The Study of Water Sorption with Hydrolysis Lignin by Solid-State NMR Spectroscopy

S.L. Shestakov*, Yu.A. Popova, A.Yu. Kozhevnikov, D.S. Kosyakov, S.A. Sypalov

Northern (Arctic) Federal University named after M.V. Lomonosov, 17 Severnaya Dvina Emb., Arkhangelsk, Russia

Article info

Abstract

Hydrolysis lignin is formed as a by-product of cellulose production and has limited industrial application. The ability of hydrolysis lignin to absorb and retain some water is an important aspect for the study of its properties and modification methods. The processes of water sorption by hydrolysis lignin were studied with solid-state NMR spectroscopy. The samples were humidified in desiccators containing different saturated salts solutions with different relative air humidity above them. The sorption capacity of the samples was determined by water sorbed from the air, and it was found that lignin absorbs the amount of water equal to 40% of sample weight at a maximum relative humidity of the air. The cross-polarization (CP) and magic angle spinning (MAS) methods were used to register solid-state NMR spectra. Using the ^{1}H-NMR spectra, it was found that the hydrolysis lignin is hydrated in the whole volume, and the water penetrates into the deep layers of polymer, however, the distribution of water at the likely sorption sites is uneven. It was obtained with the use of ^{13}C-NMR spectroscopy that hydrolysis lignin hydrates in both hydrophilic and hydrophobic regions of the macromolecule, and the bulk of sorbed water (~64%) concentrates around the hydroxyl and methoxyl groups of lignin and polysaccharide residues.

1. Introduction

Lignin is the second most abundant biopolymer in nature (after cellulose) and is a complex mixture of macromolecules based on phenylpropane structural units [1]. It can be considered as the most important renewable source of valuable aromatic compounds for chemical synthesis and raw material for the production of fuels, sorbents, and various composite materials. Despite this, the industry uses only a minor part of lignin, which is formed as a by-product of acid hydrolysis of plant feedstock with the production of bioethanol [2]. This is largely due to a lack of knowledge about the structure, properties, and reactivity of hydrolysis lignin (HL), which is a highly condensed polymer and, as a result, insoluble in organic solvents and alkalis.

The ability of HL to absorb and retain some water is an important aspect of the study of its properties and for the development of its modification methods. Water diffusing into a polymer can change its physical state (plasticization) and, under certain conditions, cause hydrolysis of the ether bonds between the structural units of macromolecules, which affects the mechanical and chemical properties of the materials obtained on the basis of HL. The study of the water sorption processes in lignin and, in particular, the identification of priority sorption centers in macromolecules is an urgent task.

The solution to this problem is possible with the use of nuclear magnetic resonance spectroscopy (NMR), which is one of the most informative method for studying the lignin structure and functional composition [3–7]. This method is highly accurate and reliable, however, studies of hydrolysis lignin are difficult, because hydrolysis lignin is insoluble in the deuterated solvents. The solution to the problem is the use of solid-state NMR spectroscopy [8, 9] using cross-polarization and rotation at a magic angle (Magic Angle Spinning, MAS) to increase the signal-to-noise ratio and improve the spectral resolution [10].

*Corresponding author. E-mail: laston85@mail.ru

Solid-state NMR spectroscopy is successfully used to analyze the state and dynamics of water molecules in natural and synthetic objects. With its help, the existence of several forms of water in polymers (free, bound, non-freezing, etc.) was established [11, 12]. The basis of this approach is the ability of water molecules to exchange protons at moisture saturation, leading to a change in the position of the signals of the ^{1}H-NMR spectra, which depends on the composition and mobility of proton complexes [12, 13]. The shape and width of the spectral bands are also associated with the mobility of nuclei, an increase in which leads to a narrowing of the corresponding peaks in the spectra, which allows conclusions to be drawn about the mobility of functional groups and the polymer matrix of various compounds. Examples of using this method to study the hydration mechanism of such polymers like polyvinyl alcohol [13], sulfonated polyetherketones [14], perfluorinated polymers like Nafion™ and Flemion™ [14–16], polybenzimidazoles [17, 18], etc. are known in the literature.

