

STEAM EXPLOSION TREATMENT OF *EUCALYPTUS GLOBULUS* WOOD: INFLUENCE OF OPERATIONAL CONDITIONS ON CHEMICAL AND STRUCTURAL MODIFICATIONS

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This study evaluates the use of steam explosion (SE) as a pre-treatment of *Eucalyptus globulus* chips, aimed at making this wood suitable as feedstock in biorefineries or alternative processes such as biopulping. Several SE treatments were applied, modifying the following variables: previous hydration of the chips, number of SE cycles, and duration of the first cycle. Chemical composition and structural properties were analyzed after all treatments. Our results show that acetone and hot water extractives contents increased between 1.0% and 6.2% and between 3.6% and 7.1%, respectively, depending on SE operational conditions. Holocellulose content was also observed to decrease (9.4 to 15.6%), while the Klason lignin underwent a slight solubilization. Furthermore, greater water retention capacity and new bigger sized pores were found in the exploded samples. It is reasonable to expect that these changes would facilitate the subsequent implementation of chemical or biological treatments of the cellulose fraction in the biorefinery design.

Keywords: *Eucalyptus globulus*; *Biorefinery*; *Steam explosion*; *Porosity*

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INTRODUCTION

Extensive research efforts to improve the processing and digestion of biomass for several applications have been pursued in the last few years. Steam explosion (SE) is one of the most widely used treatments for fractionation of lignocellulosic components. It is defined as a process by which the lignocellulosic material is exposed to high pressure steam before it undergoes very rapid decompression. This sudden decompression leads to an “explosion” of the steam inside the lignocellulosic matrix, which promotes breakdown and defibrillation of its structure, hydrolysis of the hemicelluloses, and depolymerisation/repolymerization of the lignin (Ruiz et al. 2008). Thus, the accessibility of the material to chemical or enzymatic degradation is greatly increased (Moniruzzaman 1996; Martín-Sampedro et al. 2011). The advantages of SE include a significantly lower environmental impact, low capital investment, and fewer hazardous chemicals applied in the process compared to other methods for hemicelluloses extraction (Li et al. 2001). On the other hand, the presence of degradation products from sugar and lignin is unavoidable and must be taken into account in order to minimize the potential inhibitory effect on subsequent steps such as the enzymatic hydrolysis (Tengborg et al. 2001).

The use of SE in wood processing has increased considerably in the last years due to its potential to produce chemical and structural changes in the lignocellulosic material (Ruiz et al. 2008; Viola et al. 2008; Sun and Chen 2008; Cara et al. 2008). This potential means that SE can be exploited in a way that opens new opportunities in several industrial, energetic, and environmental fields. For instance, this technology has been considered an alternative to conventional chemi-mechanical (CMP) and chemi-thermo-mechanical pulping (CTMP) (Ahvazi et al. 2007). The SE softens lignin, which favours a more intense defibrillation. However, SE pulping has not been industrially implemented yet, because the steam exploded pulps have failed so far to show any superior properties over those from CMP or CTMP (Heitner et al. 1993). Other authors have used SE as a pre-treatment in chemical pulping, and have suggested that, because it facilitates a more efficient diffusion of cooking liquor into the chips, the open structure of the exploded chips might lead to a higher yield and lower rejects in kraft pulping (Ahvazi et al. 2007; Martin-Sampedro et al. 2011a). SE has also been applied to improve delignification during the laccase treatment prior to kraft pulping or in biobleaching sequences (Martin-Sampedro et al. 2011b,c).

Similarly, a considerable research effort is being applied to develop new strategies designed to find renewable bio-fuels extracted from the biomass. The biomass types considered for ethanol production include lignocellulosic material such as wood, annual crops, and agricultural and forestry residues. It has been widely recognized that the rate and extent of enzymatic hydrolysis of lignocellulosic substrates are influenced not only by the effectiveness of the enzymes but also by the chemical, physical, and morphological characteristics of the substrates (Chandra et al. 2007). Thus, a pre-treatment of the lignocellulosic material is required to improve all these aspects. SE is suitable for this end because it is known to induce autohydrolysis and defibrillation. Therefore, several authors have reported on the use of SE in bioethanol production as pre-treatment of different biomass sources, such as sunflower stalks (Ruiz et al. 2008), wheat straw (Sun and Chen 2008), olive tree pruning (Cara et al. 2008), soybean hulls (Corredor et al. 2008), logdepole pine (Cara et al. 2008), or eel grass (*Zostera marina*) (Viola et al. 2008).

