

# Assessing the Lignin Fraction Extracted from Brazilian Energy Crops

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**Abstract:** This study quantifies the extraction performance and some properties of the lignin obtained from six crops with potential for generating thermal energy in Brazil. A full factorial design ( $2^2$ ) with four replicates including two extraction methods and two particles size distributions was adopted for assessing the fraction of lignin, as well as its Higher Heating Value (HHV) and composition given by the proximate and ultimate analysis. It was verified that in all cases the HHV of the lignin is greater than that obtained for the respective raw material, suggesting that these lignins could be also used as fuel in thermochemical processes. The ANOVA analysis showed that at 95% of confidence level the lignin yield is influenced by the extraction methods, which employ different pH values during the hydrolysis treatment. In addition, it was found that, in most cases, a smaller biomass particle size contributed to more effective lignin extraction. Response surfaces were also included in order to show tendencies of the lignin yield obtained for each raw material in function of the pH value used by the extraction method and the biomass particle size range.

**Keywords:** Biomass, Renewable Energy, Lignin Extraction Methods

## Introduction

It is well known that biologically derived fuels may replace part of the traditional fossil fuels consumption, such as oil or coal. Applying agricultural techniques and appropriate strategies for processing, biofuels may offer at least 50% of savings in atmospheric emissions, when compared to the petroleum derivatives. In addition, biofuels produced from crops are considered renewable sources of thermal energy (Serna *et al.*, 2011).

In last year's, the production of green fuels, such as ethanol and others produced from biomass has gained prominence in several countries. If the ethanol industry focused on the second generation in these nations grows, enormous amounts of lignins will be discharged as phenol wastes (Kim *et al.*, 2009). So, lignin is expected to play an important role in the near future, being utilized to produce energy and also by-products (Sena-Martins *et al.*, 2008; Hage *et al.*, 2009; Matsushita *et al.*, 2013).

Currently, lignin is of specialist's interest among the various areas of science and also to benefit the industry,

when searching it for new practical applications (Hatfield and Fukushima, 2005; Mansouri and Salvadó, 2007; Wang *et al.*, 2009). However, due to its structural complexity and difficulty of isolation, lignin still waits for more participation in thermochemical processes, such as gasification and pyrolysis (Wang *et al.*, 2009; Hashimoto *et al.*, 2011).

Several works brings contributions about yields and functional groups of lignins, by using several methods such as: Pyrolysis, spectrometry, high performance liquid chromatography, fractionations and separations, among other analytical techniques, which have been previously studied by Lalvani *et al.* (2000; Celeghini and Lanças, 2001; Demirbas, 2003; 2005; 2009; Yanhua *et al.*, 2004; Huang *et al.*, 2010; Bikovens *et al.*, 2010). Indeed, the available information about the effect of parameters or conditions during the lignin extraction from lignocellulosic materials is scattered and scarce. Some studies have been carried out for straws, husks and leafs of herbaceous crops, but most of reviews are focused on characterization of wood lignins

(Buranov and Mazza, 2008; Domínguez *et al.*, 2008; Shen *et al.*, 2009; Beis *et al.*, 2010).

In this study the performance of the lignin extraction for six Brazilian crops is in this study the performance of the lignin extraction for six Brazilian crops is thermal conversion processes. Two procedures were tested for lignin extraction using different pH values, the Klason and Willstatter methods. An experimental factorial planning ( $2^2$ ) with four replicates was utilized in order to know the relevance of the planning ( $2^2$ ) with four replicates was utilized in order to know the relevance of the yield, as well as its behavior under different experimental conditions.

## Materials and Methods

The raw material was obtained in farms and food processing industries of Santa Catarina and Paraná states at South of Brazil. Analyses were performed at laboratories of the Santa Catarina State University (UDESC) and the Federal University of Technology-Paraná (UTFPR)/Ponta Grossa. The six assessed biomass were: Wheat straw (*Triticum aestivium*), corn straw (*Zea mays*), rice husk (*Oryza sativa*), sugarcane bagasse (*Saccharum officinarum*), wood chips (*Cedrelinga catenaeformis*) and elephant grass (*Pennisetum purpureum Schumach.*).

