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Abstract: A method for the selective extraction of hemicellulose from wheat straw involving cold alkaline extraction and subsequent separation by precipitation with ethanol is proposed. Wheat straw affords selective separation of the hemicellulose fraction from the cellulose and lignin fractions with the proposed method. The hemicellulose yield was optimized by using a 2n factor design to examine the influence of temperatures from 20 to 40 ºC, operation times from 30 to 60 min and alkali concentrations from 80 to 120 g/L. The optimum conditions for cold alkaline extraction of hemicellulose from wheat straw were thus found to be a temperature of 40 C, an operation time of 90 min and an alkali concentra¬tion of 100 g/L. These conditions allowed 56.1% of all hemicellulose initially present in the raw material, and 59.1% of the lignin, to be extracted. Subsequent separation of hemicellulose in the liquid phase from the cold alkaline extraction by precipitation with ethanol provided a fraction containing 39.4% of all hemicellulose and only 12% of all lignin in the raw material.

HIGHLIGHTS

- A method for the selective extraction of cold alkaline extraction.
- The method for extraction was optimized by using a 2^n factor.
- With this method allows 56.1% of all hemicellulose present in the raw material.

Search for Optimum Conditions of Wheat Straw Hemicelluloses Cold Alkaline Extraction Process

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Abstract

A method for the selective extraction of hemicellulose from wheat straw involving cold alkaline extraction and subsequent separation by precipitation with ethanol is proposed. Wheat straw affords selective separation of the hemicellulose fraction from the cellulose and lignin fractions with the proposed method. The hemicellulose yield was optimized by using a 2^n factor design to examine the influence of temperatures from 20 to 40 °C, operation times from 30 to 60 min and alkali concentrations from 80 to 120 g/L. The optimum conditions for cold alkaline extraction of hemicellulose from wheat straw were thus found to be a temperature of 40 °C, an operation time of 90 min and an alkali concentration of 100 g/L. These conditions allowed 56.1% of all hemicellulose initially present in the raw material, and 59.1% of the lignin, to be extracted. Subsequent separation of hemicellulose in the liquid phase from the cold alkaline extraction by precipitation with ethanol provided a fraction containing 39.4% of all hemicellulose and only 12% of all lignin in the raw material.

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1. Introduction

Biorefinery is an overall concept of an integrated and diversified processing plant where lignocellulosic biomass feedstocks are converted into a wide range of valuables products, much likewise to petroleum refineries [1]. Biorefinery is one of the procedures facilitating sustainable development and resource renewal, which rests on the search for new lignocellulosic biomass resources and their proper use [2]. A number of methods enabling increasingly efficient use of agro-industrial residues such as sugarcane bagasse, wheat straw or maize stalks have been developed in recent years. These residues provide low-cost raw materials for the obtainment of end-products with a high added value [3].

Wheat straw, which is an abundant residue in many countries, has the potential for use as a low-cost raw material for the industrial production of higher-added value products. Europe alone is estimated to produce more than 170 million ton of wheat straw each year [4]. This amount is large enough for straw to be used as a source of renewable materials (particularly for the production of chemical derivatives of cellulose, hemicellulose and lignin [5].

Wheat straw contains about 32.8% cellulose, 36.8% hemicellulose and 16.8% lignin. These components cannot be easily separated without altering hemicellulose, which is closely bound to lignocellulosic components [6]. One of the prerequisites of an effective utilization of biomass such as cereal straws and grasses is the fractionation of the main components, polysaccharides and lignin, by a relatively mild procedure, ensuring their minimum physical and chemical changes as well as an acceptable extraction efficiency. [7]. So, for the full use of lignocellulosic biomass, one such possibility would be to extract the hemicellulose before cellulose to convert them to higher value-added products such as prebiotic xylooligosaccharides or polymers and molecules for chemical, pharmaceutical applications, natural barrier for packaging films, plastics and cellulosic pulp additives [8, 9].

Different treatments have been applied to hemicellulose extraction with addition of chemicals such as alkali, acid or hydrogen peroxide [10 - 14]. There are a wide variety of possible approaches for hemicelluloses extraction or pretreatment [15, 16] including pretreatments or isolation that span the complete range of pH and can use a wide range of temperatures [17, 20]. Alkali treatment at moderate temperatures is the basis of at least one approach for hemicelluloses extraction from wood prior to pulping [16, 21]. However, under alkaline-temperature conditions tested, the yield of extractable sugars from wood chips is usually low and significantly degraded xylan and very low concentrations of xylose have been obtained.

Alkali treatment of lignocellulosic substances such as cereal straw and bagasse disrupts the cell wall by dissolving hemicelluloses, lignin, and silica, by hydrolyzing uronic and acetic esters, and by swelling cellulose, decreasing the crystallinity of cellulose [22]. In addition, the treatment also cleaves the α-ether linkages between lignin and hemicelluloses and the ester bonds between lignin and/or hemicelluloses and hydroxycinnamic acids, such as *p*-coumaric and ferulic acids [23]. More importantly, the alkaline treatment has been proved to be a promising process to achieve complete utilization of lignocelluloses without impact to the environment [24].

The alkali extraction of hemicellulose succeeds in separating polymeric heteropolysaccharides from the fiber source [25]. By this process using alkaline and temperatures below 40 ºC, the wheat straw can be simply fractionated into alkali-soluble lignin and hemicelluloses and residue, which makes it easy to utilize them for more valuable products with minimal physical and chemical changes.

The end residue, mainly cellulose, can be used for either paper or cellulose derivatives. The lignin can be converted to valuable products, such as carbon fiber and adhesives From the solubilized hemicelluloses, food additives and other polymers can be produced. Recently, some important applications for hemicelluloses, such as xylans have been discovered. The current uses of xylans on an industrial scale involve their conversion to xylose, xylitol, and furfural. Xylitol is produced by hydrolysis of xylan, crystallization of xylose, and hydrogenation. This has been tested in a variety of food products [24]. In the paper industry, it has been demonstrated that the addition of hemicelluloses as additive in the cellulosic pulp can improve some mechanical properties of the paper, among other features of papermaking [8, 25].