The purpose of this research is to study the processes of water sorption by hydrolytic lignin by solid-state NMR spectroscopy.

2. Experimental part

As a research object, softwood hydrolysis lignin was used, obtained at the Kirov Biochemical Plant (Kirov, Russia) and containing 65.2% lignin (according to Klason), 14.3% of extractive substances and 12.5% of residual polysaccharides, the ash content was 7.6%. A detailed description of the structural units of macromolecules of this lignin after alkaline solvolysis is presented in [19].

2.1. Reagents and materials

Chemically pure calcium chloride, sodium bromide, sodium chloride (Vekton, St. Petersburg, Russia) and chemically pure sodium hydrogen phosphate (Neva-reaktiv, St. Petersburg, Russia) were used to create the required moisture levels. Deionized water from the Milli-Q system (Millipore, France) was used for the preparation of solutions.

2.2. Sample preparation

The sample of lignin is thoroughly crushed in an agate mortar and dried in a vacuum oven at 60 °C to constant weight. It was observed in pre-liminary experiments that raising the temperature to 90 °C did not lead to an additional loss of sample weight. Higher temperatures were not used for drying due to the possibility of undesirable changes in the structure of the biopolymer. Five weighed dry lignin samples ~ 50 mg each) were placed into desiccators with saturated salt solutions to maintain a fixed relative air humidity (RH) [20]: Na_2HPO_4 (RH = 95%), NaCl (RH = 75%), NaBr (RH = 58%), $CaCl_2$ (RH = 32%). Pure deionized water was used to create a relative humidity of 100%, and initial lignin dehydrated in the oven was used as an absolutely dry sample (RH = 0%).

Samples of hydrated lignin with the determined water content were obtained by keeping the samples in desiccators at the room temperature (20±2 °C) until the reach of constant weight (for several days). The amount of moisture absorbed by lignin was determined by the gravimetric method.

2.3. Registration of NMR spectra

The ^{1}H and ^{13}C NMR spectra were registered with NMR spectrometer Bruker AVANCE III 600 MHz (Germany). Samples with determined water content were placed in a ceramic rotor with an outer diameter of 3.2 mm. Magic angle spinning (MAS) with a frequency of 7 kHz was used in order to improve the resolution of spectra. Preliminary registration of the test spectrum with MAS frequency equal to 10 kHz showed that the increase of MAS frequency does not affect the spectrum.

^{1}H-NMR spectra were registered using a standard one-pulse sequence. To improve the signal-to-noise ratio and reduce the acquisition time of the ^{13}C-NMR spectra, we used the cross-polarization combined with magic angle spinning (CP/MAS). The parameters of the experiments are represented in Table 1.

Table 1
Parameters of solid-state NMR experiments

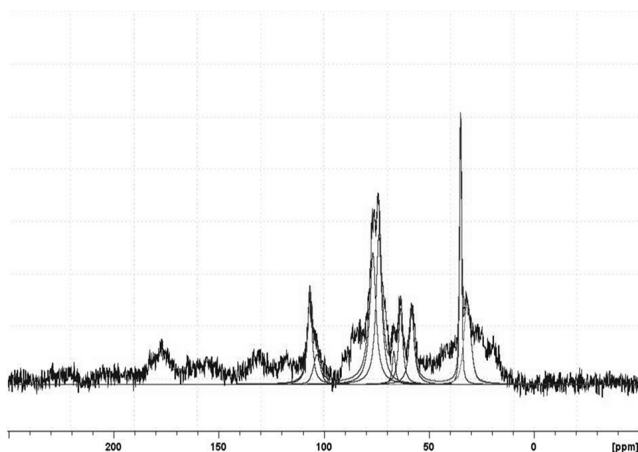| Parameter	^{1}H-NMR	^{13}C-NMR
Pulse width, µs	2.5	2000
Acquisition time, s	0.043	0.022
Delay, s	3	5
Spectral width, ppm	20	300
Number of scans	4	4096
MAS rate, kHz	7	7

Fig. 1. Partial deconvolution of ^{13}C-NMR spectrum of hydrolysis lignin sample prepared at RH = 100%.

