



# Production of Xylose from Sorghum Straw Using Hydrochloric Acid

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## ABSTRACT

Xylose is a hemicellulosic sugar mainly used for its bioconversion to xylitol. Sorghum straw is a raw material for xylose production that has been studied scarcely. The objective of this work was to study the xylose production by hydrolysis of sorghum straw with hydrochloric acid at 122 °C. Several concentrations of HCl (2–6%) and reaction time (0–300 min) were evaluated. Kinetic parameters of mathematical models for predicting the concentration of xylose, glucose, acetic acid and furfural in the hydrolysates were found. Optimal conditions for hydrolysis were 6% HCl at 122 °C for 70 min, which yielded a solution with 16.2 g xylose/L, 3.8 g glucose/L, 2.0 g furfural/L and 1.9 g acetic acid/L.

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## INTRODUCTION

Sorghum straw is a renewable, cheap and widely available resource, which is commonly used as feedstuffs. The hydrolysis of sorghum straw to produce xylose solutions could be a good alternative use for this abundant resource. The hydrolysis of sorghum straw to obtain xylose solutions has a double consequence, the elimination of a waste and the production of a value-added product that increases the economy of the process. Among the treatments tried with other lignocellulosic materials, dilute acid hydrolysis appears to be in the best position from the economic viewpoint<sup>1</sup>. In a single step, this technology causes several alterations in the raw materials, including hemicellulosic sugars release<sup>2,3</sup>.

The hydrolysis reactions in dilute-acid medium are very complex, mainly because the substrate is in

a solid phase and the catalyst in a liquid phase. The mechanism of the hydrolysis reaction includes<sup>4–6</sup>: (i) diffusion of protons through the wet lignocellulosic matrix; (ii) protonation of the oxygen of a heterocyclic ether bond between the sugar monomers; (iii) breaking of the ether bond; (iv) generation of a carbocation as intermediate; (v) solvation of the carbocation with water; (vi) regeneration of the proton with cogeneration of the sugar monomer, oligomer or polymer depending on the position of the ether bond; (vii) diffusion of the reaction products in the liquid phase if allowed by their form and size (some large oligomers cannot cross the matrix); (viii) restarting in the second step. Therefore, due to the difficult modelling of these processes, empirical models are commonly used.

Dilute acids lead to a limited hydrolysis called prehydrolysis. This consists in the hydrolysis of the hemicellulosic fraction, leaving the cellulose and lignin fractions almost unaltered. Sulphuric<sup>7,8</sup>, hydrochloric<sup>9</sup>, hydrofluoric<sup>10</sup> or acetic<sup>11</sup> acids are commonly employed as catalysts. The acid hydrolysis

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of the hemicellulosic fraction of sorghum straw can also lead to a valuable solid residue mainly formed from cellulose and lignin. This waste can be used in the production of glucose solutions<sup>12</sup> or for the production of paper pulp<sup>13</sup>. Thus, the acid hydrolysis can be conceived as the first stage of an integrated strategy for sorghum straw utilization.

The main application of xylose is its bioconversion to xylitol, a functional sweetener with important technological properties like anticarcinogenicity, low caloric value and negative heat of dissolution<sup>14–16</sup>.

The economic interest in xylitol production can be enhanced if the needed xylose solutions can be obtained from the hydrolysis of low-cost lignocellulosic wastes.

To our knowledge, there are scarce studies about the hydrolysis of sorghum straw. Only the sulphuric-acid hydrolysis has been studied<sup>17</sup> and no reports about hydrochloric-acid hydrolysis exist. The objective of this work was to study the hydrolysis of sorghum straw at 122 °C using dilute hydrochloric acid. Kinetic parameters of mathematical models for predicting the concentration of xylose, glucose, acetic acid and furfural in the hydrolysates were found and used to optimise the process.

## EXPERIMENTAL

### Materials

*Sorghum bicolor* straw collected in a local farm (Tamaulipas, Mexico) was used as raw material. The straw was air-dried, milled, screened to select the fraction of particles with a size lower than 0.5 mm and homogenized in a single lot.

