DOI:10.1021/jf901373s

JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY
ARTICLE

Concomitant Changes in Viscoelastic Properties and Amorphous Polymers during the Hydrothermal Treatment of Hardwood and Softwood

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The aim of this study was to understand how the molecular structures of amorphous polymers influence wood viscoelastic properties. Wood from oak and spruce was subjected to hydrothermal treatments at 110 or 135 °C. Wood rigidity, reflected by the wood storage modulus, showed different modification patterns according to the wood species or the temperature level. Because viscoelasticity is dependent on wood amorphous polymers, modifications of lignins and noncellulosic polysaccharides were examined. Hemicellulose degradation occurred only at 135 °C. In contrast, lignins displayed major structural alterations even at 110 °C. In oak lignins, the β-O-4 bonds were extensively degraded and wood rigidity decreased dramatically during the first hours of treatment. Spruce lignins have a lower β-O-4 content and, relative to oak, the wood rigidity decrease due to treatment was less pronounced. Wood rigidity was restored to its initial value by prolonged treatment, probably due to the formation of condensed bonds in cell wall polymers.

KEYWORDS: Hydrothermal treatment; viscoelasticity; wood; lignins; noncellulosic polysaccharides; molecular modifications

INTRODUCTION

Wood is a remarkable natural material mainly consisting of three polymers: cellulose, hemicelluloses, and lignins. Its rheological properties are dependent on the structure, proportion, and interactions of these three constituents (*1*, *2*). Cellulose is a high molecular weight and linear polymer with a uniform chain structure composed solely of glucose monomers linked together by β-1,4-glycosidic bonds. Cellulose microfibrils are partly crystalline (around 60%) and partly amorphous (*3*). Hemicelluloses, unlike cellulose, are heterogeneous polysaccharides composed of various sugar units. The main wood noncellulosic polysaccharides are galactoglucomannans in gymnosperms (softwoods) and 4-*O*-methylglucuronoxylans in dicotyledonous angiosperms (hardwoods) (*4*). Noncellulosic polysaccharides generally form an amorphous substance that has a high content of reactive hydroxyl groups and interacts both with the cellulose chains and with lignins. Lignins are complex polymers made of C_6C_3 units, namely, *p*-hydroxyphenyl (H), guaiacyl (G), and syringyl (S) units, in various proportions according to their botanical origin. Hardwood lignins are composed of S and G units, together with trace amounts of H units. Softwood lignins consist mainly of G units and low levels of H units (*5*). The most frequent interunit bond in native lignins is the β-O-4 linkage (structure A, **Figure 1**). Lignins also contain resistant linkages, referred to as condensed bonds, the main ones being the β-5, 5-5, β-β, 4-O-5, and β-1 linkages (structures B−G, **Figure 1**). The 5-5 and 4-O-5 linkages form the branching points of the polymers (*6*). Between these branching points are linear fragments consisting of units linked by β-O-4, β-5, or β-β bonds. Typical β-1 bonds are found in the spirodienone structures (F, **Figure 1**) (*7*) and the diarylpropane structures (G, **Figure 1**) obtained by rearrangement with chain cleavage of the former. The frequency of the various linkages is dependent on the proportion of H, G, and S units. Because of the availability of the aromatic C-5 position for coupling, lignins mainly composed of G units contain more β-5, 5-5, and 4-O-5 bonds than lignins containing S units (*6*). Lignins are closely associated with noncellulosic polysaccharides to form the so-called lignin−carbohydrate complexes (LCCs). These LCCs constitute the basis of the amorphous network embedding the cellulose microfibrils.

Wood properties are extremely affected by moisture and temperature levels. These two parameters are essential to many processes in the timber industry (*8*). Indeed, water acts as a plasticizer, which significantly decreases the glass transition temperature of the wood components. The elastic modulus of

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 Published on Web 07/20/2009