2.4. Spectra processing

Solid-state NMR spectra were processed in standard NMR spectrometer software "TopSpin" (Bruker, Germany). Spectra were deconvoluted (Fig. 1), i.e., the chemical shift, intensity, width and shape of the spectral line were picked for the selected signals. The parameters were changing until the spectral line calculated by the program coincided as much as possible with the contour of the recorded signal.

3. Results and discussion

3.1. ^{1}H NMR spectra of lignin

The most intense signal in the ^{1}H-NMR spectra of the studied lignin samples is observed in the region of 5–10 ppm (Fig. 2). The chemical shift, width, and relative intensity of the selected signal
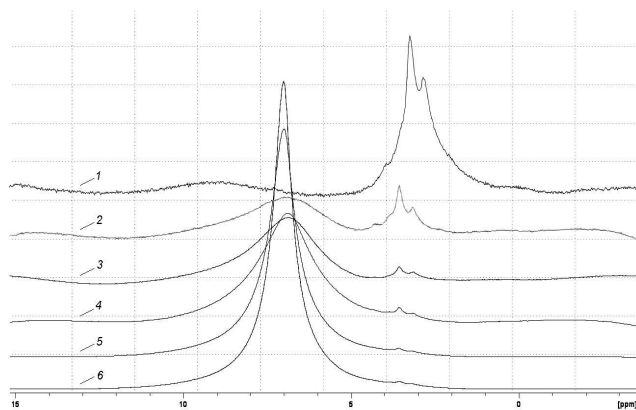
Table 2
Sorption capacity of lignin samples in relation
to water sorbed from air

RH, %	100	95	75	58	32
Sample weight, mg	49.56	50.37	50.05	49.93	50.30
Absorbed water weight, mg	19.72	15.94	7.40	5.27	3.71
Sorption capacity of 1 mg of lignin, mg	0.397	0.316	0.148	0.106	0.0074

vary with the moisture content of the sample. The set of signals is observed between 2 and 4 ppm, their chemical shift also slightly varies depending on the moisture content of the sample.

It is known that the chemical shift of water protons is about 4.7 ppm [21], and the one for isolated proton is about 12–15 ppm [12]. The ^{1}H-NMR signal is the weighted average sum of all protons signals of the sample due to fast proton exchange. The position of the ^{1}H-NMR signal is determined by the sorption capacity of the polymer relative to the water sorbed from the air at a determined relative humidity. The values of the sorption capacity are presented in Table 2.
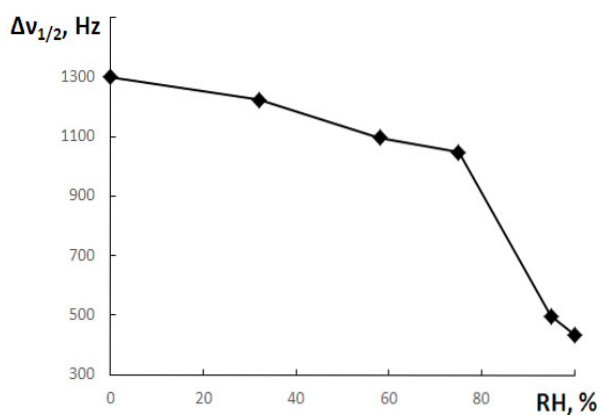
The width of the NMR signal $\Delta v_{1/2}$ is determined by the transverse relaxation time T_2 and is related to it by

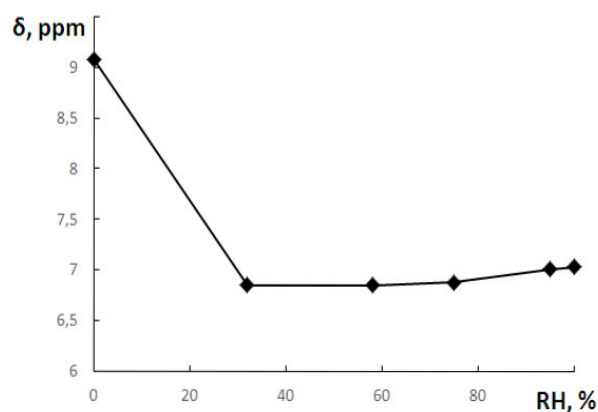$$\Delta v_{1/2} \sim \frac{1}{\pi T_2}$$

therefore, a decrease of the signal width indicates the increase in the mobility of a corresponding group of atoms, and vice versa [22].