For all the reasons above mentioned, the SE pre-treatment of lignocellulose appears to be an excellent tool for improving the production of the two main cellulose derivatives: pulp and ethanol. Consequently the aim of this study was to assess the effect of the SE pre-treatment on the chemical and structural modifications of *Eucalyptus globulus* chips. To achieve that goal the chemical composition, water retention capacity, relative porosity, and relative surface of the material were determined.

EXPERIMENTAL

Raw Material

E. globulus chips were kindly supplied by La Montañanesa pulp mill (Spain). The material was air dried until reaching constant moisture, and then homogenized in a single stock (by conditioning inside polyethylene bags) to avoid differences in composition and water content. The chips were stored in polyethylene bags at 25° C.

Steam Explosion Treatment

The SE treatment was performed in a 26 litre stainless steel digester capable of reaching a temperature of 190 °C and a pressure of 1.37 MPa (14 kg cm⁻²). The digester was equipped with a tubular heat exchanger, electrovalves for steam admission, and a ball valve of discharge. The steam generator was a Babcock Wanson VAP 250RR boiler (Erando, Spain), with a maximum steam production of 270 kg h⁻¹ and a working pressure of 1.37 MPa. A sketch of the digester is shown in Fig. 1.

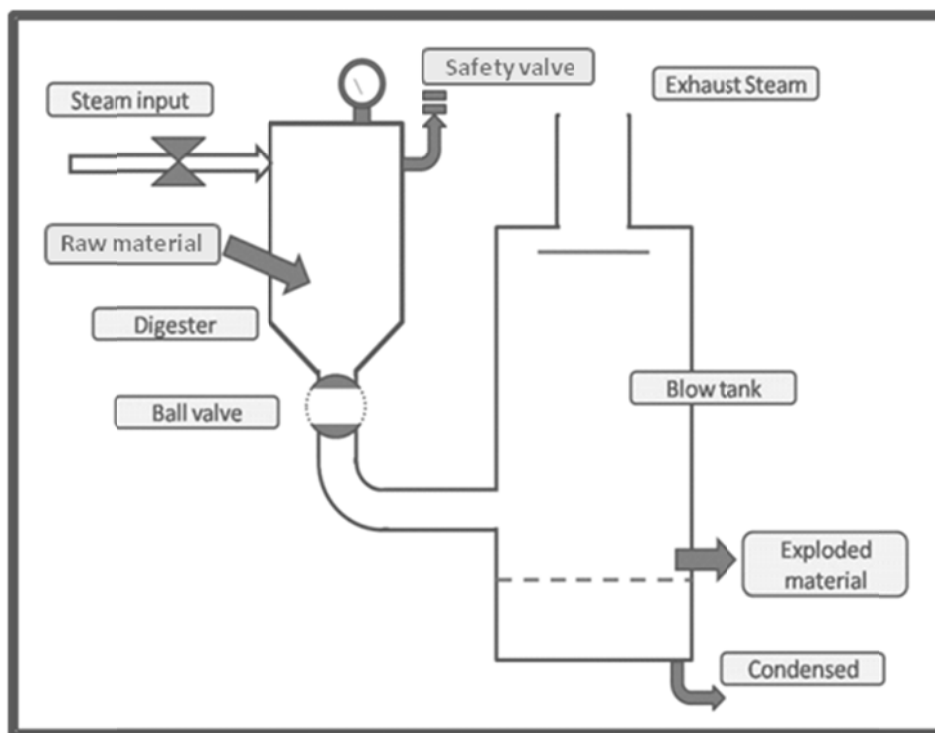


Fig. 1. Sketch of the digester used in the SE experiments

The SE conditions assayed were: number of cycles of treatment (once or twice), duration of the first cycle (5 or 10 minutes), and presence or absence of pre-hydration (presence: immersion in water for 16 hours at 25° C; absence: no previous immersion in water). This last variable was studied in order to evaluate the effect of the uniformity of impregnation on the subsequent steam explosion treatment.

The experimental design was as follows: the raw material was divided into ten samples of 500 g, which were used in eight experiments (2³) and two controls, corresponding to hydrated and non-hydrated samples. Common operational conditions to all treatments were: temperature of operation, 183 °C; treatment pressure, 0.98 MPa (10 kg cm⁻²); discharge pressure, 0.59 MPa (6 kg cm⁻²); and duration of the second cycle, 3 minutes. All the specific variable values for the two controls and all the treated samples are listed in Table 1. Chips were introduced into the digester, and operational conditions were reached and maintained by charging steam. At the end of the steam treatment (after 5 or 10 minutes, according to Table 1), the pressure was reduced to 0.59 MPa (for security reasons), and then the chips were suddenly discharged into the blowing tank at

atmospheric pressure. When a second cycle was carried out, the exploded chips obtained in the first cycle were washed with cold water and then subjected to a second cycle following the same procedure. After treatment, the samples were thoroughly washed with water, dried at room temperature and stored in sealed polyethylene bags.