The preparation of samples was performed based on the guidelines of the TAPPI (2007; 2012) and the ASABE (2011). For raw materials were determined the lignin fractions, extractives and holocellulose, the higher heating values and also the proximate and ultimate analysis. The HHV and the proximate and ultimate analysis were also carried out for the lignin fraction obtained from each raw material.

### Feedstock Properties

The biomass was previously crushed utilizing a knife mill (SOLAB) and then classified in size by using standard sieves of the Tyler mesh series. The Table 1 shows the particles size distributions, including the mean Sauter diameter ( $dm$ ) value obtained for each biomass.

For test, only two particle size groups were considered. The first group was that collected in sieves with openings from 105 to 500  $\mu\text{m}$  (150 and 32 Tyler mesh) and second one, in sieves with openings from 1000 to 2000  $\mu\text{m}$  (16 and 09 Tyler mesh). The test procedure was based on the ABNT (1982).

In the proximate analysis, the moisture content was determined by utilizing an oven, which was maintained at 110°C during 6 h until no further change in weight occurred. In the volatiles determination, individual samples were placed inside porcelain crucibles with a lid and heated at 850°C during 60 min using a vertical muffle furnace (QUIMIS), with capacity to maintain a temperature of 1100°C. The ash content was determined by using procedure based on the ASTM (2004; 2007). During the test, the samples were kept

inside a muffle furnace and heated at 750°C during 1 hour until completely carbonized, the fixed carbon was determined by weight difference.

The ultimate analysis was performed by using an automatic chemical analyzer (TRUSPEC-LECO, CHONS). The resolution of the device for the basic elements is: Carbon: 0.3 p.p.m (parts per million) or 0.5% R.S.D (Relative Standard Deviation), Hydrogen: 100 p.p.m or 1.0% R.S.D, Nitrogen/Oxygen: 40 p.p.m or 0.5% R.S.D and sulfur 5 p.p.m or 1% R.S.D. In tests, 10 mg of individual samples were evaluated.

### Determination of Extractives and Holocellulose

In the extractives determination, two extractions occurred. First, the samples were exposed to a water bath, which was maintained at 100 $\pm$ 3°C during 1 h with constant stirring, thereby removing the fractions soluble in water. Further, a second solvent extraction was performed, utilizing 10 g of individual samples and a Soxhlet extraction apparatus (QUIMILAB). The solution was composed of ethanol/cyclohexane and distilled water (1:2 v/v). After the extractions, the samples were dried utilizing an oven, which was maintained at 105 $\pm$ 3°C. The differences in weight after and before the extractions determined the total amount of extractives and the final results were established by considering the total amount of extractives (fractions soluble in water plus fractions soluble in ethanol/cyclohexane). Test procedure was based on the NREL (2008).

The determination of holocellulose was based on TAPPI (2009; ASTM (1985). In tests, 5 g of samples already free from extractives were added into a solution composed of nitric acid (HNO<sub>3</sub>), ethanol (1:4 v/v) and distilled water. During the test, the samples were kept under constant reflux stirring during 1 h. After this, samples were washed, filtered and dried by using a laboratorial oven, which was maintained at 105°C and then weighted utilizing porcelain crucibles with previous weight tare.

### Extracting Lignin

The samples were prepared according the guidelines NREL (2011; TAPPI 2002). Two hydrolysis treatments were used: Klason (H<sub>2</sub>SO<sub>4</sub>) and Willstatter (HCl). During tests, 3 mL of an acid (72%) was separately diluted and the final concentration of the solution was reduced to 4% by adding to it 84 mL of deionized water. The samples were kept in a water bath at 30°C during 60 min with constant stirring, thereby performing uniform hydrolysis. The materials were washed and transferred to flasks and at the end of the reaction solids (lignin dark crystals) were obtained using filter papers (WHATMAN-41). The lignins were discharged with final pH varying between 2 (Klason) and 4 (Willstatter). Samples were washed and dried using an oven, maintained at 105 $\pm$ 3°C during 4-6 h and then weighed.