The purpose of this work was examine and optimize the conditions for the cold alkaline extraction (soda concentration, time and temperature) of hemicellulose from wheat straw and its subsequent precipitation with ethanol.

2. Material and methods

2.1 Source and analysis of biomass

Wheat straw biomass was obtained on a plantation in Huelva (southwestern Spain).The raw material was prepared in accordance with TAPPI T-257 and analyzed for the following parameters: 1% NaOH solubles (TAPPI T 212 om-07), hot water solubles (TAPPI T 207 cm-93), ethanol–benzene extractives (TAPPI T 204 cm-07), holocellulose [26], α-cellulose (TAPPI T 203 cm-09) and ash (TAPPI T 211 om-07) contents. All treatments in this study were in a completely randomized design with four replications (variation coefficient less than 3% and less than 1% for holocellulose and cellulose contents).

In addition, the cellulose and hemicellulose composition of the material, in the form of glucan, xylan, araban and acetyl groups, were determined by high performance liquid chromatography (HPLC). Aliquots from the homogenized wood were subjected to moisture determination (drying at 105ºC to constant weight), quantitative acid hydrolysis with 5 mL of 72% sulfuric acid for an hour (TAPPI T248 sp-08), and quantitative posthydrolysis with 4% sulfuric acid at 121 ºC and 2 atm during 60 min in order to ensure quantitative conversion of oligomers into monomers [27]. Before HPLC analysis, the solid residue from posthydrolysis was recovered by filtration and considered as Klason lignin. The monosaccharides and acetic acid contained in hydrolysates were determined by HPLC in order to estimate (after corrections for stoichiometry and sugar decomposition) the contents in cellulose (as glucan), hemicelluloses (xylan + araban + acetyl groups). Chromatographic determination was performed using an Agilent 1100 HPLC equipped with an ion-exchange resin BioRad Aminex HPX-87H column under the following conditions: mobile phase, 0.005 mol·L-1 of sulphuric acid; flow rate, 0.6 mL·min-1; and column temperature, 50°C. The volume injected was 20 µL

All HPLC determinations were in a completely randomized design with three replications (variation coefficient less than 4%).

Gross heating values of raw materials were determined at a constant volume in accordance with ‘‘CEN/TS 14918:2005 (E) Solid biofuels-Method for the determination of calorific value’’ and UNE 164001 EX standards by using a Parr 6300 Automatic Isoperibol Calorimeter.

2.2. Cold Alkaline Extraction of Hemicellulose

The lignocellulosic material was ground and sieved in order to obtain a uniform chip size. The chips thus obtained were treated with variable NaOH concentrations at different temperatures for also different lengths of time. Treatments were conducted in 1 L beakers that were immersed in a thermostated bath and stirred at 5 min intervals. Once each treatment was finished, the resulting suspension was filtered and the solid washed with 2 L of water prior to neutralization with 2 N acetic acid and drying at room temperature. The liquid phase from the extraction was adjusted to pH 4.5–5.5 with 37% HCl, supplied with 4 volumes of 95% ethanol and centrifuged at 4500 rpm for 3 min. Then, the precipitate formed was washed with 95% ethanol and freeze-dried. Figure 1 depicts the operational scheme of the proposed procedure for cold alkaline extraction of hemicellulose from wheat straw. All tests were conducted in triplicate.

2.3. Experimental Design for Extraction Process, Chemical Determination

A 2^n central composite experimental design that enabled the construction of second-order polynomial in the independent variables and the identification of statistical significance in the variables was used. This allowed relating the dependent (yield, Klason lignin, glucan, xylan, araban and hemicellulose contents in solid residue, and Klason lignin, glucose and hemicellulose contents in extraction liquor) and independent (concentration of soda, temperature and time of process) variables of the extraction process with a minimum number of experiments.

Independent variables were normalized by using the following equation:

$$X_n = \frac{X - \overline{X}}{(X_{\max} - X_{\min})/2}$$

Where X is the absolute value of the independent variable concern, X is the average value of the variable, and Xmax and Xmin are its maximum and minimum values, respectively.

Based on previous studies operation conditions were selected**.** The alkali concentrations were 80, 100 and 120 $g \cdot l^{-1}$, extraction temperature and extraction time were 20, 30 and 40 ºC, and 30, 60 and 90minutes, respectively. The liquid/solid ratio was 15/1 in all experiments.

The number of tests required was calculated as $N = 2^n + 2\cdot n + nc$, 2^n being the number of points constituting the factor design, 2n that of axial points and nc that of central points. Under our conditions, N = 16.

The experimental results were fitted to the following second-order polynomial:

$$Y = a_o + \sum_{i=1}^{n} b_i X_{ni} + \sum_{i=1}^{n} c_i X_{ni}^2 + \sum_{i=1;\, j=1}^{n} d_i X_{ni} X_{nj} \quad (i < j)$$

The independent variables used in the equations relating to both types of variables were those having a statistical significant coefficient (those not exceeding a significance level of 0.05 in the student ´s-test – t < 2 - and having a 95% confidence interval excluding zero).

Aliquots from the homogenized material were subjected to moisture determination (drying at 105ºC to constant weight), quantitative acid hydrolysis with 5 mL of 72% sulfuric acid for an hour (TAPPI T248 sp-08), and quantitative posthydrolysis with 4% sulfuric acid at 121 ºC and 2 atm during 60 min in order to ensure quantitative conversion of oligomers into monomers [28]. Before HPLC analysis, the solid residue from posthydrolysis was recovered by filtration and considered as Klason lignin. The monosaccharides and acetic acid contained in hydrolysates were determined by HPLC in order to estimate (after corrections for stoichiometry and sugar decomposition) the contents in cellulose (as glucan), hemicelluloses (xylan + araban), and acetyl groups. Chromatographic determination was performed using an Agilent 1100 HPLC equipped with an ion-exchange resin BioRad Aminex HPX-87H column under the following conditions: mobile phase, 0.005 $mol\cdot L^{-1}$ of sulphuric acid; flow rate, 0.6 $mL\cdot min^{-1}$; and column temperature, 50°C.