Table 1. Variable Values for Each Control and SE Treated Sample

Sample	Previous hydration	Number of cycles of treatment	Duration the first cycle
C1	No	-	-
C2	Yes	-	-
NH-5	No	1	5
H-5	Yes	1	5
NH-10	No	1	10
H-10	Yes	1	10
NH-5+3	No	2	5
H-5+3	Yes	2	5
NH-10+3	No	2	10
H-10+3	Yes	2	10

Chemical Analysis

In order to carry out the chemical analysis, all the samples were dried at room temperature and then milled in a Wiley mill. The samples were sieved using standard sieves to obtain 20 g of wood meal sized between 0.30 and 0.40 mm. Acetone extractives (UNE-EN ISO 14453), hot water extractives (UNE 57-013-82), lignin content (TAPPI T 222 om-88), and holocellulose content (Wise et al. 1946) were then measured on the wood meal. All determinations were duplicated.

The concentrations of furfural and hydroxymethyl furfural (HMF) in the liquid fractions obtained during the experiments were also determined by High Performance Liquid Chromatography (HPLC) using the Agilent Technology 1200 series RID (Las Rozas, Spain) with an Aminex HPX-87H column (Bio-Rad, Alcobendas, Spain). The equipment was operated at 50 °C with a mobile phase containing 5 mM sulfuric acid pumped at a rate of 0.6 mL min⁻¹.

Water Retention Capacity

The water retention, or hydration capacity of the treated and untreated chips was defined as the weight of water absorbed by the chips after being immersed in water at 25 °C for 6 hours (Martin-Sampedro et al. 2011a). It was expressed as grams of water per 100 grams of oven-dry wood.

Porosity and Surface Analysis

The relative pore volume and the relative surface of the samples were determined by mercury intrusion porosimetry (MIP) using a CEI instrument, Pascal 140 / 240 porosimeter, equipped with the Software version 1.03B-2. The samples were dried at 60 °C overnight. IUPAC (Rouquerol et al. 1994) values for mercury contact angle (141°), Hg surface tension (484 m N m⁻¹), Hg density (13.534 g mL⁻¹), and Hg temperature (25°C) were used for these measurements. A cylindrical model of the pores was used in the assays.

RESULTS AND DISCUSSION

Chemical Analysis

The SE had a clear effect on the chemical composition of the treated chips. Figures 2a and 2b show the acetone and hot water extractives. As can be observed in Fig. 2a, there was a clear increase in the percentage of acetone extractives in all the exploded samples. This increase could be partly associated with the degradation of lignin caused by the treatments (Chandra et al. 2007). Longer times of treatment resulted in higher extractive percentages (increase of 1.3-3.1% and 2.4-6.2% for 5-10 min of treatment, for hydrated and non-hydrated samples, respectively), but when a second SE cycle was added, similar or even lower percentages of acetone extractives were found (increase of 1.0-4.5% and 3.2-4.2% for 5-10 min of treatment, for hydrated and non-hydrated samples respectively). This is likely to be caused by the removal effect of the second cycle. The pre-hydration of chips did not always show a meaningful effect but seemed to be more relevant during the first cycle with long times of treatment.

As Fig. 2b reveals, SE also increased the percentage of hot water extractives in all the treated samples, with differences of 3.6% to 7.1%. The increase seemed to be independent of the treatment duration, number of cycles, and previous hydration, which suggests that a threshold severity value, causing the maximum formation of water soluble compounds, was reached even with the mildest treatment.

There was a significant decrease in the holocellulose content after every treatment (Fig. 2c). The relative value was reduced from the 74-77% (controls) down to 61-65%, regardless of the SE conditions. A similar reduction of holocellulose content took place in all the treated samples, which could indicate that the severity of the treatments was enough to reach a constant degree of holocellulose degradation, causing an increase in hot water extractives, as already described. However, if steam explosion yield is taken into account, it is possible to determine the percentage of the original holocellulose that remains in the solid after treatment, and therefore, estimate the real holocellulose degradation. Table 2 shows the treatment yields and the percentages of holocellulose and Klason lignin that remain in the solid fraction after each steam explosion treatment, compared to the original amount in the raw material (C1). As can be observed, the percentage of holocellulose underwent a progressive reduction (due to hemicelluloses solubilization) when the severity of the treatment increased (longer treatment and/or addition of a second SE cycle). The pre-hydration of the chips, also contributed to the solubilization of hemicelluloses.