Table 1. Mass fraction distribution and particle mean diameter of the biomass samples

Sieve openings (mm)	$d_i$ (mm)	Mass fraction, $x_i$ (%)					
		Sugarcane bagasse	Corn straw	Wood chips	Rice husk	Elephant-grass	Wheat straw
2.000-4.000	3.0000	0.503	0.411	0.402	0.441	0.433	0.453
1.000-2.000	1.5000	0.312	0.397	0.395	0.279	0.381	0.396
0.500-1.000	0.7500	0.125	0.162	0.119	0.178	0.131	0.112
0.105-0.500	0.3025	0.060	0.030	0.084	0.102	0.055	0.039
$\Sigma x_i/d_i$		0.7400	0.617	0.832	0.907	0.753	0.692
$d_m$ (mm)		1.3500	1.620	1.200	1.100	1.330	1.450

$x_i$  = mass fraction with diameter  $d_i$

Table 2. Combinations for experimental tests

Run	Factor		Response variable, $y_i$ Lignin yield
	A Extraction method	B Particle size range ( $\mu\text{m}$ )	
1	Klason (+1)	1,000-2,000 (+1)	$y_1$
2	Klason (+1)	105-500 (-1)	$y_2$
3	Willstatter (-1)	1,000-2,000 (+1)	$y_3$
4	Willstatter (-1)	105-500 (-1)	$y_4$

### Higher Heating Values (HHV)

The higher heating values of samples of lignin (0.2-0.5 g) were measured utilizing a bomb calorimeter. Samples were burned in pure oxygen atmosphere following procedures of ABNT (1984; ASTM (2000).

### Experimental Planning for Lignin Extraction

The experiments of lignin extraction were planned according a statistical factorial design (2<sup>2</sup>). Factorial designs are useful to evaluate the effect estimative of several variables simultaneously, even when using small number of tests. Furthermore, this technique allows finding mathematical correlations between factors and responses. The ANOVA analysis and the response surfaces were generated using the program STATISTICA™ v.10. The factorial planning (Table 2) considered four replicates for each treatment or combination of controlled factors, totalizing 16 experiments.

## Results and Discussion

The major levels of extractives were obtained utilizing the wheat and corn straws, as well as elephant grass. Regarding the holocellulose content, the major levels were obtained utilizing the wheat, corn straws and rice husks. Table 3 shows the extractives and holocellulose fractions obtained from the each biomass.

Lee *et al.* (2007; Demirbas, 2009; Guo *et al.*, 2010; Haykiri-Acma *et al.*, 2010; Vargas-Radillo *et al.*, 2011; Rabemanolontsoa and Saka, 2012) found similar levels of extractives and holocellulose for several raw

materials. Table 4 shows the results of the proximate analysis for each biomass and its lignin.

The proximate analysis disclosed that the lignins extracted from wood chips and sugarcane bagasse have the highest levels of volatiles and as expected, that of the rice husk presented superior ash fraction. In addition, the lignins obtained from the elephant grass, corn and wheat straws evidenced the major levels of fixed carbon and in all cases, the fixed carbon values in the lignin were higher than those observed for the respective raw material.

Table 5 shows the results of the ultimate analysis for the biomass and lignin samples. The chemical composition of the biomass reported in this study was very similar to those previously searched by Vassilev *et al.* (2010; Rabemanolontsoa and Saka, 2013). It should be taken into account that differences on the chemical constitution of the materials studied here is conditional to several factors such as: Soil composition, climate, harvest, season, diseases, weeds presence, planting method, spacing, among others; thereby indicating differences even in plants of the same species.