3. Results and Discussion

3.1. Raw Material

Annual plants contain large amounts of hot water solubles and 1% NaOH water solubles. The wheat used here only departed from other plant materials in its 1% NaOH soluble contents, which were 19.8% higher than those previously reported by Jiménez et al. [29-31] and 8.0% higher than those reported by Pan et al. [32] (see Table 1).

This increased content in 1% NaOH solubles in the straw is consistent with its decreased holocellulose content (10% lower than that found by Pan et al. [32]) and increased hemicellulose content (the highest among those shown in Table 4 and 1–56% higher than the others). These contents differ between raw materials and also with plant variety, location, time of year and collection system, as well as with the particular analytical methods used. The α-cellulose content of our wheat straw fell in the lower end of the range of reported values and was 52.6% lower than that reported by Montané et al. [4].

The lignin content of the straw was 16.6%–57.2% lower than reported values obtained a considerably lower content. The content in soluble lignin of the straw was high in relation to wood materials.

The content in ethanol–benzene extractives was similar to reported values and also to the reference value for cereal straw (6%). As can be seen in Table 1, Pan et al. [32] reported unusually high ash content for rice straw (around 10%).

The heat power on a dry basis of our wheat straw, about 20 MJ/kg, was higher than the typical values for wood raw materials [e.g. 27.35 MJ/kg for pine wood [33], and 19.6–20.5 MJ/kg for bark, fir wood, softwood, common Oregon pine, pine woods and cedar wood [34-37]. In fact, it was closer to those for grass crops, agricultural residues or some hardwoods. Also, it was higher than that for barley straw (15.6–1 6.7 MJ/kg according to Satvanarayan et al.[36]), but lower than those for wheat straw reported by Brebu et al.[33] and Demirbas [34] ─which can be ascribed to a lower content in cellulose (the most “energetic” among the three main components of lignocellulosic materials [38].

We used High Performance Liquid Chromatography to determine major polysaccharide groups in the hemicellulose fraction. The material was found to have a high content (21.4%) in xylan, the main hemicellulosic component. The content in glucan, 33.1%, was quite consistent with that of α-cellulose as determined according to the corresponding TAPPI standard. On the other hand, the content in lignin, 18.9%, differed markedly from Klason lignin as determined in accordance with the applicable TAPPI standard ─it should be noted that the TAPPI method does not subject the raw material to two preliminary extractions with ethanol/benzene and hot water, which possibly causes the incorporation of greater amounts of soluble lignin than in the determination of lignin in Table 1. In any case, the lignin content found was within the range of previously reported values (see Table 1). The contents in araban and acetyl groups were 2.01 and 1.74%, respectively.

3.2. Cold Alkaline Extraction of Hemicellulose

Table 2 shows the results of yield (the solid phase is referred to initial raw material) and composition of solid phase after cold alkaline extraction of hemicelluloses. Also, the operational conditions in the proposed experimental were designed. Table 3 shows the characterization of liquid phase after cold alkaline extraction: lignin, glucose and hemicellulose contents (these results were calculated in terms of the composition of the raw material and the post-extraction solid phase. They were not determined experimentally owing to the extreme alkalinity of the post-extraction liquid phase. Total hemicellulose in the extract was determined as the combination of glucose, xylose, arabinose and acetyl groups in the liquid phase).

As expected, acetyl groups of hemicelluloses are totally neutralized and they are not detected in solid phase after cold alkaline extraction. Glucan content is little affected. Only a decrease between 1.1 % and 11.2 % of glucan content in raw material was observed. Hemicelluloses fraction is the most affected by cold alkaline extraction. The xylan and araban fractions, in raw material, decreases between 26.4 % to 59.6 % and 6.0 % to 38.8 % respectively. In general, the cold alkaline extraction rates are important, between 25.5 % and 38.8 % (100-yield, %) of the initial raw material. Such a high extraction yield in the liquid phase was largely due to hemicellulose (30.1–59.2%). These results are suggestive of highly selective extraction or separation of the hemicellulose fraction relative to glucan derivatives. The selectivity of the separation is probably even more selective since part of the glucose present in the liquid phase must have come from hemicellulose rather than from glucan in the raw material [39]. Also, it is important the cold alkaline extraction of lignin, between 40.4 % and 59.2 % of the lignin content in raw material.

The results of tables 2 and 3 were modeled by using the above-described multiple regression methodology. The ensuing models are shown in Table 4. The yield of the cold alkaline extraction (Y_{YS}, eq. 1) was especially influenced by the temperature and also, to a lesser extent, by the operation time and alkali concentration. The lignin content of the solid phase was also primarily influenced by the temperature. This is suggestive of thermal hydrolysis of the lignin polymer, which can hardly have been affected by the alkali concentration at this temperature level ─in fact, industrial delignification processes are conducted at much higher temperatures (150–180 °C). Although much less influential than X_T, the term X_cX_T in eq. 2 (Y_{LI}) is also significant and anticipates the effect of the alkali on the delignification efficiency at higher temperatures. This hypothesis is strongly influenced by the temperature; also, the prevalence of this effect on lignin additionally reflects in the relative values of the coefficients of equation 7 (Y_{LIEX}), where, again, the term X_T is that having the most significant coefficient.

It is not possible clear conclusions about the influence of independent variables on polysaccharide contents in solid phase after cold alkaline extraction (equations 3 to 5 in table 4, Y_{XI}, Y_{GL} and Y_{AR}). However, equations 6 (Y_{THM}) and 9 (Y_{THMEX}) shows significant influence of lineal terms and the interaction term between alkali concentration and the process temperature.