### Analytical methods

Analyses of the main fractions (cellulose, hemicelluloses and Klason lignin) were carried out using a quantitative acid hydrolysis under standard conditions<sup>18</sup>. The main composition of the sorghum straw was: cellulose, 35 ± 1%; xylan, 19 ± 1%, hemicelluloses, 24 ± 1%, lignin, 25 ± 1%.

Treatments were performed at 122 °C in media containing 2, 4 or 6 g HCl/100 g liquor using a charge of 1 g straw/10 g liquor. At given reaction times, samples of liquors were taken from the reaction media and analysed. The samples were diluted with water (1/10 v/v), centrifuged to separate the insolubilized phenolic fraction and analysed by UV-Vis spectroscopy at 280 nm for

furfural and by HPLC for glucose, xylose and acetic acid. The HPLC analyses were carried out using a Transgenomic ION-300 column (oven temperature = 45 °C) with isocratic elution (flow rate = 0.4 ml/min; mobile phase: H<sub>2</sub>SO<sub>4</sub> 0.005 N).

### Statistical analysis

All tests were carried out in triplicate and the related data were expressed as averages values. Non-linear regression analyses of experimental data were performed with a commercial optimization routine based on the Newton's method (Solver, Microsoft Excel 2000, Microsoft Corporation, Redmont, WA, USA), by minimizing the sum of the squares of deviations between experimental and calculated values as previously described<sup>19</sup>.

## RESULTS

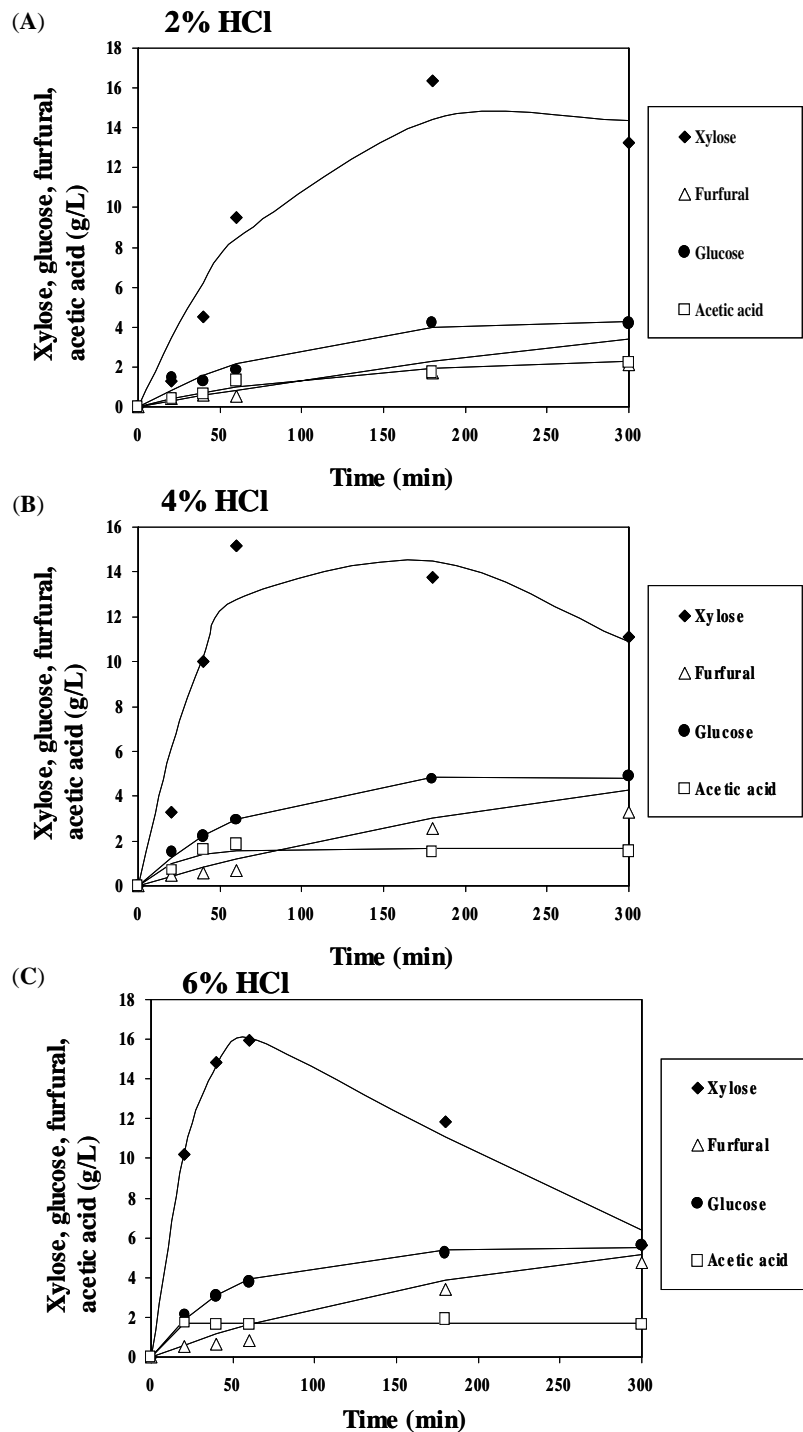
The main fractions of sorghum straw were in the range of other herbaceous materials, such as rice and barley straw<sup>18</sup>. The high content in xylan (19 ± 1%) makes this waste adequate for xylose production. The potential concentration of xylose corresponds to the quantitative conversion of xylan to xylose. The solid/liquid ratio used was 22.3 g xylose/L.