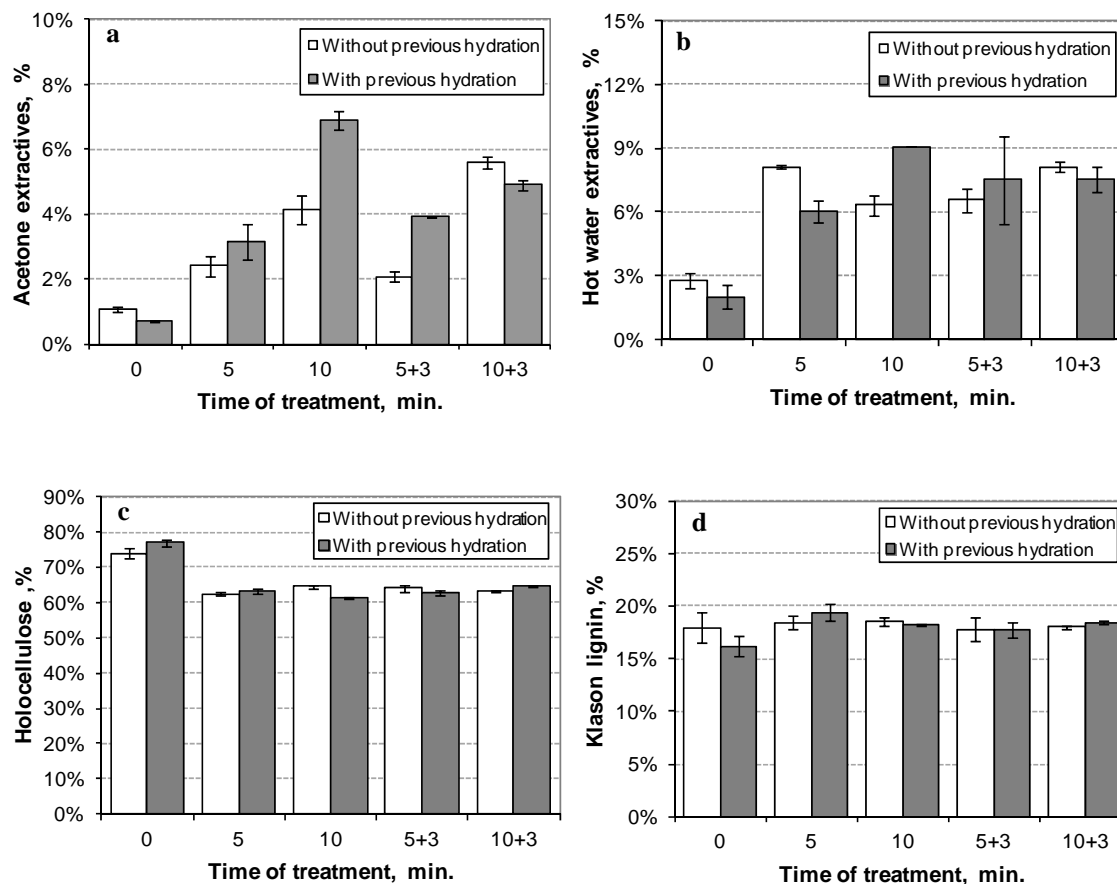


Fig. 2. Relative percentages of acetone extractives (a), hot water extractives (b), holocellulose (c), and Klason lignin (d).

Removal of hemicelluloses with SE under different severity treatments has been well reported. Hendriks and Zeeman (2009) have described the degradation of isolated hemicellulose with steam and the formation of acids that could catalyze further hydrolysis (autohydrolysis). Ruiz et al. (2008) have also reported the degradation of cellulose and mainly hemicelluloses of sunflower stalks but at higher temperatures (180 to 230 °C) than those used in our assays. Other authors have found a loss of 30% of xylose in bamboo (*Phyllostachys pubescens*) treated with SE; xylose is the major component of hemicellulose (Shao et al., 2008). A removal of 47% of xylan was also found after SE of *E. globulus* in a previous report (Martín-Sampedro et al. 2011d).

In the biorefinery, SE poses the risk of producing compounds such as furfural, HMF, and soluble phenolic compounds, which can inhibit enzyme activity (Punsuvon et al. 2008; Ximenes et al. 2010). Formation of these compounds would partly explain the changes in the percentages of acetone and hot water extractives shown in Fig. 2 and would be consistent with the decrease in holocellulose content observed in all the treated *E. globulus* samples. However, in the liquid fractions obtained in our study, neither furfural nor HMF was detected in the HPLC analysis, indicating that hydrolysis reactions

largely predominated over degradation reactions such as dehydration. This finding was probably due to the fact that milder steam explosion conditions were used in the present study (severity factor from 3.1 to 3.5) compared to those applied by other authors (from 3.5 to 4.3) (Li et al. 2005). Therefore, the use of a less intensive steam explosion pre-treatment before enzymatic hydrolysis in bioethanol production could be an alternative option to reduce the inhibiting products. Nevertheless, it is also possible that these volatile products were not quantitatively recovered in the liquid fraction because they were partially lost to the atmosphere after explosive decompression (Emmel et al. 2003).