Results of the major added levels of carbon and hydrogen, which influence on the thermal energy potential, were obtained from wood chips. Also, lignins evidenced major levels of carbon than the raw material, as expected. The ultimate analysis discloses that generally, the carbon content of lignins ranges between 50-60% and from biomass ranges between 40-50%. However, the biomass evidenced major quantities of oxygen 35-45% and hydrogen 4-7% than lignins, which presented results ranging from 30-40 and 4-6%, respectively.

Regarding the possibility of formation of harmful compounds, the ultimate analysis disclosed that remaining fractions of sulfur (from Klason method) and chlorine (from Willstatter method) in lignins could be released when used in thermochemical conversion

processes. So, studies focused in quantifying these emissions become essential for the appropriated use of lignin as a fuel during processes of combustion, gasification or pyrolysis. The HHV obtained from both biomass and lignin is showed in Table 6.

Table 3. Extractives and holocelluloses yields

	Sugarcane bagasse	Corn straw	Wood chips	Rice husks	Elephant-grass	Wheat straw
Extractives* (%)	13.190	13.380	9.7100	10.050	13.670	14.170
Holocelluloses* (%)	63.662	66.187	62.862	65.987	61.862	65.712

\*dry weight basis

Table 4. Proximate analysis of raw materials and lignins (dry basis)

Sample, method and analysis	Sugarcane bagasse	Corn straw	Wood chips	Rice husks	Elephant-grass	Wheat straw
<b>Biomass</b>						
Moisture (%)	12.90	12.64	13.55	10.91	11.18	11.90
Volatiles (%)	72.33	82.77	85.61	81.63	83.25	83.90
Ash (%)	10.80	1.58	0.18	10.32	10.70	9.30
Fixed carbon (%)	16.87	15.72	14.22	8.11	6.10	6.80
<b>Klason lignin</b>						
Volatiles (%)	72.81	70.18	73.46	70.02	68.44	71.28
Ash (%)	1.00	1.80	0.50	4.01	1.05	1.73
Fixed carbon (%)	26.19	28.02	26.04	25.97	30.51	26.99
<b>Willstatter lignin</b>						
Volatiles (%)	72.08	69.81	74.11	71.52	67.30	68.25
Ash (%)	0.79	1.54	0.04	3.84	1.51	1.69
Fixed carbon (%)	27.16	28.75	25.85	24.64	31.19	30.06

Table 5. Ultimate analysis of raw materials and lignins (dry basis)

Sample, method and analysis	Sugarcane bagasse	Corn straw	Wood chips	Rice husks	Elephant-grass	Wheat straw
<b>Biomass (%)</b>						
C	46.80	45.40	48.06	40.10	46.70	47.50
H	5.90	6.70	6.50	4.70	5.70	5.40
O	45.70	45.12	44.26	38.30	46.50	35.10
N	0.50	0.90	0.80	0.40	0.20	0.60
S	0.30	0.30	0.20	0.20	0.20	0.10
<b>Klason lignin (%)</b>						
C	60.34	55.04	60.21	56.40	52.74	57.42
H	4.10	5.58	5.28	4.82	5.14	5.10
O	34.10	37.64	32.30	37.44	40.60	35.90
N	0.40	0.50	0.30	0.30	0.10	0.40
S	1.06	1.24	1.91	1.04	1.42	1.18
<b>Willstatter lignin (%)</b>						
C	59.32	55.70	62.21	57.02	53.66	58.01
H	4.02	5.45	4.50	4.39	4.62	6.19
O	36.16	38.15	32.89	38.09	41.42	35.40
N	0.30	0.40	0.10	0.30	0.10	0.30
S	0.20	0.30	0.30	0.20	0.30	0.10