Hydrolysates were obtained using HCl at 122 °C. The range of HCl concentration (2–6%) and temperature studied (122 °C) were selected according to the values reported for dilute acid hydrolysis of lignocellulosic materials<sup>3,17,20</sup>.

Figure 1(A) shows the concentration of xylose, furfural, glucose and acetic acid released during the hydrolysis of sorghum straw performed with 2% HCl at 122 °C. Xylose concentration reached a maximum value (16.38 g/L) after 180 min and then decreased with increasing the reaction time while furfural concentration progressively increased up to 2.09 g furfural/L. These results suggest that undesired secondary reactions could have been responsible for xylose decomposition to furfural.

During the hydrolysis of sorghum straw, other sugars were released to liquors, mainly glucose, which can proceed from the cellulosic fraction or from some heteropolymers of the hemicellulosic fraction. Glucose concentration progressively increased up to 4.21 g/L, even after long time, which suggests that decomposition reactions of glucose were negligible using HCl.

Acetic acid, which derives from the hydrolysis of the acetyl groups bound to the hemicellulosic



**Figure 1** Experimental and predicted dependence of the products generate during the hydrolysis at 2, 4 or 6% HCl at different time.

monomers, reached a maximum concentration of 2.2 g acetic acid/L using 2% HCl and a reaction time of 300 min.

Figure 1(B) shows the concentration of xylose, furfural, glucose and acetic acid released during

the hydrolysis of sorghum straw at 122 °C with 4% HCl. Xylose concentration reached a maximum (15.14 g/L) at 60 min while glucose and furfural concentrations grew up to 4.9 and 3.29 g/L, values that are 17.5 and 57% higher than using 2%

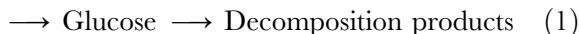
HCl, respectively. Acetic acid concentration kept at 1.6 g/L.

Figure 1(C) shows the concentration of products released during the hydrolysis of sorghum straw at 122 °C with 6% HCl. Xylose concentration had a maximum (15.96 g/L) at 60 min and then decreased drastically with a slope higher than using 4% HCl. Xylose concentration obtained was 71.6% of the potential concentration. Glucose concentration reached up to 5.64 g/L, 35% higher than using 2% HCl. Furfural and acetic acid also increased up to 4.7 and 1.6 g/L, respectively.