Finally, Fig. 2d shows that there were no significant changes in the Klason lignin content. The results could indicate that no quantitative changes occurred in the lignin fraction after SE. However, the darkening of the treated chips suggests that some kind of reaction, possibly condensation, took place, making the lignin macromolecule more resistant to solvent extraction. Several authors (Donaldson et al. 1988; Michalowicz et al. 1991) have reported that SE did not remove lignin but induced a redistribution of lignin as a result of melting and agglomeration due to surface tension effects generated during the SE treatment. However, other authors (Excoffier et al. 1991) have reported that lignin softens under the heat and slowly depolymerizes, causing the solubilization of a small part of the original lignin. Thus, Table 2 shows that the percentage of Klason lignin remaining in the solid (compared to the original amount in the raw material) decreased when the severity of the treatment increased. This lignin solubilization agrees with the increase in acetone extractive above mentioned.

Table 2. Steam Explosion Yield and % of Holocellulose and Klason Lignin Remained in the Solid after the SE Treatment, Compared to Raw Material (C1)

Sample	Steam explosion Yield	% Holocellulose remained	% Klason lignin remained
C1	100	100	100
C2	99.9	100	96.2
NH-5	94.6	80.0	97.2
H-5	94.8	81.3	100
NH-10	93.8	81.9	96.6
H-10	85.5	71.0	86.5
NH-5+3	90.4	78.4	89.7
H-5+3	88.1	74.8	86.9
NH-10+3	87.6	74.9	87.9
H-10+3	79.7	69.8	81.8

Water Retention Capacity

The hydration capacity of the steam-exploded chips during a time interval gave evidence of changes in the internal structure of the material and the macro-structural effects of the different SE treatments. As shown in Fig. 3, there was a significant increase in the water retention capacity of all samples after SE, both in non-hydrated and hydrated

samples. This effect was more pronounced for more intense treatments: two cycles or longer durations.

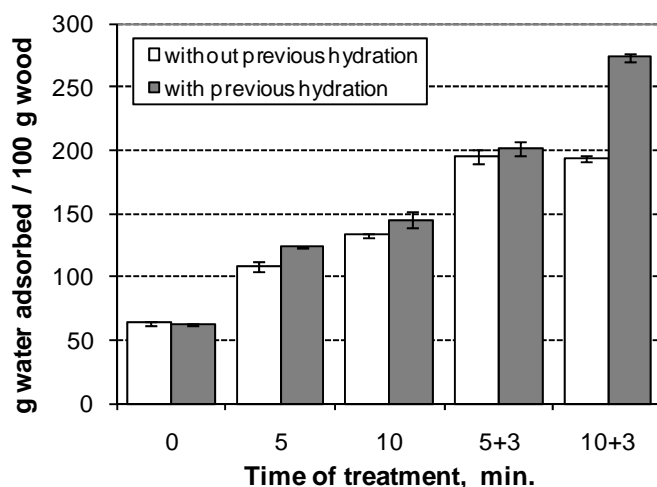


Fig. 3. Water retention capacity

Longer treatments enabled a higher amount of saturated steam to penetrate into the chips, and so decompression resulted in a more intense defibration of the chips. Two cycles of SE also increased the internal rupture of wood, and its effect was even more outstanding than that of extending the treatment. The consequence, in both cases, was an improved capacity of the eucalyptus chips to retain water because there was more available surface inside them.

An increase in water retention was also observed when the chips were hydrated before SE. Previous hydration replaced air by water inside the wood chips. It is supposed that, in non-hydrated wood chips, occluded air partially prevents steam penetration into the material. As a result, decompression brings about a less intense decomposition in non-hydrated chips, which consequently, show lower water retention. Pre-hydration is also likely to have caused a removal of extractives, making steam penetration easier and increasing the hydrophilicity of the material.

Since the increase in water retention capacity is a consequence of the opening of new spaces within the material, SE can be used either to improve the accessibility of chemicals in kraft pulping, as previously reported (Martín-Sampedro et al. 2011a), or, as some postulate (Ruiz et al. 2008), to facilitate enzyme penetration into the lignocellulose, in the biorefinery concept. In this last scenario, a more detailed description of the size and distribution of new pores in the wood will be needed to estimate the probability of enzyme penetration (see next section).