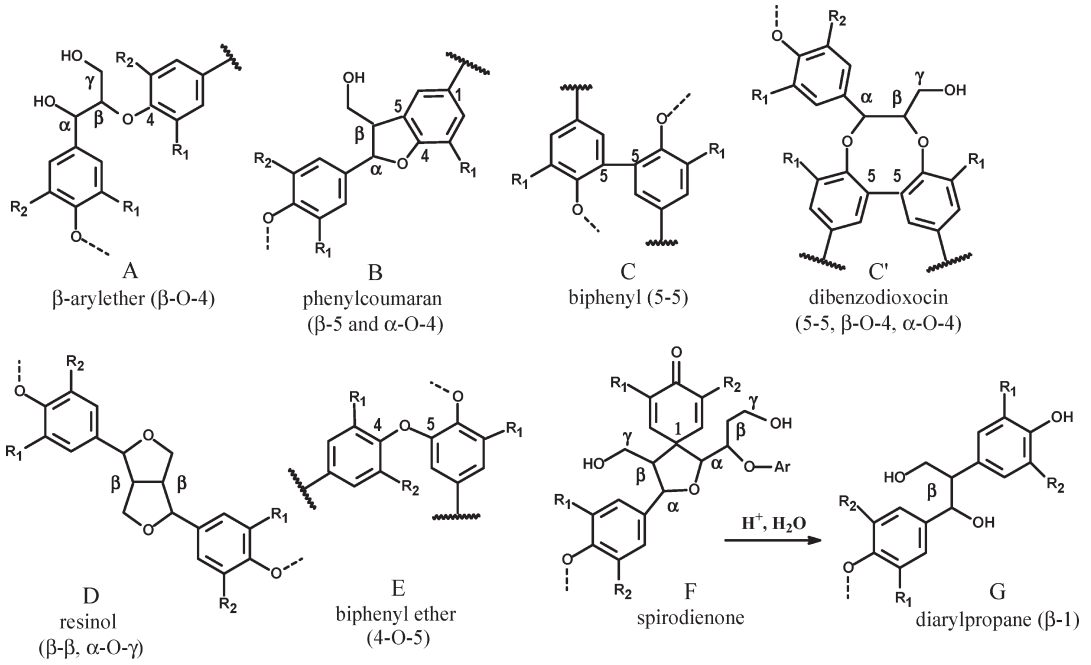

Figure 1. Main bonding patterns evidenced in native lignins ($R_1 = R_2 = H$ in *p*-hydroxyphenyl units; $R_1 = OMe$, $R_2 = H$ in guaiacyl units, $R_1 = R_2 = OMe$ in syringyl units). The 5-5 bonds are encountered in ordinary biphenyl structures C as well as in macrocyclic dibenzodioxocin structures C′. The β-1 bonds occur in the spirodienone structure F and in the diarylpropane structures.

wood and of its constituents is also affected by water adsorption at positive temperatures. Moderate heating at below 100 °C brings about wood softening. The temperature level provides the necessary activation energy to break weak or even covalent intermolecular linkages, hence permitting crawling movements between the macromolecular chains. It is generally accepted that the softening of wet wood occurs between 50 and 100 °C, depending on the wood species and type and on the material direction (*1*). The transition occurring in this temperature range for soaked samples is attributed to lignins (*9*, *10*). The viscoelastic properties of wet wood have been previously studied above 100 °C and mainly by quasi-static methods (*11*−*16*). Heating wood at higher temperatures generates thermal degradation, which also depends on the moisture level. Significant thermal degradation of dry wood occurs only above 180−200 °C for a duration of several hours, but with soaked samples this threshold drops to around 100 °C (for similar treatment durations) (*17*, *18*). The effect of thermal treatment on dry solid wood has been widely studied. Thermal treatment is an interesting process and confers on wood remarkable properties (durability, dimensional stability) to the detriment of the mechanical properties. A dry thermal treatment between 150 and 200 °C provokes noticeable structural modifications of the wood polymers. An increase in cellulose content and crystallinity (*19*, *20*) is reported, which is assigned to preferential degradation of the amorphous polysaccharide fraction. In addition, thermolysis of the noncellulosic polysaccharides is observed, with mainly the removal of acetyl groups (*21*, *22*). The kinetics and extent of noncellulosic polysaccharide degradation are dependent on the types and amounts of acetyl groups. The impact of dry thermal treatments on lignins has also been studied (*19*, *22*−*24*). According to these literature data, the main thermally induced reactions in lignins are the cleavage of β-O-4 bonds and the formation of free phenolic groups and vinyl ether structures, as well as some condensation reactions (*22*−*24*). The impact of a wet thermal treatment is less known. The presence of water affects degradation by promoting hydrolysis, particularly in the presence of acetic acid, which is released from acetylated noncellulosic polysaccharides and which catalyzes their depolymerization (*25*, *26*). Funaoka et al. (*17*) showed that the water contained in wood was favorable to lignin condensations. The presence of hemicellulose-derived acetic acid is also a factor promoting condensation reactions in lignins (*27*).

The aim of this study was to evaluate the effects of hydrothermal treatments on concomitant changes in rheological properties and the amorphous polymers of wood samples. Because the viscoelastic properties of wood are widely determined by these amorphous polymers, viscoelasticity was continuously measured throughout the hydrothermal treatment and the most relevant treatment durations for chemical analyses of the noncellulosic polysaccharides and lignins were determined. As plant taxa display large structural variations in these polymers, wood samples from a gymnosperm (spruce) and a dicotyledonous angiosperm (oak) were studied.

MATERIALS AND METHODS

Wood Sampling. Wood from a hardwood (oak, *Quercus sessiliflora*) and a softwood (spruce, *Picea abies*) were harvested in eastern France, from plantations belonging to the Forestry School of Nancy (AgroParisTech/ENGREF). Test samples were cut in a radial direction from the heartwood of green logs. The sample cross sections were 6 × 10 mm^2 (longitudinal × tangential) and 100 mm long (radial). Care was taken to select a zone of "perfect" wood, that is to say, mature wood without any reaction wood or singularity.

Rheological Measurements. Viscoelastic properties were measured in the WAVE^T apparatus (Environmental Vibration Analyzer for Wood). This device, developed by the biomaterials team of LERFOB (AgroParisTech/INRA), allows a rigorous dynamic mechanical analysis (DMA) of samples in water-saturated conditions (**Figure 2**). This patented apparatus has been described previously (*28*, *29*). Samples were immersed in a temperature-controlled water bath throughout the test, at a pressure level of up to 0.5 MPa bar, which is required to prevent the water from boiling at 100 °C under normal conditions. Specimens were tested in a bending configuration based on a rigorous single cantilever mode. Sinusoidal solicitations were applied to the sample with a zero mean stress to avoid any deflection due to long-term creep. In all cases, samples were