Table 6. Higher Heating Values (HHV) of raw materials and lignins, MJ/kg

Determination	Sugarcane bagasse	Corn straw	Wood chips	Rice husks	Elephant-grass	Wheat straw
Biomass	18.07	17.63	19.78	15.84	17.50	17.99
Klason lignin	24.64	21.48	25.73	22.16	23.17	20.61
Willstatter	22.34	20.60	24.83	21.53	21.92	20.11

Higher heating values for biomass close to those reported in this study were presented by Yan *et al.* (2009; De Ramos e Paula *et al.*, 2011). Results indicate that the all raw materials have lower HHV values than their lignin fractions, as expected. So, by using appropriated environmental emission control devices, lignin fractions can be important as source of thermal energy. It was noticed that the high carbon and hydrogen levels verified in the ultimate analysis for wood chips and its lignin were in accordance with the most elevated HHV values attained in both materials.

Table 7 and Fig. 1 show the results of the lignin yield and the effect estimate associated to the two controlled factors for each biomass, respectively.

Figure 1 suggests that for all biomass and conditions analyzed in this study the effect produced by the extraction method is more important that by the particle size. It is also noticed that in most of cases, the group of particle size ranged from 105 to 500  $\mu\text{m}$  tends to be more efficient for lignin extraction, once that the effect estimate in such cases was a negative value. This can be explained due to the more exposed superficial area when small particles of biomass are used during the lignin extraction.

In order to know the statistical significance of the extraction method and the biomass particle size on the lignin yield, a ANOVA analysis is presented in Table 8.