The dependences of the width and chemical shift of the most intense ^{1}H-NMR signal on moisture content are presented in Fig. 3.

According to Fig. 3a, the proton mobility increases with increasing moisture content, abruptly increasing at high values of relative humidity (95 and 100%). The change in mobility with an increase of moisture content can be explained by the expansion of free space between the polymer chains of lignin during swelling, and the abrupt increase in mobility at high relative humidity can be interpreted as the water penetration into deeper structures of the natural polymer, which are inaccessible at low moisture contents. The assumption is confirmed by a sharp increase in the sorption capacity of the samples at high moisture contents (Table 2).

Fig. 2. ^{1}H-NMR spectra of the hydrolysis lignin samples prepared at RH: 1 − 0%; 2 − 32%; 3 − 58%; 4 − 75%; 5 − 95%; 6 − 100%.

(a)

(b)

Fig. 3. The dependencies of width (a) and chemical shift (b) of the most intensive peak in ^{1}H-NMR spectrum on water content.

The components of the considered ^{1}H-NMR signal in an absolutely dry sample are mostly signals of hydroxyl and carboxyl protons [23], which explains the position of the signal in the region of ~9 ppm. (Fig. 3b). A significant decrease in the chemical shift of the ^{1}H-NMR signal with increasing moisture content shows the appearance of partially bound and free water in the sample, which is involved in proton exchange. At high moisture contents (corresponding to RH = 95% and RH = 100%), an increase in chemical shift is observed, which can also be explained by the fact that under these conditions water penetrates into the deep layers of natural polymer particles, with the result that the proportion of bound water slightly increases. The reason for this process may be the competition between physical and chemical sorption.

We can conclude that the entire sample volume is hydrated during moisture saturation, and the majority of protons are involved in exchange processes due to one powerful signal is observed in the ^{1}H-NMR spectra of hydrolysis lignin, which is the weighted average sum of the signals of most protons. Naturally, with increasing moisture content, the rate of proton exchange increases, according to the dependence of the signal width on moisture content (Fig. 3a). Because of this, the mechanism of hydration and, particularly the distribution of water sorption sites in the lignin seems to be interesting.

3.2. ^{13}C-NMR spectra of lignin

After registration of the ^{13}C NMR spectra (Fig. 4) of the samples, it was observed that the structure of the obtained spectra is similar to the structure of the ^{13}C NMR spectra presented in [24]. Thus, considering the content of residual polysaccharides in the samples, it can be assumed that they play a significant role in the processes of water sorption.

Figure 5 shows the structure of a cellulose unit as a polysaccharide, predominantly present in the technical lignin. With the further assignment of spectrum bands, the presence of polysaccharides was taken into account.

Despite the complex composition of the sample, in the ^{13}C-NMR spectra, it is possible to trace the dependence of the parameters of some signals on the moisture content. When the relative humidity changes, the chemical shifts of the ^{13}C-NMR signals remain almost constant, but the resolution of the spectra improves as the moisture content increases, which means that the width of the signals changes. Thus, the mobility of groups of atoms corresponding to the selected signals also changes. Figure 6 shows the dependence of the linewidth of some ^{13}C-NMR signals on the moisture content of the samples.
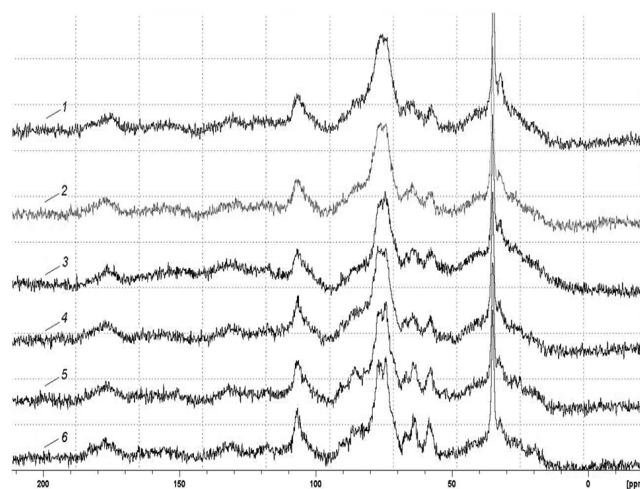