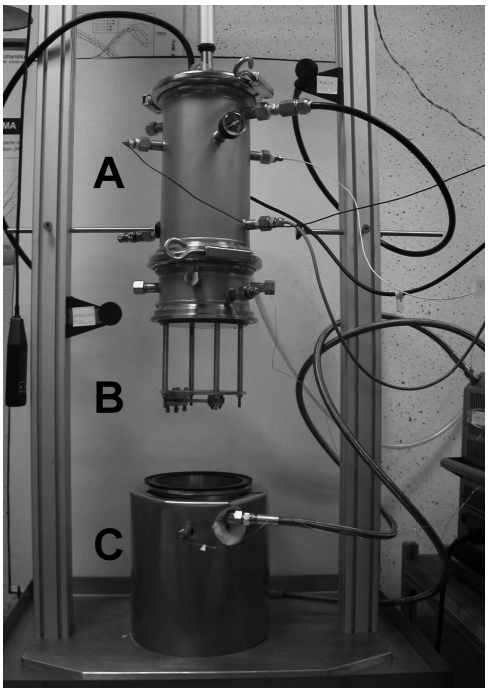

Figure 2. Partial view of the WAVE^T apparatus (Environmental Vibration Analyzer for Wood (*29*)) with its principal specifications: (A) instrumented part of the test bench (the sinusoidal load applied to the sample during the hydrothermal treatment has a frequency which can be adjusted between 5×10^{-3} and 10 Hz); (B) sample holder (specimen tested in a bending configuration with a specific clamp ensuring that a pure vertical force is applied); (C) conditioning room (temperature-controlled water bath working in the 5−140 °C range and at a pressure increasing up to 0.5 MPa).

tested within the linear viscoelastic range. The maximum strain value was between 0.01 and 0.1%.

Results of dynamic mechanical analyses are commonly expressed as storage modulus (E'), loss modulus (E''), and loss factor ($\tan \delta = E''/E'$). The storage modulus describes the capacity of a material to store mechanical energy and represents the elastic part of the sample. The loss modulus is the viscous response of the sample and is proportional to the dissipated energy. The loss factor characterizes the damping capacity of the material. When scanning in temperature, the softening temperature is characterized by a decrease in storage modulus and a peak of E'' and $\tan \delta$ (*30*−*32*).

In this work, the WAVE^T was used at constant temperature to study the viscoelastic properties over time. The samples were therefore subjected to a sinusoidal load at a constant frequency of 1 Hz for a period of hydrothermal treatment varying from 1 to 24 h, at two temperature levels (110 and 135 °C). Viscoelastic properties were measured every 4 min. The heating rate used to attain the plateau temperature was 1.25 °C min^{-1}. A minimum of three samples of each species was analyzed to check measurement repeatability. As these rheological measurements revealed large variations in the storage modulus over time, we were then able to select the most relevant treatment durations for chemical analyses of the noncellulosic polysaccharides and lignins.

Soxhlet Extractions. The thermally treated and untreated samples were oven-dried at 140 °C overnight and then ground using an Ikar grinder (to pass through a 0.5 mm sieve) before exhaustive extraction in a Soxhlet apparatus with toluene/ethanol, 2:1 (v/v), ethanol, and then water. Polysaccharide and lignin analyses were performed on the dried and extract-free cell wall materials, referred to hereafter as the cell wall residue (CWR).

Carbohydrate Analyses. Five milligram samples of CWR were subjected to acid hydrolysis using 12 M H_2SO_4 (125 μL, 2 h at room temperature) and then 1 M H_2SO_4 (1.625 mL) for 2 h at 100 °C (*33*). Acid-soluble samples were filtered and injected into a CarboPac PA1 anion exchange column (4 × 250 mm, Dionex) and detected by pulsed amperometry (PAD 2, Dionex) using a postcolumn addition of 300 mM NaOH as previously described (*34*). Monosaccharides were quantified using 2-deoxy-D-ribose as the internal standard. The noncellulosic glucose content was determined by performing a milder acid hydrolysis at 121 °C using 2 M trifluoroacetic acid. The monosaccharides were then separated and quantified as above (*35*).

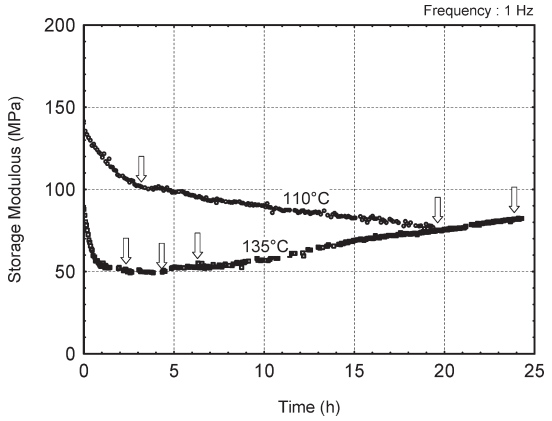

Figure 3. Changes in the storage modulus of oak samples in a radial direction (1 Hz) during two hydrothermal treatments (110 and 135 °C). Arrows indicate the different times at which chemical analyses were performed on samples.

Lignin Analyses. The lignin content in 300 mg of the extract-free samples was determined by Klason method, according to the standard procedure (*36*). The Klason lignin content was calculated as percentage weight of the extract-free wood and expressed as the average of two independent determinations on the same sample. Thioacidolysis and subsequent gas chromatography−mass spectrometry analyses of the lignin-derived monomers were performed according to previously published procedures (*37*, *38*). The thioacidolyzed lignin-derived dimers were determined after desulfurization as previously described (*39*).