Structural Properties

A structural characterization, assessing the changes in the porous network and the surface configuration of the material, was developed as part of our research. This part of our study was intended to reveal structural modifications in the treated material that might be related with changes in the pore size distribution and accessible surface. As the

accessibility of the material to penetration of chemical reagents and enzymes can be directly related with the effectiveness of the chemical or biological processes, these structural changes would have a great relevance.

The results showed a heterogeneous and broad band of relative pore volumes, with pore size diameters ranging from 20 to 100 micrometers (data not shown) that would correspond to the lumen space of the cell and vessels.

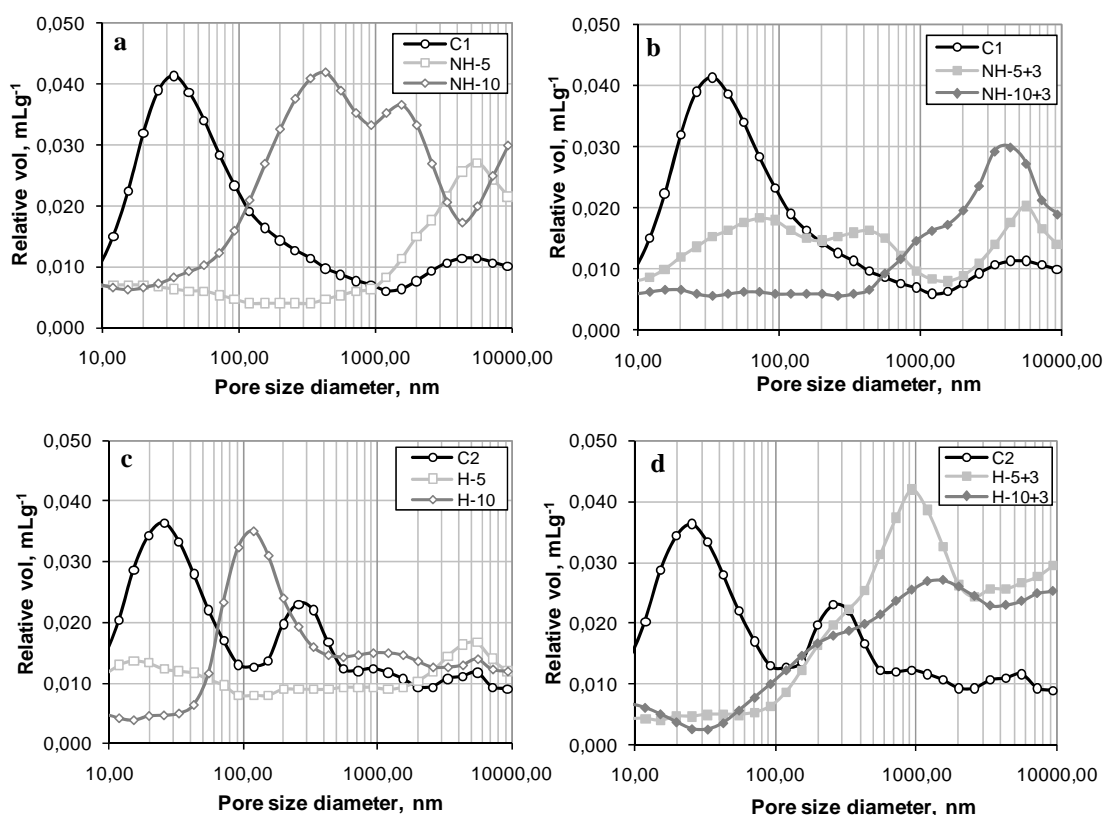


Fig. 4. Relative pore volume versus pore size diameter. Samples without previous hydration (NH) (a and b), samples previously hydrated (H) (c and d). C1 and C2 indicate control without and with previous hydration respectively. The numbers after NH or H indicate the duration of the cycles in minutes.