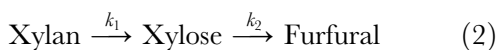
## DISCUSSION

Due to the difficulty to find a strict mechanism for hydrolysis reactions, it is usual to use simplified models to determine the kinetics of the hydrolysis of lignocellulosic materials. The models proposed in the literature use pseudo-homogeneous irreversible first-order reactions. The first model used successful was proposed by Saeman<sup>21</sup> in 1945:

Cellulose



This model was designed for the hydrolysis of cellulose from fir wood using sulphuric acid. The model was also applied to the hydrolysis of the hemicellulosic fraction<sup>17,22</sup>. Therefore, it can be generalized for hemicellulosic hydrolysis to Eq. (2):



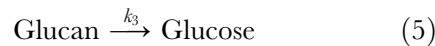
where  $k_1$  is the rate of xylose release ( $\text{min}^{-1}$ ) and  $k_2$  is the rate of xylose decomposition ( $\text{min}^{-1}$ ). Based on this reaction model and solving differential equations leads to Eq. (3), which expresses xylose concentration  $[X]$  as a function of time ( $t$ ):

$$[X] = \frac{k_1[Xn_0]}{k_2 - k_1} (e^{-k_1 t} - e^{-k_2 t}) + [X_0]e^{-k_2 t} \quad (3)$$

The initial xylan and xylose concentrations are defined at time 0 as  $[Xn_0]$  and  $[X_0]$ , respectively. Assuming that  $[X_0]$  is close to 0, a simplification of the model yields:

$$[X] = \frac{k_1[Xn_0]}{k_2 - k_1} (e^{-k_1 t} - e^{-k_2 t}) \quad (4)$$

A similar model can be developed to describe the increase in glucose concentration, but in this case the decomposition reactions were negligible and the model can be simplified as follow:



where  $k_3$  is the rate of glucose release ( $\text{min}^{-1}$ ).

Solving differential equations leads to Eq. (6), which expresses the glucose concentration  $[G]$  as a function of time ( $t$ ).

$$[G] = [G_0] \cdot (1 - e^{-k_3 t}) \quad (6)$$

where  $[G_0]$  is the potential glucose concentration which was estimated by regression.

A similar model has also been developed for furfural concentration. In this case, the rate for furfural generation,  $k_2$ , is the same that for xylose decomposition (Eq. (2)). The equation that expresses furfural concentration  $[F]$  as function of time is:

$$[F] = [F_0] \cdot (1 - e^{-k_2 t}) \quad (7)$$

where  $[F_0]$  is the potential concentration of furfural (g/L) that was determined by regression analysis. The modelling of acetic acid can be developed on the basis of the equation:



where the regression parameter  $k_4$  is the rate of its generation ( $\text{min}^{-1}$ ).

Acetic acid concentration (Ac) can be expressed as a function of time ( $t$ ) as:

$$\text{Ac} = \text{Ac}_0 \cdot (1 - e^{-k_4 t}) \quad (9)$$

where the regression parameter  $\text{Ac}_0$  is the potential concentration of acetic acid and  $k_4$  the rate of acetic acid generation ( $\text{min}^{-1}$ ).

Equations (4), (6), (7) and (9) have been applied to model the hydrolysis of sorghum straw with hydrochloric acid. Non-linear regression analyses were performed to obtain the kinetic parameters and constants. The results were statistically evaluated with the coefficient of determination  $r^2$  to establish the reliability of the models.

### Kinetic modelling of xylose concentration

Xylan concentration,  $[Xn_0]$ , was fixed at 22.27 g/L, taking into account the solid/liquid ratio and the stoichiometric ratios. Table I shows the kinetic and statistical parameters obtained for xylose released during the hydrolysis of sorghum straw at 122 °C.

Figure 1 show the experimental and predicted data for these hydrolyses at each concentration of HCl. Comparing the values of  $k_1$  and  $k_2$ , the reactions of xylose generation appear to be faster than the decomposition reactions, even though both accelerated with increasing HCl concentration. The determination coefficient ( $r^2$ ) showed a good agreement between experimental and predicted data for all regressions. The models predict that increasing the HCl concentration the maximum xylose concentration can be obtained after less reaction time.

A generalized model for predicting all products was developed. Kinetic parameters were correlated with the HCl concentration ( $C_a$ ) by means of the empirical equation:

$$k_j = k_0 C_a^n \quad (10)$$

where  $j$  is an integer in the range 1–4,  $k_0$  and  $n$  are regression parameters and  $C_a$  is the acid concentration expressed as % (w/w). In the xylose model,  $k_1$  correlated with HCl concentration as shown in Eq. (11) of Table II and  $k_2$  can be determined by Eq. (12) of the same table. The coefficient  $r^2$  showed good values for all regressions.

