro, L. C. Duarte and F. M. Gírio, *Ind. Res.*, 2008, **67**, 849–864.
8 F. B. Sedlmeyer, *Food Hydrocolloids*, 2011, **25**, 1891–1898.
9 S. F. Zhang and H. Yang, *Adv. Mater. Res.*, 2011, **236–238**, 1174–1177.
10 C. Vila, J. Romero, J. L. Francisco, G. Garrote and J. C. Parajó, *Bioresour. Technol.*, 2011, **102**, 5251–5254.
11 J. C. García, M. A. M. Zamudio, A. Pérez, F. López and J. L. Colodette, *Environ. Prog.*, 2011, **30**, 92–101.
12 B. Zhang and A. Shahbazi, *J. Pet. Environ. Biotechnol.*, 2011, **2**, 108.
13 Z. Liu, P. Fatehi, M. S. Jahan and Y. Ni, *Bioresour. Technol.*, 2011, **102**, 1264–1269.
14 B. Yang and C. E. Wyman, *Environ. Eng.*, 2008, **2**, 26–40.
15 F. Carvalheiro, *Bioresour. Technol.*, 2004, **91**, 93–100.
16 F. Carvalheiro, T. Silva-fernandes, H. Pereira, Nordic Wood Biorefinery Conference (NWBC), Helsinki, 2009, pp. 96–102.
17 G. Garrote, H. Dominguez and J. Parajo, *J. Food Eng.*, 2002, **52**, 211–218.
18 G. Garrote, E. Falqué, H. Domínguez and J. C. Parajó, *Bioresour. Technol.*, 2007, **98**, 1951–1957.
19 A. Romaní, G. Garrote, F. López and J. Carlos, *Cellulose*, 2011, **102**, 5896–5904.
20 A. Romaní, G. Garrote, J. L. Alonso and J. C. Parajó, *Bioresour. Technol.*, 2010, **101**, 8706–8712.
21 G. Garrote, H. Domínguez and J. C. Parajó, *Holz Roh- Werkst.*, 1999, **57**, 191–202.
22 F. M. Gírio, C. Fonseca, F. Carvalheiro, L. C. Duarte, S. Marques and R. Bogel-Łukasik, *Bioresour. Technol.*, 2010, **101**, 4775–4800.
23 N. Mosier, C. Wyman, B. Dale, R. Elander, Y. Y. Lee, M. Holtzapple and M. Ladisch, *Bioresour. Technol.*, 2005, **96**, 673–686.
24 R. F. Branco, J. C. Santos, L. Y. Murakami, S. I. Mussatto, G. Dragone and S. S. Silva, *Process Biochem.*, 2007, **42**, 258–262.
25 J. Zhang, X. Wang, D. Chu, Y. He and J. Bao, *Bioresour. Technol.*, 2011, **102**, 4480–4488.
26 M. G. A. Felipe, M. Vitolo, I. M. Mancilha and S. S. Silva, *Biomass Bioenergy*, 1997, **13**, 11–14.
27 S. I. Mussatto and I. C. Roberto, *Process Biochem.*, 2004, **39**, 1433–1439.
28 B. Rivas, J. M. Dominguez, H. Dominguez and J. C. Parajo, *Enzyme Microb. Technol.*, 2002, **31**, 431–438.
29 S. Lima, M. Pillinger and A. A. Valente, *Catal. Commun.*, 2008, **9**, 2144–2148.
30 G. Marcotullio and W. De Jong, *Green Chem.*, 2010, **12**, 1739–1746.
31 R. Weingarten, J. Cho, W. C. Conner and G. W. Huber, *Green Chem.*, 2010, **12**, 1423–1429.
32 C. R. Silva, T. C. Zangirolami, J. P. Rodrigues, K. Matugi, R. C. Giordano and R. L. C. Giordano, *Enzyme Microb. Technol.*, 2012, **50**, 35–42.
33 C. F. Crespo, M. Badshah, M. T. Alvarez and B. Mattiasson, *Bioresour. Technol.*, 2012, **103**, 186–191.
34 H. Tamakawa, S. Ikushima and S. Yoshida, *Biosci., Biotechnol., Biochem.*, 2011, **75**, 1994–2000.
35 E. R. Borges and N. Pereira, *J. Ind. Microbiol. Biotechnol.*, 2011, **38**, 1001–1011.

36 A. Escalante, A. Goncalves, A. Bodin, A. Stepan, C. Sandstrom, G. Toriz and P. Gatenholm, *Carbohydr. Polym.*, 2012, **87**, 2381–2387.
37 B. S. Kayserilioglu, U. Bakir, L. Yilmaz and N. Akkas, *Bioresour. Technol.*, 2003, **87**, 239–246.
38 A. Hoeije, E. Sternemalm, S. Heikkinen, M. Tenkanen and P. Gatenholm, *Biomacromolecules*, 2008, **9**, 2042–2047.
39 B. S. Kayserilioglu, U. Bakir, L. Yilmaz and N. Akkas, *Bioresour. Technol.*, 2003, **87**, 239–246.
40 K. Hettrich and C. Fanter, *Macromol. Symp.*, 2010, **294**, 141–150.
41 I. Gabrielii and P. Gatenholm, *J. Appl. Polym. Sci.*, 1998, **69**, 1661–1667.
42 J. L. Ren, X. W. Peng, F. Peng and R. C. Sun, *Adv. Mater. Res.*, 2011, **239–242**, 463–467.
43 J. S. Stevanic, C. Joly, K. S. Mikkonen, K. Pirkkalainen, R. Serimaa, C. Remond, G. Toriz, P. Gatenholm, M. Tenkanen and L. Salmen, *J. Appl. Polym. Sci.*, 2011, **122**, 1030–1039.
44 S. Dammstrom and P. Gatenholm, in *Characterization of the Cellulosic Cell Wall*, ed. D. D. Stokke and L. H. Groom, Wiley, 1st edn, 2006, ch. 5, pp. 53–63.
45 A. Saxena, T. J. Elder, S. Pan and A. J. Ragauskas, *Composites, Part B*, 2009, **40**, 727–730.
46 A. Saxena and A. J. Ragauskas, *Carbohydr. Polym.*, 2009, **78**, 357–360.
47 T. J. Heinze, *Abstr. Paper Am. Chem. Soc*, 2002, **223**, U142–U143.
48 A. Ebringerova and T. Heinze, *Macromol. Rapid Commun.*, 2000, **21**, 542–556.
49 M. Saska and E. Ozer, *Biotechnol. Bioeng.*, 1995, **45**, 517–523.
50 G. Garrote, H. Domínguez and J. C. Parajó, *J. Chem. Technol. Biotechnol.*, 1999, **74**, 1101–1109.
51 S. E. Jacobsen and C. E. Wyman, *Ind. Eng. Chem. Res.*, 2002, **41**, 1454–1461.
52 F. Carvalheiro, G. Garrote, J. C. Parajó, H. Pereira and F. M. Gírio, *Biotechnol. Prog.*, 2008, **21**, 233–243.
53 R. Sánchez, A. Rodríguez, J. C. García, A. Rosal and L. Jiménez, *Bioresour. Technol.*, 2011, **102**, 1308–1315.
54 J. X. Sun, X. F. Sun, R. C. Sun and Y. Q. Su, *Carbohydr. Polym.*, 2004, **56**, 195–204.