Fig. 4. ^{13}C-NMR spectra of the hydrolysis lignin samples prepared at RH: 1 – 0%; 2 – 32%; 3 – 58%; 4 – 75%; 5 – 95%; 6 – 100%.
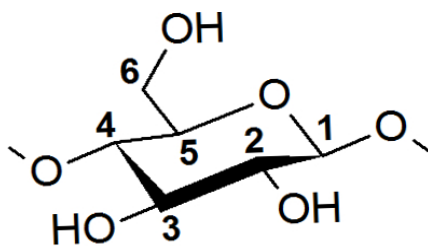
Fig. 5. Structure of the cellulose monomer.

Based on the tables of chemical shifts [23] and assignment of spectrum bands [24], it was established that the observed signals correspond to the following atomic groups:

I) protonated carbon in the aromatic ring and the C-1 atom (Fig. 5) in the cellulose unit (imposition of signals with a chemical shift of ~ 106.7 ppm in the spectrum of ^{13}C-NMR);

II) ^{13}C-NMR spectrum signals in the region of 76.7 ppm and ~ 73.9 ppm presumably correspond to C-2, C-3 and C-5 atoms in the cellulose unit (Fig. 5), as well as to carbon atoms in the aliphatic chains of lignin associated with hydroxyl groups;

III) chemical shift signal ~ 66.6 ppm in the ^{13}C-NMR spectrum is assumed to correspond to the C-6 atom in the cellulose unit (Fig. 5);

IV) methoxyl groups on the aromatic ring (a signal with a chemical shift of 63.5 ppm);

V) methoxyl groups CH3 –O– in aliphatic chains of lignin (a signal with chemical shift 58.3 ppm).

VI) signal in the region of ~31.8 ppm presumably corresponds to the methyl acetate group in hemicellulose residues.

The presence of all these fragments in the structure of lignin is confirmed by literature data [1, 2, 24].

The correspondence of signals I and IV to the designated groups is indirectly confirmed by an analysis of the dependence of the band widths on moisture content (Fig. 6). With a moisture content of the sample corresponding to a relative humidity of 75%, groups I and IV reach the maximum of their mobility, which indicates the formation of a hydration shell around the entire aromatic group. In this case, the width of the band corresponding to the methoxyl group changes monotonically until the specified moisture content is reached, and the change in the width of the signal corresponding to the carbon atom in the aromatic ring is abrupt. Consequently, the methoxyl group is first hydrated, and then the entire aromatic ring, which is consistent with known information about the structure of lignin. It can be assumed that the width of the

band corresponding to the C-1 atom (Fig. 5) changes similarly to the band width of the carbon atoms of the aromatic ring since the width of their sum monotonously decreases with increasing moisture content. An increase in the mobility of the C-1 atom may be due to the formation of a hydrogen bond between the water molecule and the oxygen atom.

The decrease in the width of peaks II, depending on the moisture content, is probably due to the formation of hydrogen bonds between water molecules and hydroxyl groups, which leads to an increase in their mobility (Fig. 6). This assumption may explain the absence of limit value of mobility even with the maximum possible moisture saturation of the sample (corresponding to RH = 100%).

The correspondence of signal III to carbon atoms C-6 is confirmed by the smallest initial peak width and the dynamics of its change depending on moisture content. A monotonous increase in the mobility of the group over the entire range of moisture content may be associated with hydration of the terminal hydroxyl group (Fig. 6).