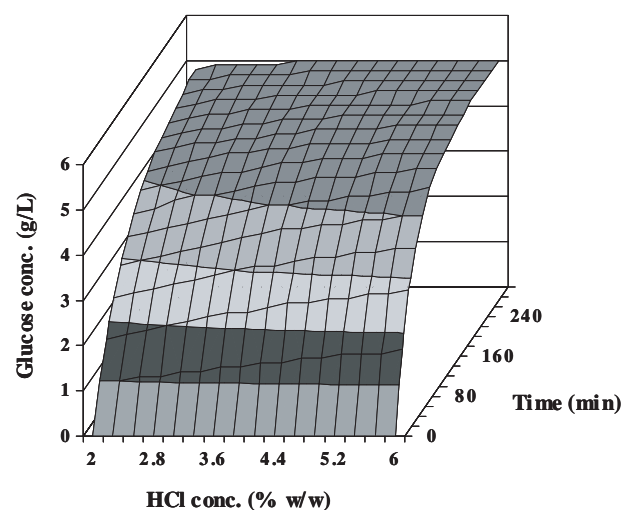
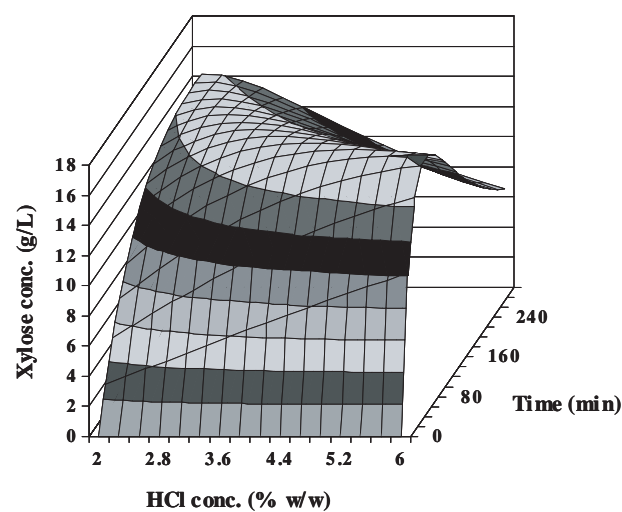
**Table I** Kinetic and statistical parameters of products released during the HCl hydrolysis of sorghum straw at 122 °C

	2% HCl	4% HCl	6% HCl
<b>Xylose</b>			
$k_1$ (min <sup>-1</sup> )	0.00856	0.01678	0.03327
$k_2$ (min <sup>-1</sup> )	0.00177	0.00299	0.00468
$r^2$	0.9494	0.9265	0.9942
<b>Glucose</b>			
$k_3$ (min <sup>-1</sup> )	0.01082	0.01543	0.02041
$[G_0]$	4.53	4.97	5.54
$r^2$	0.9644	0.9946	0.9959
<b>Furfural</b>			
$k_2$ (min <sup>-1</sup> )	0.00177	0.00299	0.00468
$[F_0]$	8.29	7.22	6.82
$r^2$	0.9668	0.9841	0.9773
<b>Acetic acid</b>			
$k_4$	0.00856	0.04672	0.19921
$[Ac_0]$	2.45	1.66	1.73
$r^2$	0.9562	0.9035	0.9711

Combining Eqs (11) and (12) of Table II with the kinetic models, it is possible to predict the xylose concentration for any time and acid concentration over the range under consideration (0–300 min and 2–6% HCl). Figure 2 shows how the generalized models predict the dependence of xylose

**Table II** Generalized models for the prediction of kinetic parameters of sorghum straw hydrolysis with HCl

Products	Models	$r^2$
Xylose	$k_1 = 0.00261 C_a^{1.41}$ (11)	0.9833
Xylose or furfural	$k_2 = 0.0009 C_a^{0.93}$ (12)	0.9891
Glucose	$k_3 = 0.00702 C_a^{0.59}$ (13)	0.9929
Acetic acid	$k_4 = 0.00037 C_a^{3.51}$ (14)	0.9868