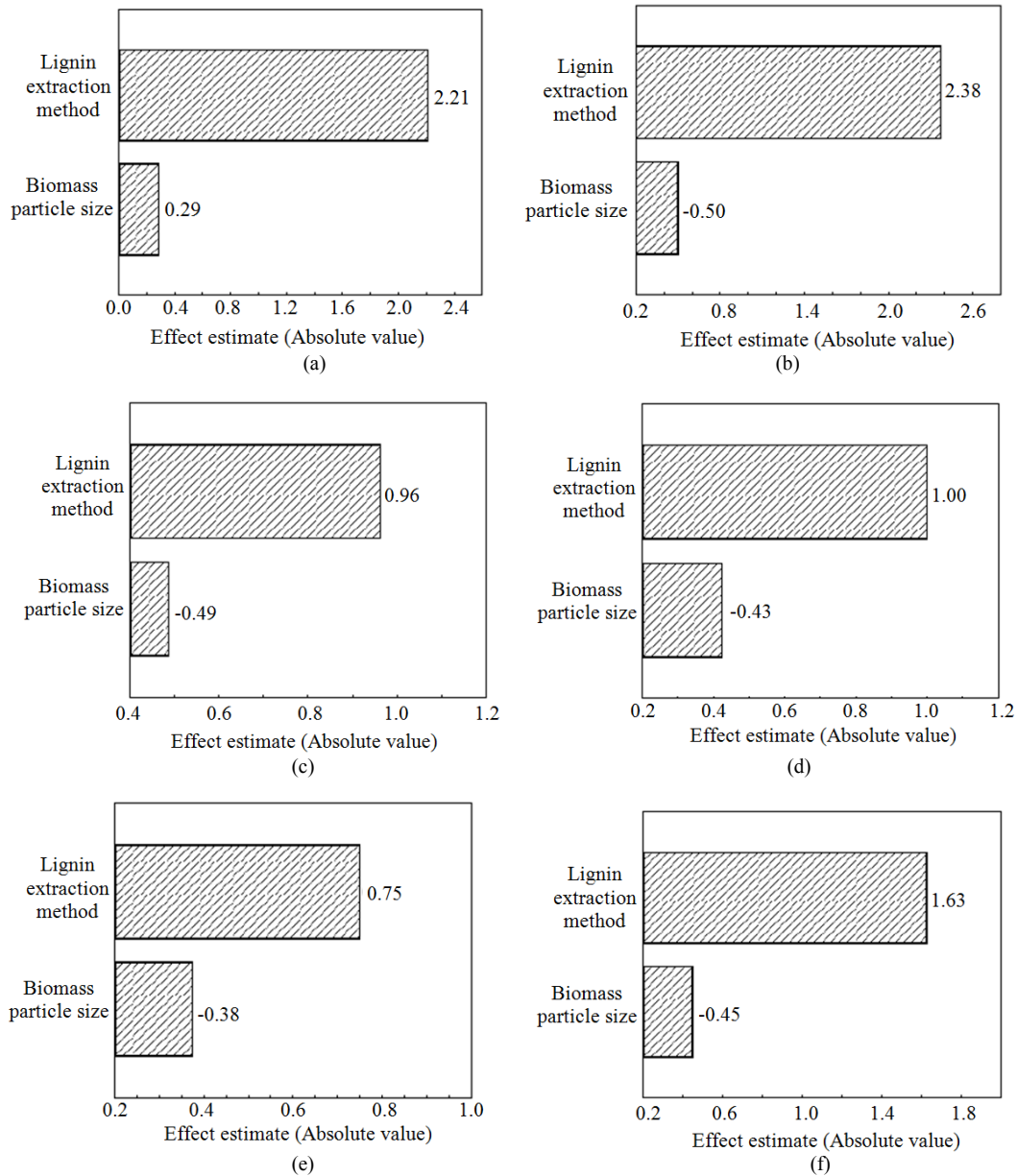


Fig. 1. Pareto charts of effects: (a) sugarcane bagasse; (b) elephant grass; (c) rice husks; (d) corn straw; (e) wheat straw and (f) wood chips