In order to determine the values of the independent variables in cold alkaline extraction (alkali concentration and operation temperature and time) giving the optimum values of total hemicelluloses in solid phase post-autohydrolysis, the response surfaces for each dependent variable were plotted (Figs. 2 to 3**).** Through the three-dimensional plot it is very easy and convenient to understand the interactions between two or three variables and to locate their optimum ranges. Since operation temperature is the most influential variable, the response surfaces are showed at outliers (Temperature: +1 and -1) of the proposed experimental design.

As can be seen from Fig. 2, the highest extraction yields for the liquid phase will be obtained at high temperatures ─decreased solid yields─ and long operation times. As regards alkali concentration, once the central levels in the operating range (100 g/L) are surpassed, the extraction yield ceases to increase. Therefore, such alkali levels should not be exceeded if the process is to be economical. These conditions, however, result in substantial extraction of lignin into the liquid phase.

Figure 3 shows the variation of the total amount of hemicellulose extracted in the liquid phase, which changed similarly to the overall extraction yield. The independent variables operation time and alkali concentration were considerably less influential than the temperature. Interestingly, the equations contained quadratic terms and interactions involving the alkali concentration; above a given, medium level of this variable, hemicellulose extraction failed to increase in response to an increase in the other variables. Again, this suggests the need to use medium alkaline concentrations (90–100 g/L) for economy. Identical conclusions can be drawn from the response surfaces for the variation of the extraction yield of xylan and araban derivatives in the liquid phase or the permanence of the xylan and araban fractions in the solid phase (results not shown).

The graphs in Fig. 3, which show the amount of hemicellulose extracted in relation to its content in the raw material, facilitated optimization of hemicellulose extraction. Three different values of the independent variables leading to the maximal hemicellulose extraction yield (about 45%) were used. The third, which resulted in substantial alkali savings, decreased the hemicellulose yield in the liquid phase by only 1.3% with respect to the maximum value obtained at point [0.6 (alkali concentration), +1 (temperature), +1 (time)] in the experimental design.

The optimum operating conditions (determined with JMP 8.0 statistical software) were used to conduct a cold alkali extraction process the solid and liquid phases from which were characterized following acid post-hydrolysis as described under Experimental. The strong alkalinity of the medium precluded direct determination of sugars and oligomers by HPLC. The analytical results are shown in Table 5, which includes the theoretical composition of the solid phase as determined with the models of Table 4. As can be seen, the calculations, which provided for intrinsic variability in the process and raw material, were highly consistent with the experimental results. In fact, the mass balance of araban and acetyl groups in both fractions (liquid phase + solid phase) was 100% consistent with the composition of the raw material. Thus, the glucan content in the combined fractions was 32.7% (versus 33.1% in the raw material) and the xylan content 19.2% versus 21.4%. It should be noted that part of the glucose quantified as a product of glucan degradation was in fact a product of hemicellulose [40,41] and that part of the xylose formed was dehydrated to furfural (by effect of the hydrolytic reaction used in the analyses) and other degradation products [42].

The total amount of hemicellulose extracted in the liquid phase was 56.1% of that initially present in the raw material ‒which contained 3.7% glucan. Therefore, the proposed cold alkaline extraction method affords selective extraction of the hemicellulose fraction, in addition to also highly efficient delignification ‒59.1% of all lignin present in the raw material was extracted.

The liquid phase from a cold alkaline extraction with 100 g/L alkali, a temperature of 40 °C and an operation time of 90 min was precipitated with ethanol as described under Experimental. A test performed in quadruplicate provided an average yield of 21.9% (0.56) in the solid phase; this contained 10.3% (0.53) lignin and 0.022% (0.0014) soluble lignin, which jointly accounted for 12.0% of all lignin initially present in the raw material. The total content in hemicellulose was 45.2% (0.45); therefore, the separation yield obtained by precipitating hemicellulose in an alcoholic medium was 70.2% and the cold alkaline extraction yield 39.4%.

4. Conclusions

Wheat straw, which contains more than 25% hemicellulose (xylan + araban + acetyl groups) and less than 19% lignin, provides an effective raw material for the selective separation of the hemicellulose fraction from the cellulose and lignin fractions by cold alkaline extraction.

The cold alkaline extraction of hemicelluloses from wheat straw develops optimally at a temperature of 40 °C, and operation time of 90 min and an alkali concentration of 100 g/L. These conditions allow 56.1% of all hemicellulose and 59.1% of all lignin initially present in the raw material to be extracted in the liquid phase.

Subsequent separation of hemicellulose in the liquid fraction from the cold alkaline extraction by precipitation with ethanol allows 39.4% of all hemicellulose present in the raw material to be recovered; also, the extracted–precipitated fraction contains only 10.3% lignin, which accounts for 12% of all present in the raw material.

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References

[1] F. Carvalheiro, L.C. Duarte, F.M. Gírio. Hemicellulose biorefineries: a review on biomass pretreatments. J. Sci. Ind. Res. 67 (2008) 849-864.

[2] H. Clark. Green chemistry for the second generation biorefinery - Sustainable chemical manufacturing based on biomass. J. Chem. Technol. Biotechnol. 82 (2007) 603-607.

[3] D. S. Ruzene, D.P. Silva, A. A. Vicente, A. R. Gonçalves, J. A. Teixeira. An Alternative Application to the Portuguese Agro-Industrial Residue: Wheat Straw. Appl. Biochem. Biotechnol. 147 (2008) 85–96.

[4] D. Montané, X. Farriol, J. Salvadó, P. Jollez, E. Chornet. Fractionation of wheat straw by steam explosion pretreatment and alkali delignification. Cellulose pulp and byproducts from hemicellulose and lignin. J. Wood Chem. Technol. 18 (1998) 171- 191.

[5] D. S. Ruzene, D. P. Silva, A. A. Vicente, A. R. Gonçalves, J. A. Teixeira. An Alternative Application to the Portuguese Agro-Industrial Residue: Wheat Straw. Appl. Biochem. Biotechnol. 147 (2008) 85–96.

[6] J. Lundqvist, A. Teleman, L. Junel, G. Zacchi, O. Dalman, F. Tjernel. Isolation and characterization of galactoglucomannan from spruce (*Picea abies*). Carbohydr. Polym. 48 (2002) 29–39.