**Figure 2** Prediction of the generalized model for the dependence of xylose and glucose concentration on HCl concentration and time.

concentration on HCl concentration and time. This kind of response surface allows evaluating several conditions to achieve the same result. Therefore, it is possible to select conditions leading to the highest xylose concentration and the minimum concentrations of degradation product by comparing response surfaces for different products of sorghum hydrolysis. The response surface can be also used to obtain the optimum xylose concentration. The models predicted that maximum xylose concentration (16.15 g/L) is achieved using 6% HCl after 70 min. Higher xylose concentrations can not be obtained even if longer reaction times were used, because the degradation reactions tend to consume xylose.

### Kinetic modelling of glucose concentration

In the model for glucose,  $[G_0]$  cannot be obtained experimentally because the glucose released in the hydrolysis can proceed from both hemicellulosic heteropolymers and cellulose, even though cellulose is not usually hydrolysed over the range of operational conditions tested; therefore,  $[G_0]$  has been introduced as a new regression parameter. Both values obtained by regression for  $[G_0]$  and  $k_3$  increased with the concentration (Table I). The glucan susceptible of hydrolysis was in the range 4.53–5.54 g/L, which represents 11.6–14.2% of total glucan in sorghum straw. This result compares very well with those reported for the hydrolysis of sorghum straw with sulphuric acid<sup>17</sup>. In that case, the glucan concentration susceptible of hydrolysis was in the range (6.76–7.22 g/L), corresponding to 17.3–18.5% of the total glucan. It can be inferred that the treatment with hydrochloric is more selective, being able to hydrolyse the hemicellulosic fraction leaving cellulose and lignin in the solid phase. This is adequate for subsequent processing of the solid residue.

The kinetic parameter  $k_3$  increased with the HCl concentration and  $r^2$  showed a good agreement between experimental and predicted data. Figure 1 shows the experimental and predicted data for these hydrolyses at each concentration of HCl.

A generalized model for predicting glucose was also developed. Kinetic parameters  $k_3$  were correlated with the HCl concentration ( $C_a$ ) by mean of the empirical equation (13) showed in Table II. Figure 2 shows how the generalized models predict the dependence of glucose concentration on HCl concentration and time. It can be observed a continuous increase of this parameter with the time course and

no clear dependence on HCl concentration. The models predict a maximum glucose concentration (5 g/L) at the most severe conditions (6% HCl and 300 min), but less than 4 g/L were obtained after  $t < 60$  min for any HCl concentration. The generalized models proposed here are useful tools for selecting operational conditions that ensure the lower glucose concentration and the maximum xylose concentration.

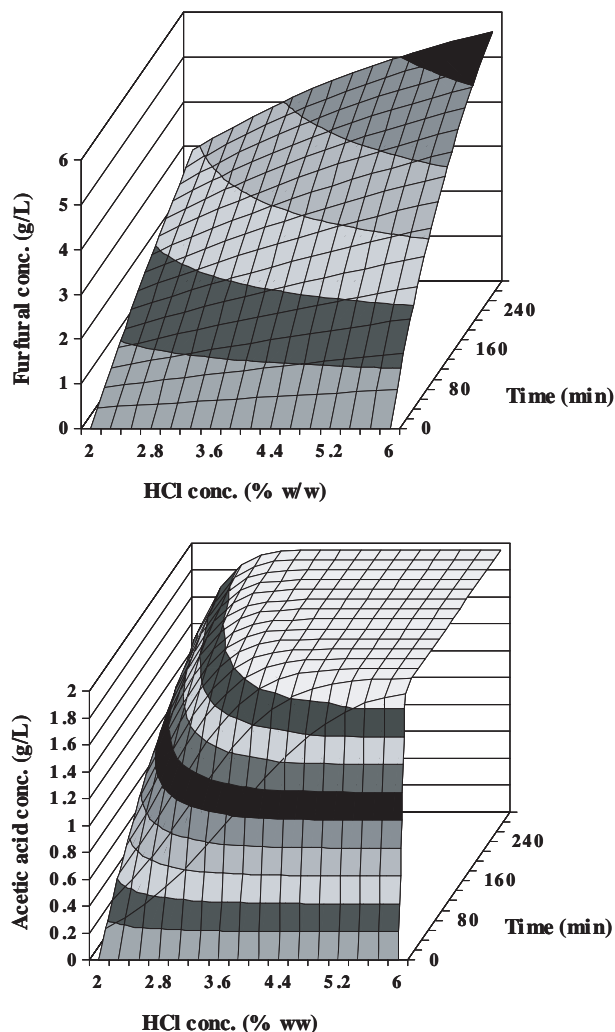
### Kinetic modelling of furfural concentration