Figure 4 shows changes in relative pore volume (mL g^{-1}) versus pore diameter of samples without previous hydration, and with one (Fig. 4a) and two cycles (Fig. 4b) of SE. It should be noted that the control is a unique sample in both figures, and that it exhibits a band with a maximum ca. 33 nm. This broad band included the greater pore volume of the control sample and ranged from 10 to 200 nm of pore diameter. The control also showed a lower peak, between 3 and 4 μm , which corresponds to bigger but less abundant pores. In the two samples treated with one cycle of SE (5 and 10 minutes), there was a significant displacement of the band to greater pore size diameters. In this pore size diameter range, the 10 minute treatment produced the biggest pore volume. The addition of a second cycle generated pores with diameters between 3-4 and 6 μm . Again,

the biggest pore volume in this range resulted from the longest treatment. The structural analysis of previously hydrated samples (Figs. 4c and 4d) showed also a significant displacement of the pore volume band from a smaller pore size to a greater pore diameter. In comparison to the non-hydrated control, the hydrated control sample reveals an additional band around 300 nm. These pores would have been produced during pre-hydration of the sample, caused by solubilization of some wood components in water (Romero et al. 1992). This new type of pores is related to the greater effect that SE has on hydrated samples, as penetration of steam into these newly formed pores leads to the formation of bigger pores than those originated when there is no previous hydration.

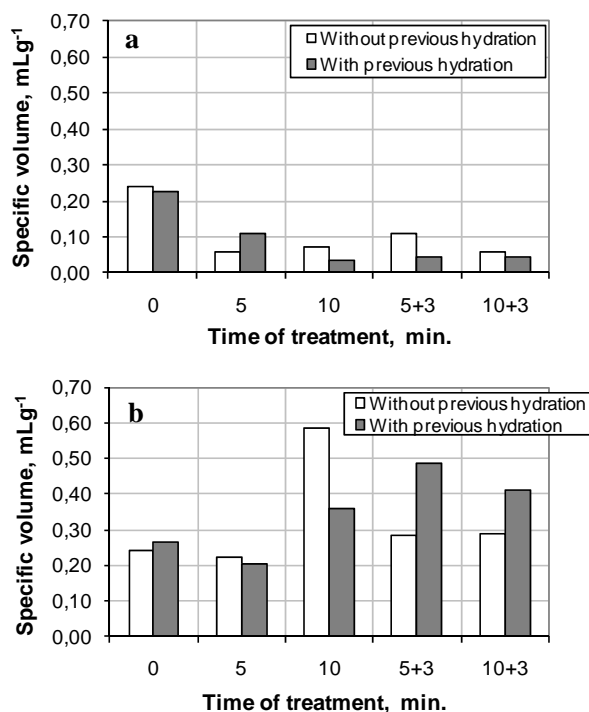


Fig. 5. Mesopore (a) and macropore (b) specific volume for every sample

These results revealed a redistribution of the pore network in the treated material, with a decrease of volume of small pores and an increase of volume of bigger pores. Since there was not an increase of the total pore volume in the pore size range between 10 and 10,000 nm (data not shown), these results can be explained by assuming the formation of pores of longer diameters derived from small pores. This can be observed in Fig. 5, which shows the mesopore (pore diameter: 7-50 nm) and macropore (pore diameter: 50-10,000 nm) specific volume (mL g⁻¹) for every sample analyzed. The results reveal a clear decrease of the mesopore relative volume in all treated samples, both previously hydrated and non-previously hydrated (Fig. 5a). The relative volume of macropores (Fig. 5b) increased in all treated samples, except for those exposed to the mildest SE. In these cases (1 only cycle of 5 minutes), the results were similar to those obtained with the control samples. These results confirm the possibility that there is a reorganization of the pore network rather than creation of new pore volume, in this pore size range. Thus, we may conclude that there is a global positive effect of SE on the

formation of new bigger sized pores that improves the accessibility of chemical reactants and enzymes to the wood chips.

Considering the fact that the analysis of the hydration capacity (Fig. 3) revealed greater water absorption conferred by SE, and taking into account that there is no significant increase of the total pore volume in the diameter range studied here, it is reasonable to conclude that there must be a direct relationship between the new pore matrix of the material and the kinetics of water penetration. The increase in water retention/hydration capacity of the exploded samples indicates that water absorption lasts long enough (6 hours) for water to fill the new bigger pores formed. In the control samples, this phase is not long enough for water to penetrate into the smaller pores, which manifests itself as a clear difference between control and treated samples.