[7] A.X. Jin, J.L. Ren, F. Peng, F. Xu, G.Y. Zhou, R.C. Sun, J.F. Kennedy. Comparative characterization of degraded and non-degradative hemicelluloses from barley straw and maize stems: Composition, structure, and thermal properties. Carbohydr. Polym. 78 (2009) 609–619.

[8] D.U. Lima, R.C. Oliveira, M.S. Buckeridge. Seed storage hemicelluloses as wet-end additives in papermaking. Carbohydr. Polym. 52(2003) 367-373.

[9] J. Hartman, A.N. Albertsson, M.S. Lindblad, J. Sjöberg. Oxygen barrier materials from renewable sources: material properties of softwood hemicelluloses bases films. J. Appl. Polym. Sci., 100 (2006) 2985-2991.

[10] J.X. Sun, X.F. Sun, R.C. Sun, Y.Q. Su. Fractional extraction and structural characterization of sugarcane bagasse hemicelluloses. Carbohydr. Polym. 56 (2004) 195-204.

[11] F.Xu, J.X. Sun, F.C. Liu, R.C. Sun. Comparative study of alkali and acid organic solvent-soluble hemicelluloses polysaccharides from sugar cane bagasse. Carbohydr. Res. 341 (2006) 253-261.

[12] C. Martín, H.B. Klinke, A.B. Thomsen. Wet oxidation as a pretreatment method for enhancing the Enzyme & Microbial Technolgy enzymatic convertibility of sugarcane bagasse. Enzyme Microb. Technol. 40 (2007) 426-432.

[13] Y.J.Lee, C.H. Chung, F. Donal. Sugarcane bagasse oxidation using a combination of hypochlorite and peroxide. Bioresour. Technol. 100 (2008) 275-279.

[14] M. Brienzo, A.F. Siqueira, A.M.F. Mialgres. Search for optimum conditions of sugarcane bagasse hemicelllulose extraction. Biochem. Eng. J. 46 (2009) 199-204.

[15] W. W. Al-Dajani, U. W. Tschirner. Pre-extraction of hemicelluloses and subsequent kraft pulping Part 1: alkaline extraction. TAPPI J. 7 (2008) 3-8.

[16] G. Schild, H. Sixta, L. Testova. Mutifuncional alkakine pulping, delignification and hemicellulose extraction. Cell Chem. Technol. 44 (2010) 35-45.

[17] M. Ibrahim, W.G. Glasser. Steam-assisted biomass fractionation. Part III: a quantitative evaluation of the "clean fractionation" concept. Bioresour. Technol. 70(1999) 181-192.

[18] F. Peng, J.L. Ren, F. Xu, J. Bian, P. Peng, R.C. Sun. Comparative Study of Hemicelluloses Obtained by Graded Ethanol Precipitation from Sugarcane Bagasse. J. Agric. Food Chem. 57 (2009) 6305–6317.

[19] H.J. Huang, S. Ramaswamy, W.W. Al-Dajani, U. Tschirner.. Process modeling and analysis of pulp mill-based integrated biorefinery with hemicellulose pre-extraction for ethanol production: A comparative study. Bioresour. Technol. 1012(2010) 624-631.

[20] J. Bian, F. Peng, X.P. Peng, F. Xu, R.C. Sun, J.F. John F. Kennedy, J.F. Isolation of hemicelluloses from sugarcane bagasse at different temperatures: Structure and properties. Carbohydr. Polym, 88 (2012) 638–645.

[21] C. Heli, Z. Huaiyu, F. Shiyu, A.L. Lucian. Alkali extraction of hemicellulose form depithed corn stover and effects on soda-aq pulping. Bioresources, 11 (2010) 196-206.

[22] M.G. Jackson. The alkali treatment of straws. Anim. Feed Sci. Technol. 2 (1977) 105–130.

[23] R.R. Spencer, D.E. Akin. Rumen microbial-degradation of potassium hydroxide-treated coastal bermudagrass leaf blades examined by electron-microscopy. J. Anim. Sci., 51 (1980) 1189–1196.

[24] F. Peng, J.-L. Ren, F. Xu, J. Bian, P. Peng, R.-C.Sun.. Comparative Study of Hemicelluloses Obtained by Gradedv Ethanol Precipitation from Sugarcane Bagasse. J. Agric. Food Chem. 57 (2009) 6305–6317.

[25] W.G. Glasser, W.E. Kaar, R.K. Jain, J.E. Sealey. Isolation Options For Non-Cellulosic Heteropolysaccharides (Hetps). Cellulose 7 (2000) 299–317.

[25] B. Saake, T. Busse, J. Puls. The effect of xylan adsorption on the properties of sulfite and kraft pulps. Appita J. 2(2005) 141–146.

[26] L.E. Wise, M. Marphy, A. D‘Adieco.. Chlorite holocellulose, its fractionation and bearing on summative wood analysis and on studies on the hemicelluloses. Trade Journal. 122 (1946) 35-43.

[27] G. Garrote, M.E. Eugenio, M.J. Díaz, J. Ariza, F. López.. “Hydrothermal, pulp processing of eucalyptus wood,” Bioresour. Technol. 88(2003) 61-68.

[28] J.C. García, M.A.M. Zamudio, A. Pérez, F. López, J.L. Colodette.. Search for optimum conditions of Paulownia autohydrolysis process and influence in pulping process. Environ. Prog. Sustain. Energy. 30 (2011) 92-101.

[29] L. Jiménez F. López, C. Martínez. Biological Pretreatments for Bleaching Wheat-Straw Pulp.Process Biochem. 29 (1994) 595-599.

[30] L. Jiménez, A. Rodríguez, J.L. Ferrer, A. Pérez, V. Angulo. Paulownia, a fast-growing plant, as a raw material for paper manufacturing. Afinidad 62 (2005) 100-105.

[31] L. Jiménez, A. Pérez, A. Rodríguez, M.J. De la Torre..New raw materials and pulping processes for production of pulp and paper. Afinidad 63 (2006) 362-369.