RESULTS AND DISCUSSION

Changes in Rheological Properties over Time during the Hydrothermal Treatment. In a previous paper (*40*), it was shown that the temperature level served as an activator of the viscoelastic properties and also as a degradation factor in the case of soaked samples. In this study, the viscoelastic properties of various wood species were shown to be strongly affected by the temperature treatment. The variations in wood rigidity were speculatively assigned to the hydrolysis of hemicelluloses and to the condensation of lignins, without corresponding experimental support. The WAVE^T apparatus is able to distinguish the characteristic mechanical time (test frequency) from the characteristic degradation time (duration of the plateau at the selected temperature) over a wide temperature range. This unique feature enabled us to prove that, in water-saturated conditions, the material itself was altered at temperatures as low as 80−90 °C provided that this level was maintained for several hours.

Figures 3 and **4** depict the changes that occurred in the storage modulus for oak and spruce samples in the radial direction at two plateau temperatures (110 and 135 °C) and when sinusoidal solicitations were applied at a frequency of 1 Hz. For each wood species, three measurements were repeated, which provided profiles similar to the ones outlined in **Figures 3** and **4**. The observed variations in storage modulus values were indicative of changes in material rigidity. These rigidity changes were most likely related to modifications in the chain mobility of the cell wall polymers. Indeed, molecular mobility requires massive and simultaneous bond cleavage, which is less likely to occur at 110 or 135 °C. In addition to bond cleavage, the partial removal of some hemicellulosic components may also enhance the mobility of the cell wall polymers.

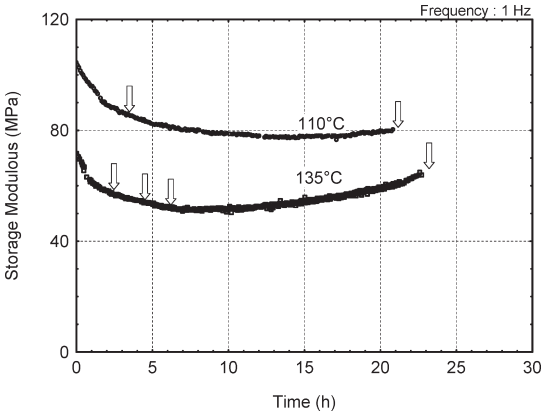

Figure 4. Changes in the storage modulus of spruce samples in a radial direction (1 Hz) during two hydrothermal treatments (110 and 135 °C). Arrows indicate the different times at which chemical analyses were performed on samples.

The rigidity of oak wood at 110 °C (**Figure 3**) decreased as the experiment duration increased. Two degradation time constants could be distinguished: a rapid decrease during the first 4 h was followed by a second phase in which the decrease with time was almost linear. The results obtained at 135 °C were quite different. The storage modulus for oak wood showed a pronounced decrease during the first 2 h, followed by an increase in rigidity during subsequent hours. In fact, despite the reduction of 45% that occurred during the first 3 h of hydrothermal treatment, this storage modulus returned to its initial value after 24 h.

For spruce, the kinetics were quite different (**Figure 4**). The decrease in storage modulus was smaller than for oak. In view of the different cell wall structure and organization in softwoods and hardwoods, this is not surprising. Spruce lignins contain fewer β-O-4 bonds and more 5-5 branching structures than oak lignins. As β-O-4 bonds are the main targets of hydrolytic degradation (*24*) and the 5-5 linkages are stiffening structures (*41*), these structural traits produce a more rigid lignin polymer that is less susceptible to thermal treatment. An increase in the spruce storage modulus was observed during the final phase even at 110 °C. This final increase may represent the thermally induced accumulation of condensed bonds in lignins, as discussed hereafter.

Chemical Analyses. *Lignins and Polysaccharides in Extract-free Oak and Spruce Wood.* As shown in **Table 1** and before thermal treatment, the cell walls of extract-free oak and spruce displayed important compositional differences. Not unexpectedly, the lignin content of the spruce sample was higher. In addition, the spruce lignins consisted almost exclusively of G units, with only low amounts of H units and a lower frequency of units solely involved in β-O-4 bonds. These latter are hereafter referred to as noncondensed lignin units. This lower frequency was apparent as a 2-fold lower yield of lignin-derived thioacidolysis monomers in relation to the oak sample. In contrast, native oak lignins had high contents of both S units and β-O-4 bonds. Their high yield in thioacidolysis monomers meant that, like other hardwood lignins (*39*), about 66% of the oak lignins consisted of noncondensed units solely involved in β-O-4 bonds. According to model studies (*41*), these β-O-4 rich lignin structures display conformational flexibility, which allows them to be adsorbed flat against the surface of polysaccharide chains.

The monosaccharide composition of the noncellulosic wood polysaccharides was determined by acid hydrolysis of the cell wall material and by summing the xylose, mannose, arabinose, galactose, rhamnose, and galacturonic acid contents. The sum of these sugars represented almost 30% of the samples in both wood species (**Table 1**). As expected, xylose was the main sugar (representing 69%), released from noncellulosic oak polysaccharides. The xylan skeleton of hardwoods is known to carry different decorations, including uronic and acetic acids (*42*). When released during hydrothermal treatment, these acidic groups could participate in chemical degradation of the wood cell walls. The second and third most important noncellulosic oak polysaccharides were found to contain mannose and galacturonic acid, these accounting for 14 and 11% of the total noncellulosic sugars, respectively. Finally, arabinose and galactose were released from oak wood in relatively minor amounts (around 2% of the sugar components).