Furfural is the main degradation product in the hydrolysis of lignocellulosic materials to obtain xylose. Table I shows the kinetic and statistical parameters fitting the model for furfural generated in the hydrolysis of sorghum straw at 122 °C, whereas Figure 1 shows the experimental and predicted data for furfural at different HCl concentrations. The coefficient  $k_2$  was fixed with the value previously obtained in the fitting of the model for xylose concentration while  $[F_0]$  was the regression parameter that varied over the range 8.29–6.82 g/L for each regression. These values are higher than the values obtained using sulphuric acid as catalyst (4 g/L)<sup>17</sup>.

Equation (12) of Table II was used for furfural modelling because the formation of this inhibitor occurred at the same rate as xylose degradation. Figure 3 shows that the model predicts a maximum furfural concentration (5.6 g/L) at the most severe conditions (6% HCl and 300 min). These results suggest that a low reaction time should be selected to minimize the formation of furfural, which is an inhibitor of the growth of microorganisms involved in the subsequent fermentation steps.

### Kinetic modelling of acetic acid concentration

Table I also shows the kinetic and statistical parameters fitting the model for the acetic acid generated during the hydrolysis of sorghum straw at 122 °C, whereas Figure 1 shows the experimental and predicted data for acetic acid concentration at different HCl concentrations.  $[Ac_0]$  varied slightly over the range 1.66–2.45 g/L. The high values of the determination coefficients ( $r^2$ ) confirm that the models fit very well ( $r^2 = 0.95$  and  $0.97$ ) with the exception of the test at 4% H<sub>2</sub>SO<sub>4</sub> ( $r^2 = 0.90$ ). The values of  $k_4$  showed a quick increase with HCl concentration. Figure 3 shows that the model predicts a stable concentration (1.95 g/L) for a large area in the graph



**Figure 3** Prediction of the generalized model for the dependence of furfural and acetic acid concentration on HCl concentration and time.

corresponding with a wide pool of operational conditions.

### Overall optimization

If the hydrolysates of sorghum straw are going to be used as fermentation media and the solid residue have to be processed for glucose production it is important to obtain sugar solutions (mainly xylose) with low concentration of inhibitors (furfural and acetic acid). The generalized models show that a complex situation take place: (i) more than 13 g xylose/L can be obtained using several combinations of time and HCl concentration; (ii) low reaction time permits to reduce the release of glucose and

generation of furfural, (iii) acetic acid achieves a stable concentration (1.95 g/L) after 100 min and with 4% HCl.

Based on these considerations, the condition ensuring the maximum xylose concentration can be considered as optimum for the overall hydrolysis of sorghum straw with HCl, if the concentrations of inhibitors are not critical for the microorganism that will be grown on the hydrolysates. This operational condition is achieved at 6% HCl and 70 min and the models predict hydrolysates containing 16.18 g xylose/L, 3.79 g glucose/L, 2.05 g furfural/L and 1.95 g acetic acid/L. Since the concentrations of furfural and acetic acid could be enough to inhibit the growth of some yeasts, in this case, other conditions should be selected. For instance, for 2% HCl and 170 min, the models predict hydrolysates with 13.08 g xylose/L, 4.18 g glucose/L, 1.82 g furfural/L and 1.00 g acetic acid/L.

The developed models are useful tools for further technical and economic studies. The application of HCl for the hydrolysis of sorghum straw showed a very good selectivity.

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