[32] X. Pan, C. Arato, N. Gilkes, D. Gregg, W. Mabee, K. Pye, Z. Xiao, X. Zhang, J. Saddler. Biorefining of Softwoods Using Ethanol Organosolv Pulping: Preliminary Evaluation of Process Streams for Manufacture of Fuel- Grade Ethanol and Co- Products. Biotechnol. Bioeng. 90 (2005) 473- 481.

[33] M. Brebu, S. Ucar, C. Vasile, J. Yanik. Co-pyrolysis of pine cone with synthetic polymers Fuel 89 (2010) 1911-1918.

[34] A. Demirbas. Products from lignocellulosic materials via degradation processes. Energy Sources, Part A: Recovery, Utilization and Environmental Effects 30 (2008) 27-37.

[35] C. Telmo, J. Lousada, N. Moreira. Proximate analysis, backwards stepwise regression between gross calorific value, ultimate and chemical analysis of wood. Bioresour. Technol. 101 (2010) 3808-3815.

[36] N. Satyanarayan, V.G. Vaibhav, K.R. Prasant, J. Kathlene, K.D. Ajay. Characterization of Canadian biomass for alternative renewable biofuel. Renew. Energy 35 (2010) 1624–1631.

[37] M.J. Feria, A. Rivera, F. Ruiz, E. Grandal, J. C. García, A. Pérez, F. López.. Energetic Characterization Of Lignocellulosic Biomass From South-West Spanish. Int. J. Green Energy 8 (2011) 631-642.

[38] M. Erol, H. Haykiri-Acma, S. Küçü kbayrak, S. Calorific value estimation of biomass from their proximate analyses data. Renew. Energy 35 (2010) 170–173.

[39] R.C. Sun, J.M. Lawther, W.B. Banks. Influence of alkaline pre-treatments on the cell wall components of wheat straw. Ind. Crop. Prod. 4 (1995) 127–145.

[40] R.C. Sun, J.M. Lawther, W.B. Banks. Fractional and structural characterization of wheat straw hemicelluloses. Carbohydr. Polym. 29 (1996) 325–331.

[41] E. Escarnot, M. Aguedo, M. Paquot. Characterization of hemicellulosic fractions from spelt hull extracted by different methods. Carbohydr. Polym. 85 (2011) 419–428.

[42] S. Caparros, G. Garrote, J. Ariza, M.J. Diaz, F. Lopez. Xylooligosaccharides Production from Arundo donax. J. Agric. Food Chem.55 (2007) 5536 - 5543.

Figure 1

Figure 1: Scheme for extraction of hemicelluloses from wheat straw.

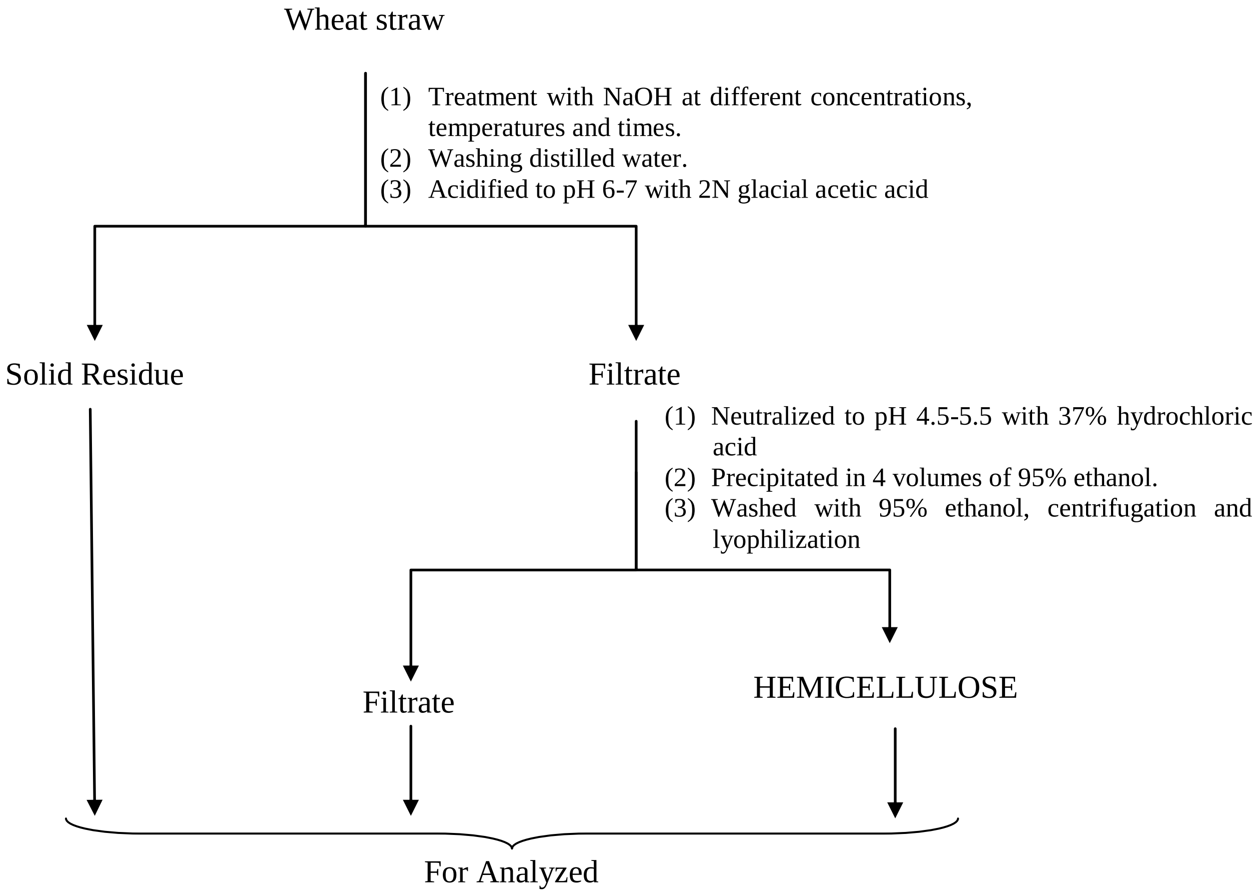

Figure 2: Solid residue yield variation as a function of independent variables of extraction process.