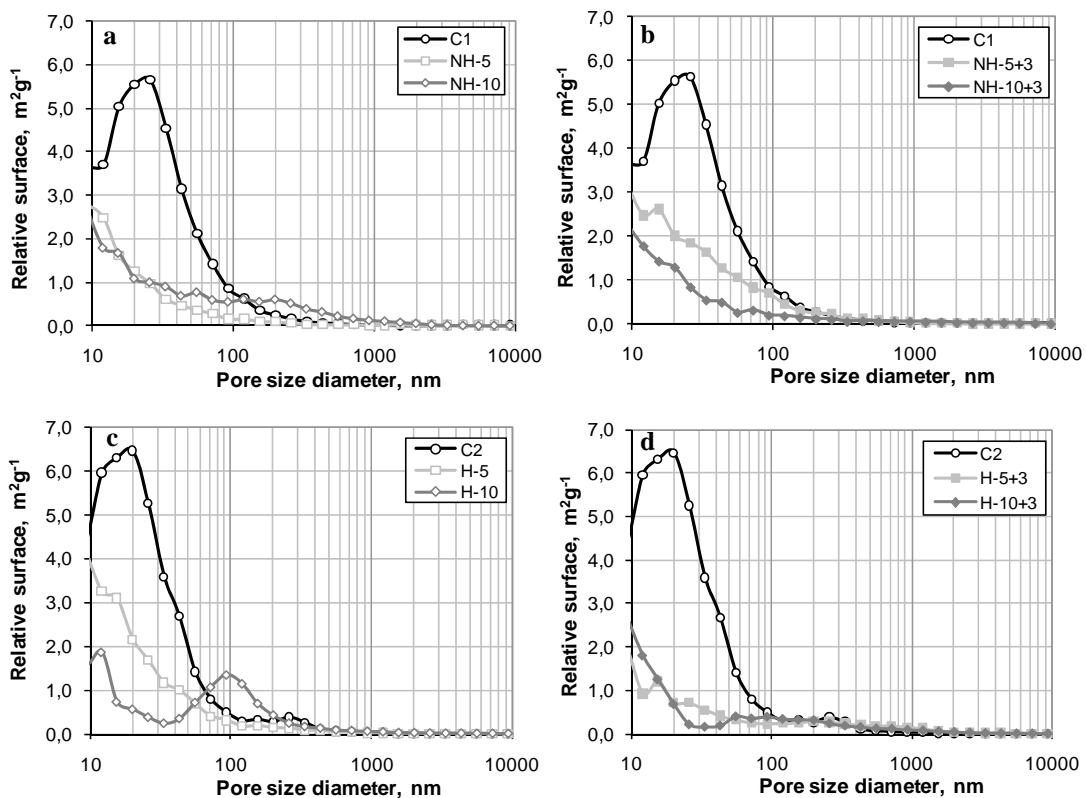


Fig. 6. Relative surface versus pore size diameter. Samples without previous hydration (NH) (a and b), samples previously hydrated (H) (c and d). C1 and C2 indicate control without and with previous hydration respectively. The numbers after NH or H indicate the duration of the cycles in minutes.

The relation between pore size and accessible surface was also evaluated in order to get clear knowledge of the cost, in terms of surface, posed by the formation of bigger pores derived from smaller pores. In Fig. 6 the relative surface area ($\text{m}^2 \text{g}^{-1}$) has been plotted versus pore size diameter (nm) for every sample tested. When the pore diameter ranged between 10 and 100 nm, there was a clear decrease in the relative surface in all the treated samples, which corresponds to the disappearance of small pores in these

samples, as it has been explained above. These results correlated with those shown in Figs. 4 and 5 and confirm the decrease in the relative pore volume for pore size diameters between 10 and 100 nm.

Formation of macropores within the pore size diameter range from 100 to 1000 nm produced only a slight increase of relative surface area, as the ratio surface/volume decreases as pore volume increases. Therefore the relative surface area decreased significantly in all treated samples when the pore size diameter was within the 10 to 10,000 nm range. A direct consequence of these results may be in relation to the balance between greater accessibility of the newly created pore network and the reduction of surface accessible to the subsequent attack of chemical reagents or macromolecules such as enzymes. Depending on the nature of the reaction or process that the material would be subjected to, the porosity and surface changes would have a different effect.

CONCLUSIONS

1. Steam explosion is an appropriate alternative treatment in the fractionation of *E. globulus* wood for different applications, as it produces modifications that could enhance subsequent processes.
2. An increase in acetone and hot water extractives (between 1.0-6.2% and 3.6-7.1%, respectively), and a decrease in holocellulose content (9.4-15.6%), were observed after SE. However, there was only a minor variation of the relative Klason lignin content when comparing the composition of samples before and after steam explosion treatment.
3. Results of studying the water retention capacity, the relative porosity, and the relative surface suggest that the wood structure was redistributed, resulting in a decrease of the volume of mesopores and an increase of macropores in the exploded samples.

ACKNOWLEDGMENTS

The authors wish to thank the Spanish Ministry of Science and Innovation for funding this study via Projects CTQ 2006-00385 and CTQ 2009-11268, the “Ramón y Cajal” Programme, and the FPI fellowship (BES2007 – 14528).

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Article submitted: September 4, 2011; Peer review completed: October 5, 2011; Revised version received and accepted: October 14, 2011; Published: October 18, 2011.