Table 1. Composition of the Extract-free Woods before Hydrothermal Treatment[a]

	wood species	
cell wall component	spruce	oak
lignins		
Klason lignin KLa	27.6 (0.3)	23.0 (0.1)
thioacidolysis monomers (H + G + S) (μmol/g of KL)	1480 (10)	2600 (50)
relative frequency H/G/S (% molar)	1.0/99.0/−	0.8/29.0/70.2
noncellulosic polysaccharides		
total[b]	27.2 (0.9)	28.8 (1.0)
xylose	6.3 (0.2)	20.3 (0.8)
mannose	12.7 (0.4)	3.7 (0.3)
arabinose	1.4 (0.0)	0.5 (0.0)
galactose	1.6 (0.1)	0.7 (0.0)
halacturonic acid	2.8 (0.1)	3.1 (0.1)

[a] All of the compositional data (expressed as weight percentage of extract-free woods) are the mean values of duplicate analyses (standard errors in parentheses). [b] The total noncellulosic carbohydrate content was calculated by summing, arabinose, galactose, rhamnose, xylose, mannose, galacturonic acid and glucuronic acid released after sulfuric acid hydrolysis.

The main sugar released by acid hydrolysis of spruce was mannose (48%, **Table 1**) with xylose second (22% of the total noncellulosic carbohydrate content). Arabinose and galactose were released in similar amounts (5 and 6% of the sugars, respectively). A small proportion of the noncellulosic fraction, determined by mild acid hydrolysis, was due to glucose (8%, data not shown.). This overall sugar analysis was consistent with the fact that galactoglucomannans are the main noncellulosic polysaccharides in softwoods, whereas xylans are the second most abundant (*43*).

Effect of Hydrothermal Treatments on the Amorphous Cell Wall Polymers. Chemical analyses, based on the dynamic mechanical data, were carried out at relevant treatment times that corresponded to large variations of the storage modulus in both wood species. The selected times are marked with arrows in **Figures 3** and **4**. For the tests at 110 °C, 3 and 20 h were selected, which corresponded to the end of the successive exponential and linear decreases of the storage modulus. For the tests at 135 °C, four treatment durations were selected (2, 4, 6, and 24 h) to catch the observed trends in the viscoelastic signature of the two species.

Lignin and noncellulosic polysaccharide analyses were run on the dry and extract-free samples obtained after exhaustive solvent extraction in a Soxhlet apparatus. The yield in extract-free material recovered from the control or hydrothermally treated samples varied from 83 to 89% for oak and from 91 to 95% for spruce, without any clear-cut effect of the hydrothermal treatment. It is, however, very likely that most of the low molecular weight and soluble components formed during treatment of the cell walls were lost in the water in which the samples were immersed, resulting in a sample weight decrease. Sample densities were therefore calculated both before and after the 4, 6, and 24 h

Table 2. Changes in Lignins and Noncellulosic Carbohydrates Occurring during Various Hydrothermal Treatments of Oak and Spruce Species, in Relation to Control Samples[a]

						thioacidolysis lignin-derived monomers		
wood species	T (°C)	time (h)	infradensity (kg m^{-3})	decrease of the storage modulus[b] (%)	Klason lignin[c] (%)	total yield [d]	S/G	noncellulosic polysaccharides [c] (%)
oak	control		590		23.0 (0.1)	2620 (50)	2.42 (0.02)	28.8 (0.9)
	110	3		28	23.2 (0.3)	1870 (10)	2.16 (0.03)	29.9 (1.0)
		20	520	46	23.4 (0.1)	2080 (15)	2.09 (0.01)	27.0 (0.8)
	135	2		38	25.0 (0.2)	1600 (270)	1.97 (0.12)	21.3 (0.3)
		4	479	44	24.4 (0.1)	1370 (40)	1.73 (0.01)	17.2 (0.1)
		6	374	40	24.4 (0.0)	1140 (30)	1.63 (0.00)	15.2 (0.1)
		24	392	7	28.4 (0.1)	730 (80)	1.45 (0.09)	13.0 (0.5)
spruce	control		290		27.6 (0.3)	1480 (10)		27.2 (0.9)
	110	3		16	27.6 (0.5)	1490 (70)		25.6 (0.5)
		20	266	27	27.0 (0.3)	1400 (20)		23.9 (0.4)
	135	2		19	28.1 (0.1)	1210 (30)		23.9 (0.4)
		4	277	24	28.4 (0.1)	1210 (70)		23.5 (0.3)
		6	246	27	28.8 (0.1)	1260 (90)		21.8 (1.0)
		24	221	11	31.1 (0.1)	1170 (3)		15.8 (0.8)

[a] The data are mean values of duplicate analyses (standard errors in parentheses). [b] Calculated from the initial value of the storage modulus that was measured at the temperature treatment after water temperature stabilization. [c] Weight percent of the extract-free material. [d] Total yield of lignin derived monomers expressed in micromoles per gram of Klason lignin.

treatments to determine these weight variations, the pressure applied during treatment tending to promote water impregnation of the wood and mask them (**Table 2**). Because the volumes of the water-saturated samples were quite constant, the density variations very likely paralleled the sample weight variations. The density loss increased with treatment severity (temperature and duration). The density losses with the harshest treatment (24 h at 135 °C) were 34 and 24% in oak and spruce samples, respectively (**Table 2**). These losses suggest that the cell wall polymers were substantially hydrolyzed and/or solubilized during the hydrothermal treatment.