The nature of the change in the peak width of the signal V indicates that the mobility of the corresponding groups almost does not change with an increase in moisture content, which is probably due to steric hindrance.

The width of the signal corresponding to group VI increases with increasing moisture content, which may be due to the dimerization of carboxyl groups and the formation of hydrogen bonds that limit the movement of the terminal methyl groups. This assumption is indirectly confirmed by the fact that the peak width of the signal corresponding to carboxyl groups (~ 175 ppm) does not change with increasing moisture content.
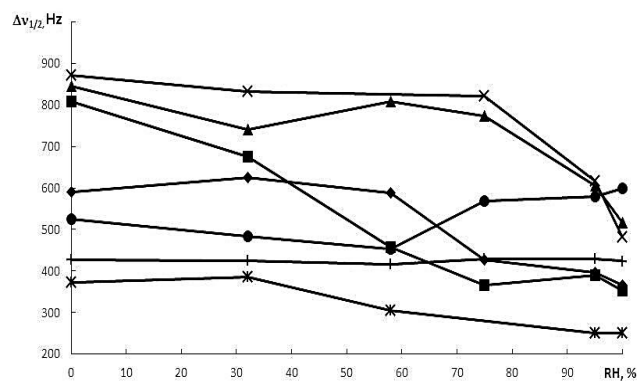

Fig. 6. The dependencies of band widths in the ^{13}C-NMR spectra on relative humidity with chemical shifts 106.7 ppm (•), 76.7 ppm (▲), 73.9 ppm (×), 66.6 ppm (*), 63.5 ppm (■), 58.3 ppm (+), 31.8 ppm (●).

Table 3

$\Delta v_{1/2}$ (Hz) band widths in the ^{13}C-NMR spectra of hydrolyzed lignin with different moisture content (RH)

Groups / RH,%	-CH-(aryl.), C-1 (δ=106.7 ppm)	C-2, C-3, C-5, C-OH (aliph.) (δ=76.75 ppm)	C-2, C-3, C-5, C-OH (aliph.) (δ=73.9 ppm)	C-6 (δ=66.6 ppm)	-OCH3 (aryl.) (δ=63.5 ppm)	-OCH3 (aliph.) (δ=58.3 ppm)	CH3COO- (hemicell.) (δ=31.8 ppm)
0	589.8	845.3	872.0	372.0	807.2	427.7	525.4
100	366.5	516.7	482.2	250.7	353.5	423.8	599.1
Width change multiplicity	1.609	1.636	1.808	1.484	2.283	1.009	0.877

Table 3 shows the relative changes in the peak width of the selected signals when the moisture content varies from 0 to 100%.

Based on the relative changes in the peak width of the selected signals, we can conclude that the sample fragments are not uniformly hydrated, and some groups absorb water more actively than others. To obtain, in the first approximation, a semi-quantitative picture of the distribution of water in a sample, it is possible to consider only those signals whose width decreases with increasing sample moisture content.

Assuming that all sorbed water accumulates around these functional groups, it is possible to calculate the relative shares of the total amount of water per type of group. For this, the relative changes in the peak widths were normalized to the smallest value, the results were summed to give the total number of parts by weight of sorbed water. Then, the proportion of water molecules per group type was determined. The results of the calculations are presented in Table 4.

According to calculations, the largest amount of water is accumulated around the methoxyl and hydroxyl groups of lignin and polysaccharides. Since the mobility of atoms that are not associated with these groups also increases with increasing moisture content, it is assumed that hydrophobic fragments of macromolecules are hydrated to a certain extent.

4. Conclusion

We proposed the approach to the study of water binding by hydrolysis lignin, based on the use of solid-state NMR spectroscopy. It has been established that technical hydrolysis lignin can sorb up to 40% of water by mass from the gas phase. Hydration of high-molecular compound proceeds through both the hydrophilic and hydrophobic regions of the macromolecules, with the bulk of the adsorbed water (~ 64%) associated with the hydroxyl and methoxyl groups of lignin and polysaccharide residues.