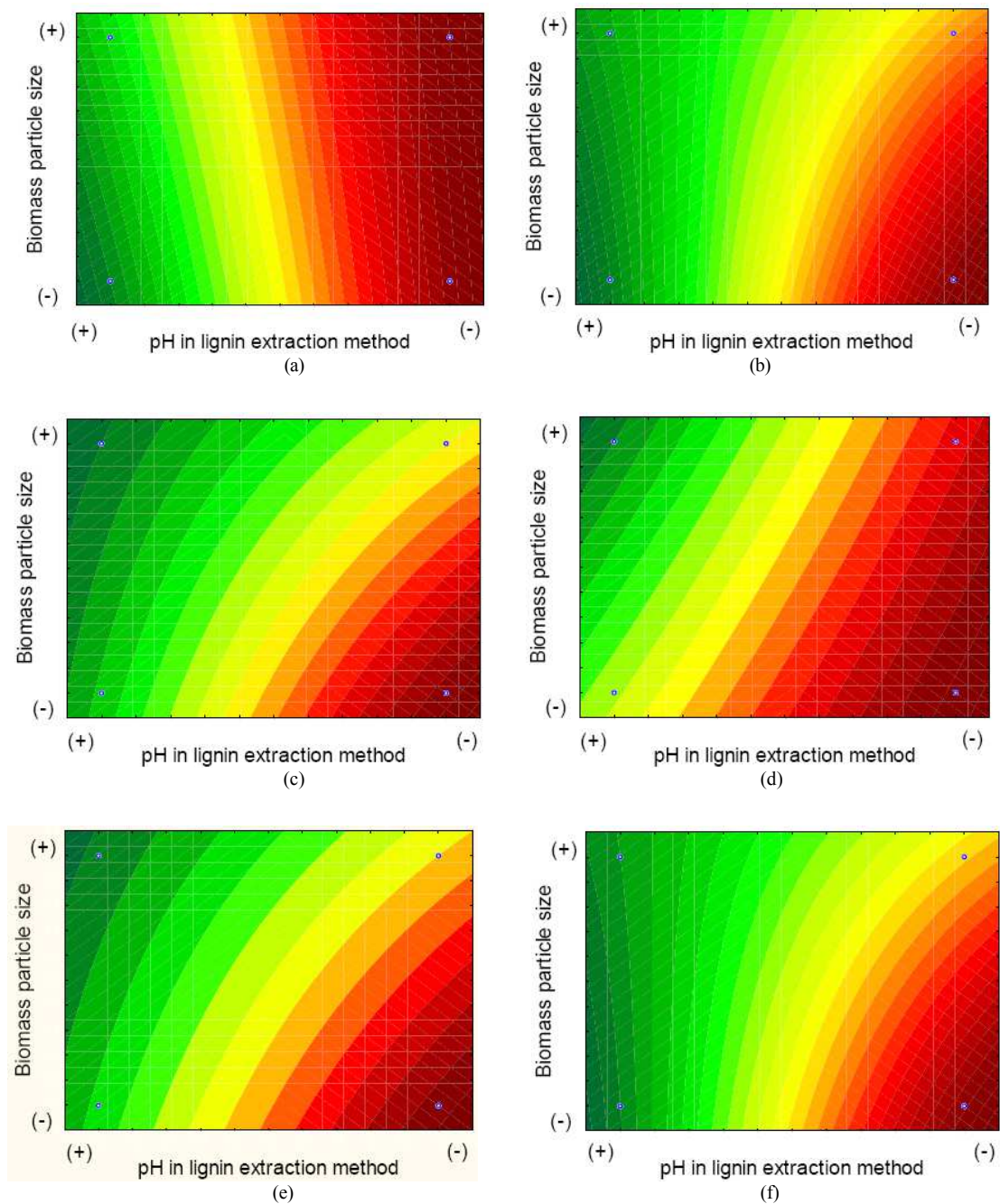


Fig. 2. Response surfaces: (a) sugarcane bagasse; (b) elephant grass; (c) rice husks; (d) corn straw; (e) wheat straw and (f) wood chips

Results indicate that, at 95% of confidence level ( $p\text{-value} \leq 0.05$ ), the lignin yield is affected only by the extraction method in all of cases. In this sense, the extraction carried out through Klason method was always more efficient, once that the effect estimates of Fig. 1 were positive values. This suggests that the lower pH used in the hydrolysis with the Klason method could improve the lignin extraction. In previous works, Zhu and Theliander (2011;

Agarwal *et al.*, 2011) noticed variations in lignin yields and HHV values by changing the pH and temperature during the hydrolysis treatment.

Figure 2 illustrates the surface responses with only tendencies of the lignin yield variation for each biomass in terms of pH used in the hydrolysis for lignin extraction and the mean biomass particle. More studies are needed in order to make available a more accurate prediction for the lignin yield behavior.

Table 7. Lignin yields in raw materials (%)

Run	Extraction method	Particle size range (µm)	Sugarcane Bagasse	Corn straw	Wood Chips	Rice Husk	Elephant-grass	Wheat straw
1	Klason	1000-2000	25.4	20.9	26.5	24.9	24.4	20.0
2	Klason	105-500	25.1	21.1	28.5	25.9	25.3	20.7
3	Willstatter	1000-2000	22.5	20.0	25.1	23.2	22.6	19.4
4	Willstatter	105-500	22.2	20.1	25.9	23.4	22.5	19.6
5	Klason	1000-2000	25.0	20.5	27.3	23.6	24.3	20.1
6	Klason	105-500	24.9	20.7	28.4	24.1	25.3	20.5
7	Willstatter	1000-2000	22.1	19.4	26.9	23.0	22.5	19.5
8	Willstatter	105-500	22.4	19.7	26.5	23.4	21.9	19.7
9	Klason	1000-2000	24.7	20.7	27.0	23.6	23.4	20.2
10	Klason	105-500	24.5	20.9	28.0	24.3	24.0	21.6
11	Willstatter	1000-2000	23.3	19.4	25.9	22.6	21.8	20.0
12	Willstatter	105-500	22.6	20.2	25.4	22.9	21.8	20.2
13	Klason	1000-2000	24.5	20.5	27.1	22.7	23.4	20.5
14	Klason	105-500	24.5	21.4	27.0	23.5	26.0	20.2
15	Willstatter	1000-2000	23.4	19.6	25.7	23.2	22.2	19.6
16	Willstatter	105-500	22.4	20.3	25.4	23.2	21.8	19.8