Oak Wood Amorphous Polymers Displayed High Susceptibility to Hydrothermal Treatment. No marked effect on lignin and hemicellulose contents or the density of oak wood samples was apparent at 110 °C, whatever the treatment duration (**Table 2**). However, it was apparent from thioacidolysis that marked changes in oak lignin structure were induced by the 3 h treatment at 110 °C. This result was all the more unexpected as in some previous studies carbohydrates were reported to be the most reactive cell wall components of wood at the beginning of hydrothermal treatment (*21*, *44*). The substantial decrease in thioacidolysis yield indicated that about 33% of the noncondensed lignin structures disappeared within 3 h. This implied a formation of resistant bonds as no substantial loss of material occurred during this period. It can be seen that, along with the decrease in thioacidolysis yield, the S/G ratio after thioacidolysis was also decreased (**Table 2**), which meant that the degradation of β-O-4 linked structures specifically involved the S lignin units. The frequency of H monomers after thioacidolysis remained low (in the 0.5−1% range, data not shown in **Table 2**), whatever the sample. These changes in lignin structure were accompanied by a considerable decrease in wood rigidity (**Figure 3**). No further change in lignin structure was apparent after the linear decrease of the storage modulus (i.e., after 20 h of treatment). In contrast, a further slight reduction in noncellulosic fraction content was apparent, leading to an additional decrease of almost 20% of the rigidity. The composition of the noncellulosic polysaccharides was substantially changed by the treatment. After 20 h at 110 °C, recovery of the minor arabinose, galactose, and galacturonic acid fractions was decreased by 76, 34, and 28%, respectively (**Table 3**).

At 135 °C, during the first 4 h of treatment, the storage modulus of oak wood samples decreased by 44%. This decrease was associated with a considerable loss of noncellulosic polysaccharides (41% relative reduction, **Table 2**). In addition, the composition of the residual noncellulosic polysaccharides was dramatically modified, with almost 90% of the arabinose or galacturonic acid and 68% of the galactose disappearing while the main sugar components of the oak noncellulosic polysaccharides were altered to a lesser extent (35% and 25% decrease in xylose and mannose respectively) (**Table 3**). The lignins were moderately enriched, but displayed substantial structural alterations. The dramatic reduction of thioacidolysis yield (from 2600 to 1370 μmol/g of lignin) signified that 51% of the noncondensed lignin units had been degraded (**Table 2**). In accordance with the results obtained at 110 °C, the S/G ratio was greatly decreased by hydrothermal treatment. This specific degradation of S lignin units could be due either to the higher susceptibility of S units to hydrothermal treatment or to the fact that degradation was more extensive in the secondary cell walls which contain S-rich lignins (*45*, *46*).

Between 4 and 6 h of treatment, the proportion of β-O-4 linked lignin structures and the hemicellulose content decreased to a still greater extent, without any further change in the storage modulus. From this stage to the end of treatment, the storage modulus increased, surprisingly, to attain a value similar to that of the untreated samples (**Figure 3**). Because the frequency of the noncondensed units in lignins was reduced by 74% during this period, it is suggested that, in addition to β-O-4 cleavage, the lignins undergo severe condensation reactions that would lead to a highly rigid material (*17*). This latter point was confirmed by the viscoelasticity measurements that indicated a shift in the softening temperature of treated samples toward higher temperatures. The hydrothermal degradation of hemicelluloses tends to reduce the rigidity and increase the loss factor, whereas the condensation of lignins would reduce their mobility and increase the softening temperature. These two successive effects are clearly observed when a sample undergoes heating cycles up to 135 °C (**Figure 5**). The first cycle increases the loss factor but does not affect much the transition temperature, whereas the second cycle leads to a reduced loss factor but significantly increases the transition temperature. The third cycle has no significant additional effect. For each cycle, it is obvious that the time is only important at high temperature levels. This explains why the heating curve is almost identical to the previous cooling curve (curve 2 is similar to curve 3,

Table 3. Variation of Some Noncellulosic Polysaccharide Contents[a] during Various Hydrothermal Treatments of Oak and Spruce Species

wood species	T (°C)	time (h)	arabinose	galactose	galacturonic acid	xylose	mannose
oak	control		0.5 (0.0)	0.7 (0.0)	3.1 (0.1)	20.3 (0.8)	3.7 (0.3)
	110	3	0.3 (0.0)	0.6 (0.0)	3.1 (0.1)	22.0 (0.8)	3.4 (0.0)
		19	0.1 (0.0)	0.4 (0.0)	2.2 (0.1)	20.4 (0.6)	3.5 (0.1)
	135	2	0.1 (0.0)	0.3 (0.0)	1.8 (0.0)	15.9 (0.2)	3.0 (0.1)
		4	0.1 (0.0)	0.2 (0.0)	0.4 (0.0)	13.3 (0.1)	3.0 (0.0)
		6	0.1 (0.0)	0.1 (0.0)	0.3 (0.0)	11.7 (0.0)	2.9 (0.1)
		24	0.1 (0.0)	0.0 (0.0)	0.3 (0.0)	9.6 (0.2)	3.0 (0.3)
spruce	control		1.4 (0.0)	1.6 (0.1)	2.8 (0.1)	6.3 (0.2)	12.7 (0.4)
	110	3	1.4 (0.0)	1.6 (0.1)	2.7 (0.0)	6.3 (0.2)	13.2 (0.2)
		21	1.1 (0.0)	1.5 (0.0)	2.4 (0.2)	6.2 (0.0)	12.4 (0.2)
	135	2	0.5 (0.0)	1.6 (0.0)	1.0 (0.1)	5.6 (0.1)	11.9 (0.1)
		4	0.4 (0.0)	1.4 (0.0)	0.8 (0.0)	5.8 (0.1)	12.0 (0.1)
		6	0.2 (0.0)	1.3 (0.1)	0.7 (0.0)	5.3 (0.3)	11.4 (0.4)
		24	0.1 (0.0)	0.5 (0.0)	0.7 (0.1)	3.8 (0.2)	8.1 (0.1)

[a] Weight percent of the extract-free material.

and curve 4 is similar to curve 5). Olsson and Salmén (*47*) have previously shown that the more the lignin structure is cross-linked, the higher is the glass transition temperature of in situ lignin.