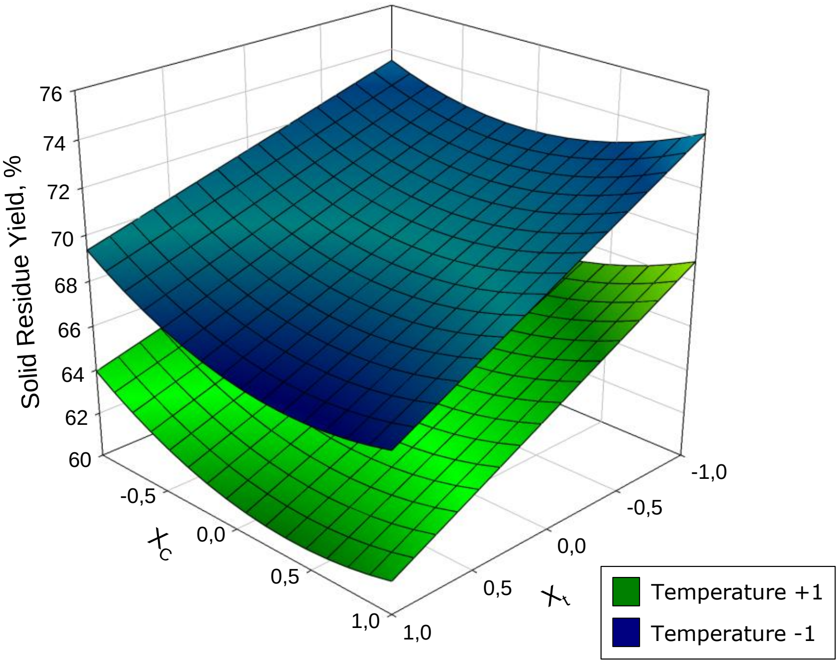

Figure 3

Figure 3: Liquor Extraction Hemicellulose variation as a function of independent variables of extraction process.

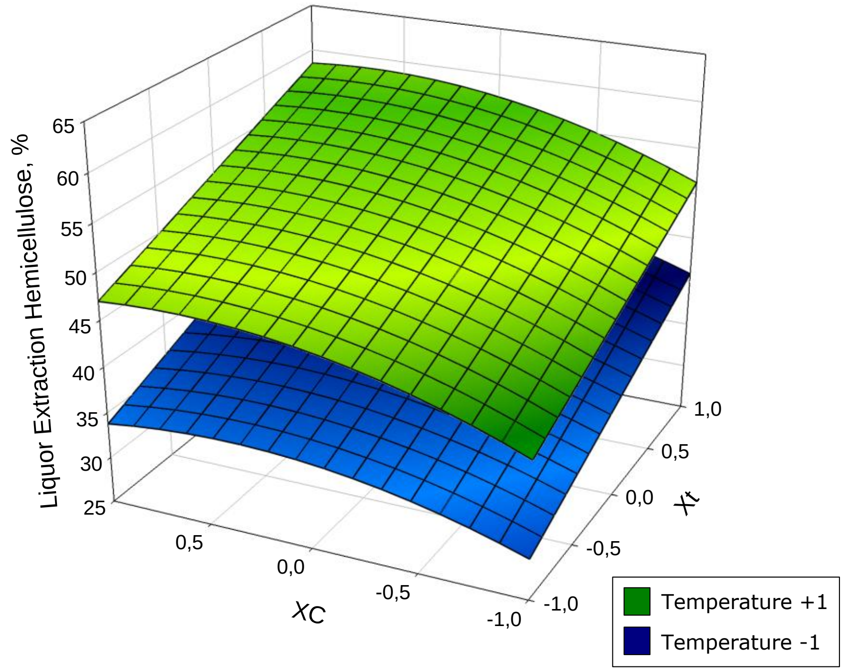

Table 1 Shows the results of chemical and energetic characterization of wheat straw and results from other authors

Results in percentages, to respect on raw material (over dry basis)	Results in this work	Pan et al 2005	Jimenez et al.1994, 2005, 2006	Montané et al., 1998
Hot water solubles	15.0	17,5	12,27-12,49	-
1% NaOH solubles	49.7	46	41,49-43,58	-
Etanol-benzene extractives	5.24	5,3	4,01	-
Ash	5.9	9,6	6,49	-
Holocellulose	69.6	76,6	76,2	-
α-Cellulose	32.8	-	39,72	32,3-50
Hemicellulosa	36.8	-	36,48	23,6-35,6
Klason lignin	14.2	16,5	17,28	22,8
Soluble lignina	2.8	2,5	-	-

Table 2

Table 2. Normalized operational conditions and composition of solid phase after cold alkaline extraction of hemicelluloses. The concentrations are referred to initial raw material.

Normalized values of alkali concentrtion (X_C) temperature (X_T) and operation time (X_t)			Yield (%)	Klason Lignin (%)	Glucan (%)	Xylan (%)	Araban (%)	Total Hemicelluloses (%)
0	0	0	65,2	9,8	31,3	12,2	1,5	13,7
0	0	0	65,9	9,7	31,2	12,1	1,6	13,7
1	1	1	61,2	8,2	31,1	8,6	1,6	10,3
1	1	-1	66,9	10,2	30,7	12,1	1,8	13,9
1	-1	1	68,5	9,3	31,8	12,1	1,5	13,6
1	-1	-1	74,5	11,2	30,5	14,5	1,9	16,4
-1	1	1	64,3	7,7	32,3	10,7	1,8	12,4
-1	1	-1	69,3	10,3	31,6	13,1	1,7	14,8
-1	-1	1	68,6	9,0	32,7	13,4	1,6	15,0
-1	-1	-1	73,3	11,2	31,9	15,8	1,8	17,6
1	0	0	67,1	9,7	31,7	11,9	1,6	13,5
-1	0	0	67,1	9,7	32,8	13,3	1,8	15,1
0	1	0	62,8	9,3	30,7	10,3	1,5	11,8
0	-1	0	68,5	10,1	31,1	13,6	1,5	15,1
0	0	1	63,7	8,5	31,3	10,7	1,2	12,0
0	0	-1	69,4	10,8	29,4	13,5	1,8	15,3