Table 8. Analysis of variance for lignin yields

Biomass of the lignin	Factor	Sum of squares	Degrees of Freedom	Mean square	F0	p-value
Sugarcane bagasse	Extraction method	19.58	1	19.58	117.63	0.000
	Biomass particle size	0.33	1	0.33	1.98	0.184
	Error	1.99	12	0.16		
	Total	21.98	15			
Elephant-grass	Extraction method	22.56	1	22.56	72.68	0.000
	Biomass particle size	1.00	1	1.00	3.22	0.097
	Error	3.72	12	0.31		
	Total	29.69	15			
Rice husk	Extraction method	3.70	1	3.70	7.38	0.018
	Biomass particle size	0.95	1	0.95	1.89	0.193
	Error	6.01	12	0.50		
	Total	10.94	15			
Corn straw	Extraction method	4.00	1	4.00	58.18	0.000
	Biomass particle size	0.72	1	0.72	10.50	0.007
	Error	0.82	12	0.06		
	Total	5.55	15			
Wood chips	Extraction method	10.56	1	10.56	29.78	0.000
	Biomass particle size	0.81	1	0.81	2.28	0.156
	Error	4.25	12	0.35		
	Total	16.83	15			
Wheat straw	Extraction method	2.25	1	2.25	16.41	0.001
	Biomass particle size	0.56	1	0.56	4.10	0.065
	Error	1.64	12	0.13		
	Total	4.58	15			

## Conclusion

In this study the yield and some properties of lignins obtained from several energy crops grows in Brazil were assessed in function of the lignin extraction method and the biomass particle size. For wheat straw, corn straw, rice husk, sugarcane bagasse, wood chips and elephant grass the proximate and ultimate analysis, as well as, the HHV values were in accordance with those found in literature. As expected, all biomass showed lower HHV

values than their lignin fractions. So, even though important restrictions could be imposed for burning the lignin due to presence of corrosive and pollutant compounds, the HHV values ranged from approximately 20.5 to 24.0 MJ/kg shows that the lignin obtained from the raw materials has potential as source of thermal energy to be used in thermochemical processes.

By means of a 2<sup>2</sup> factorial design with 16 runs for planning the lignin extraction tests was possible to establish the statistical significance of the effects

produced by the extraction method and the biomass particle size on the lignin extraction performance. In this sense, it was found that, at 95% of confidence level, only the extraction method is relevant, suggesting that the Klason procedure is more effective than the Willstatter one. The higher lignin extraction yield obtained with the Klason method was attributed to the lower pH used in the hydrolysis treatment. On the other hand, the biomass particle size had significant effect on the lignin extraction for some raw materials. In such cases, samples with minor particle size lead to more lignin yield, suggesting the influence of the superficial area in contact during the hydrolysis.

More studies focused in quantifying the emissions and other thermochemical properties of lignins become necessary in order to find alternatives for the safe use as fuel or intensify its application in the petrochemical industry.

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## Author's Contributions

This study is a result of the full collaboration of all the authors.

**Meanwhile, Diogo José Horst:** responsible for the collection of samples and laboratorial essays.

**Jhon Jairo Ramírez Behainne:** Coordinated the data analysis and statistical treatment.

**Pedro Paulo de Andrade Junior:** Interpreted the data and organized the study by reviewing it critically.

**Luís Filipe Serpe:** Contributed with the writing and was responsible for the grammar check and final artwork.

## Ethics

The authors declare that this is an original research and do not have any ethical issues or copyrights conflict.

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