Interestingly, thioacidolysis revealed that thermal treatments in the 200−280 °C range (*24*, *48*) and the hydrothermal treatment studied in the present work had both similar and different effects on lignins. When performed under severe conditions (prolonged duration and/or high temperature), both treatments increased the lignin contents of the samples, due mainly to extensive loss of the hemicellulose fraction. In both cases, the extent of lignin structure alteration was dependent on treatment severity and essentially consisted of β-O-4 cleavage and condensation reactions. However, the lignin degradation mechanisms prevailing in dry conditions displayed some peculiarities. The thermal treatment of lignins, in both air and inert atmosphere, induced the formation of vinyl ether structures, as revealed by thioacidolysis (*24*, *48*). The formation of these resistant vinyl ether structures was associated with loss of the terminal hydroxymethyl groups of the lignin side chains, released as formaldehyde, which could efficiently contribute to condensation reactions (*17*). We did not find any substantial formation of vinyl ether structures under our hydrothermal treatments. It is therefore very likely that the condensation reactions associated with hydrothermal treatment were mainly due to acid-catalyzed condensations involving benzylic carbocations released from the hydrolytic cleavage of β-O-4 bonds.

Additional analyses were carried out to characterize the various resistant interunit linkages in lignins and the variations induced by the 24 h treatment at 135 °C (**Table 4**). The main lignin-derived thioacidolysis dimers recovered from hardwood lignins were representative of 5-5, 4-O-5, β-1, β-5, and syringaresinol (β-β) bonding patterns. Their relative frequencies (expressed as molar percentages of the total) were substantially altered by the severe hydrothermal treatment. The most dramatic change was the large relative decrease in β-1 dimers. This implied that the spirodienone or diarylpropane structures (F and G in **Figure 1**) were degraded to a greater extent than the other condensed lignin bonding patterns. This can be assigned to the fact that diarylpropane structures are terminal units with free phenolic groups, which could make them more susceptible to hydrothermal degradation. We also suggest the following hypothesis. The lignin fragments associated with water-soluble LCCs have been shown to be particularly enriched in β-1 linkages (*49*−*51*). As the treatment induces a massive loss of hemicellulose components, it is very likely that those lignin structures most closely associated with such components, namely, the lignin domains rich in β-1 bonding patterns, were preferentially lost. Analysis of the

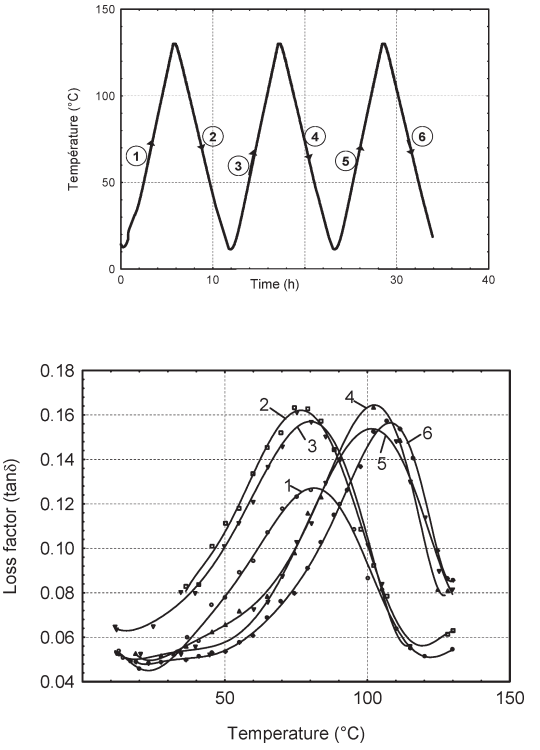

Figure 5. Dynamic mechanical analysis of oak sample subjected to heating/cooling cycles in water-saturated conditions (temperature, 10−135 °C; frequency of mechanic solicitations, 1 Hz, radial direction). Note the changes in the loss factor and particularly the positive shift of the softening temperature as the cycles progress.

lignin-derived dimers also revealed a substantial reduction of the β-β aryltetralin dimers derived from syringaresinol bonding patterns (**Table 4**). This relative reduction could be assigned to the selective loss of the syringaresinol structures incorporated into lignins solely by β-O-4 bonds, which were cleaved by the treatment. This hypothesis was further supported by the recent identification of syringaresinol in the soluble components formed after thermal treatment of beech wood (*22*). In contrast to these relative reductions in β-1 and syringaresinol bonding patterns, the hydrothermally treated oak lignins were substantially enriched in β-5 interunit bonds and, to a lesser extent, in 5-5 and 4-O-5 linkages (**Table 4**).