X_c X_t X_T Yield (%) Lignin (%) Glucan (%/%), Xylan (%/%), Araban (%/%), Total hemicelluloses (%/% refered to xylan + araban + acetyl groups in raw material) X_c: Alkali concentration, X_t: Operation time; X_T: Operation temperature

Table 3. Normalized operational conditions and composition of liquid phase after cold alkaline extraction of hemicelluloses. The concentrations are referred to each polymer content (glucan, xylan, araban) in initial raw material.

Normalized values of alkali concentrtion (X_C) temperature (X_T) and operation time (X_t)			Klason Lignin (%)	Glucan (%)	Total Hemicelluloses (%)
0	0	0	47,9	5,3	45,6
0	0	0	48,7	5,6	45,6
1	1	1	56,7	6,1	59,2
1	1	-1	46,1	7,3	44,8
1	-1	1	50,9	4,0	45,9
1	-1	-1	40,4	7,8	34,6
-1	1	1	59,2	2,3	50,5
-1	1	-1	45,1	4,6	40,9
-1	-1	1	52,0	1,1	40,4
-1	-1	-1	40,4	3,7	30,1
1	0	0	48,5	4,3	46,2
-1	0	0	48,5	1,0	39,7
0	1	0	50,6	7,1	53,0
0	-1	0	46,4	6,1	40,0
0	0	1	54,8	5,3	52,3
0	0	-1	42,5	11,2	39,0

X_c X_t X_T Yield (%) Lignin (%) Glucan (%/%),Total hemicelluloses (%/% refered to xylan + araban + acetyl groups in raw material). X_c: Alkali concentration, X_t: Operation time; X_T: Operation temperature

Table 4

Table 4. Equations yielded for each dependent variable for solid and liquid phases after cold alkaline extraction of wheat straw.

Equation		R^2	F- Snedecor
Residual Solid			
[1]	$Y_{YI}= 65{,}58 - 0{,}44\ X_C -2{,}89\ X_t -2{,}70\ X_T + 1{,}64X_C^2 + 1{,}06\ X_T^2 - 0{,}82\ X_C\cdot X_t$	0,992	192,00
[2]	$Y_{LI} = 9{,}67 - 0{,}52\ Xt - 1{,}12\ X_T + 0{,}11\ Xc\cdot X_T$	0,989	371,93
[3]	$Y_{XYS} = 12{,}07 - 0{,}75\ X_C - 1{,}39\ X_t - 1{,}29\ X_T + 0{,}42\ X_C^2$	0,980	138,41
[4]	$Y_{GLS} = 31{,}08 - 0{,}56\ X_C + 0{,}52\ X_T + 1{,}16\ X_C^2 - 0{,}69\ X_T^2$	0,901	24,99
[5]	$Y_{ARS} = 1{,}50 - 0{,}07\ X_C - 0{,}18\ X_T + 0{,}16\ X_C^2 - 0{,}10\ X_C\cdot X_T$	0,839	13,65
[6]	$Y_{HEM} = 13{,}58 - 0{,}738\ X_C - 1{,}448\ X_t -1{,}48\ X_T + 0{,}68\ X_C^2 - 0{,}18\ X_C\cdot X_T$	0,990	207,75
Liquid phase			
[7]	$Y_{LEX} = 48{,}47 + 2{,}49\ X_t + 5{,}86\ X_T + 0{,}25\ X_C^2$	0,995	814,71
[8]	$Y_{GLEX} = 6{,}07 + 1{,}68\ X_C - 1{,}48\ X_T + 0{,}48X_t - 3{,}53\ X_C^2 + 2{,}08\ X_T^2$	0,924	24,34
[9]	$Y_{HEX} = 45{,}92 + 2{,}91\ X_C + 5{,}74\ X_t + 5{,}88\ X_T - 2{,}70\ X_C^2 + 0{,}73\ X_C\cdot X_T$	0,990	207,75

Y_{YS}, Y_{LI}, Y_{XY}, Y_{GL}, Y_{AR}, Y_{THM} denote yield, lignin, xylan, glucan, araban and total hemicelluloses contents in solid phase after cold alkaline extraction respect initial raw material (dry basis).

X_C, X_t and X_T denotes normalized alkaline concentration, operation time and operation temperature respectively.

Y_{LIEX}, Y_{GLEX}, Y_{THMEX} denote lignin, glucan and total hemicelluloses extraction respect initial lignin, glucan and totalhemicellulose content in raw material (dry basis).

The differences between the experimental values and those estimated by using the previous equations never exceeded 10 % of the former.

Table 5. Chemical characterization of solid and liquid phases after a cold alkaline extraction process (operation temperature: 40 ºC, operation time: 90 min, alkali concentration: 100 g/L). * 0 +1 +1 point in de experimental design. ** Each value is a mean of four replicates. Variation coefficient less than 5 %.

Percentages refered to 100 g of raw material	Calculated composition of solid phase (models in table 4)*	Experimental composition of solid phase *	Calculated extraction of polymers in liquid phase (models in table 4)*	Extraction of polymers in liquid phase (refered to raw material / total hemicelluloses in raw material) *
Yield	61.1	64.0		
Lignin	8.0	7.7	56.8	
Glucan	30.9	31.4		1.2
Xylan	9.4	8.0		11.2
Araban	1.3	1.0		1.1
Acetyl Groups		0.0		1.8
Total hemicelluloses			57.5%	14.1/56.1 (xylan + araban + acetyl groups)