The hydrothermal treatment caused significant alterations in oak lignin structure. The key reaction was degradation of the

Table 4. Relative Frequency (Percent Molar) of the Lignin-Derived Dimers Recovered after Thioacidolysis and Raney Nickel Desulfurization of the Control and of the Hydrothermally Treated (24 h, 135°C) Oak Samples[a]

dimer type	control	treated
5-5	11.5 (0.2)	13.8 (0.4)
4-O-5	8.3 (0.0)	9.7 (0.6)
β-1	32.5 (0.2)	18.4 (0.3)
β-5	20.5 (0.5)	35.8 (2.3)
β-β in syringaresinols	27.2 (0.4)	22.3 (1.2)

[a] The data are mean values of duplicate analyses (standard errors in parentheses).

β-O-4 structures, which first led to a decrease of the wood storage modulus. Secondary condensation reactions, concomitant with β-O-4 cleavage, also occurred, and this reorganization, which increased with treatment severity, finally produced a more rigid material. In addition, noncellulosic polysaccharides were partially hydrolyzed and/or dissolved, which led to the selective loss of arabinose, galactose, and galacturonic acid units.

Spruce Wood Amorphous Polymers Displayed Moderate Susceptibility to Hydrothermal Treatment. Very little structural modification of spruce cell wall polymers was apparent with the hydrothermal treatment at 110 °C, whereas the storage modulus decreased by 27% (**Table 2**). The lower susceptibility of spruce lignins to moderate hydrothermal treatment can be assigned to their lower content of units solely involved in β-O-4 bonds (36% for native spruce lignins versus 66% for native oak lignins). The absolute content of noncellulosic polysaccharides did not vary to a pronounced extent (< 6%), but their composition was modified with a preferential loss of the minor arabinose and galacturonic acid units (proportion in the cell wall decreased by 22 and 14%, respectively) (**Table 3**). These discrete but nevertheless noticeable changes in the structure of the cell wall polysaccharides might partially account for the decrease in rigidity (27%). It is also very likely that the hydrothermal treatment altered the weak interactions between cell wall polymers, and this would not be apparent from conventional chemical analyses.

A 2 h treatment at 135 °C led to a slight loss of noncellulosic polysaccharides and a moderate degradation of the β-O-4 linkages (18 and 11%, respectively) of spruce lignin, which could be connected to the 19% decrease of the storage modulus. No further modification of lignin structure was observed between 2 and 6 h of treatment, although the storage modulus continued to decrease (to 27%). These changes were most likely due to the degradation of noncellulosic polysaccharides. The content of these latter remained relatively constant, whereas the composition was considerably modified. The galacturonic acid and arabinose contents decreased by 70 and 73%, respectively. Galacturonic acid is mainly associated with pectins in the middle lamella region (*43*). The observed changes in the viscoelastic properties of spruce wood could be due to the hydrothermally induced hydrolysis of the pectins and to their resulting molar mass reduction as well as to alterations in the interactive capabilities of the cell wall polymers. Beyond 6 h of treatment, and as observed in oak samples, the storage modulus rose to a value close to that of untreated samples. This was concomitant with a considerable loss of noncellulosic polysaccharides (50% reduction). Inversely, the lignin content and thioacidolysis yields remained stable, implying that no further reduction in the frequency of noncondensed structures occurred. Although the levels of lignin and of noncondensed lignin units remained constant, analysis of the thioacidolysis dimers recovered from spruce samples confirmed that the severe hydrothermal treatment (24 h, 135 °C) induced substantial changes in lignin structure. As in oak lignins, this treatment caused a dramatic reduction in the relative frequency of the β-1 interunit linkage (reduced from 32% in the control to 19% in the treated sample). Conversely, the relative frequencies of the 5-5, 4-O-5, and β-5 representatives were increased (data not shown).

Conclusion. The variations in the viscoelastic properties of the two hydrothermally treated woods were due not only to structural alterations of each type of cell wall polymer (i.e., lignins and noncellulosic polysaccharides) but also to changes in the interactions established between the various wall polymers. The analytical degradations used in the present study could not provide information about these interactions. Nevertheless, they revealed considerable changes in the amorphous polymers that could be linked to the variations in viscoelastic properties of these woods.

This study demonstrated that hardwood lignins were more sensitive to hydrothermal treatment than softwood lignins. The considerable decrease in wood rigidity seemed to be primarily due to degradation of the β-O-4 linkages, which required milder conditions than the degradation of noncellulosic polysaccharides, a result that contrasts with previous literature data (*20*, *43*). The most severe treatment (24 h at 135 °C) not only induced extensive degradation of β-O-4 structures but also selective loss of the β-1 structures known to be preferentially associated with hydrosoluble polysaccharides. The galacturonic acid and arabinose substituents of the noncellulosic polysaccharides seemed to be the most reactive sugar units during hydrothermal treatment. The various changes in the amorphous polymers led to substantial reorganization within the hydrothermally treated cell walls so that rigidity, which decreased at the beginning of treatment, was eventually almost fully restored to its initial value. Future efforts will focus on spectroscopic analyses as these should provide information about local changes in structure and interactions of the cell wall polymers.

ABBREVIATIONS USED

CWR, cell wall residue; E', storage modulus; E'', loss modulus; G, guaiacyl; H, *p*-hydroxyphenyl; LCCs, lignin−carbohydrate complexes; S; syringyl; tan δ, loss factor; WAVE^{T}, Environmental Vibration Analyzer for Wood.

ACKNOWLEDGMENT

We very sincerely thank Laurent Cézard (UMR 206, INRA) and Frédéric Legée (UMR 206, AgroParisTech) for running the lignin analyses.

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Received for review April 27, 2009. Revised manuscript received July 6, 2009. Accepted July 7, 2009.