Acknowledgments

Table 4

Mass fractions of water sorbed by various functional groups of hydrolysis lignin macromolecules

Groups	-CH-(aryl.), C-1 (δ = 106.7 ppm)	C-2, C-3, C-5, C-OH (aliph.) (δ = 76.75 ppm)	C-2, C-3, C-5, C-OH (aliph.) (δ = 73.9 ppm)	C-6 (δ = 66.6 ppm)	-OCH3 (aryl.) (δ = 63.5 ppm)
Width change multiplicity	1.609	1.636	1.808	1.484	2.283
Amount of weight parts	1.08	1.10	1.22	1.00	1.51
Water mass fraction, %	18.24	18.55	20.50	16.82	25.89

References

[1]. C. Heitner, D.R. Dimmel, J.A. Schmidt. Lignin and lignans: advances in chemistry (Ed. by C. Heitner). Boca Raton: CRC Press, 2010. 683 p.

[2]. K.G. Bogolitsyn, V.V. Lunin, D.S. Kosyakov, A.P. Karmanov, T.E. Skrebets, N.R. Popova, A.V. Malkov, N.S. Gorbova, A.N. Pryahin, A.N. Shkaev. Fizicheskaya khimiya lignina [Physical chemistry of lignin]. Ed. by K.G. Bogolitsyn, V.V. Lunin. Arkhangelsk: Arkhangelsk State Technical University, 2009. 489 p. (in Russian).

[3]. M. Balakshin, E. Capanema, *J. Wood Chem. Technol.* 35 (2015) 220–237. DOI: 10.1080/02773813.2014.928328

[4]. I.F. Fiţigău, F. Peter, C.G. Boeriu, *International Journal of Chemical, Molecular, Nuclear, Materials and Metallurgical Engineering* 7 (2013) 167–172.

[5]. L.V. Kanitskaya, A.F. Gorotov, D.T.T. Khai, A.V. Rokhin, *Russ. J. Bioorg. Chem.* 38 (2012) 720–725. DOI: 10.1134/S1068162012070102

[6]. Y. Pu, S. Cao, A.J. Ragauskas, *Energy Environ. Sci*. 4 (2011) 3154–3166. DOI: 10.1039/C1EE01201K

[7]. S.L. Shestakov, D.S. Kosiakov, A.Yu. Kozhevnikov, N.V. Ul'ianovskii, Yu.A. Popova, *Chemistry of plant raw material* [Khimija Rastitel'nogo Syr'ja] 2. (2017) 81–88 (in Russian). DOI: 10.14258/jcprm.2017021641

[8]. N. Terashima, J. Hafrén, U. Westermark, D.L. VanderHart, *Holzforschung* 56 (2002) 43–50. DOI: 10.1515/HF.2002.008

[9]. L. Fu, S.A. McCallum, J. Miao, C. Hart, G.J. Tudryn, F. Zhang, R.J. Linhardt, *Fuel* 141 (2015) 39–45. DOI: 10.1016/j.fuel.2014.10.039

[10]. M.J. Duer, Solid-State NMR Spectroscopy Principles and Applications, 2002, Blackwell Science Ltd. DOI: 10.1002/9780470999394

[11]. G.L. Brown, *Water in polymers*, Chapter 26, 441–450. ACS Symposium Series 127 (1980). DOI: 10.1021/bk-1980-0127.ch026

[12]. V.I. Volkov, E.A. Sidorenkova, S.F. Timashev, S.G. Lakeev, *Zhurnal fizicheskoi khimii* [Russian Journal of Physical Chemistry A] 67 (1993) 1014–1018 (in Russian).

[13]. V.I. Volkov, A.I. Rebrov, E.A. Sanginov, E.M. Anokhin, S.L. Shestakov, A.A. Pavlov, A.V. Maksimychev, Yu.A. Dobrovolskii, *Russ. J. Electrochem.* 45 (2009) 374–381. DOI: 10.1134/S102319350904003X

[14]. G. Ye, N. Janzen, G.R. Goward, *Macromolecules* 39 (2006) 3283–3290. DOI: 10.1021/ma0523825

[15]. M. Takasaki, K. Kimura, K. Kawaguchi, A. Abe, G. Katagiri, *Macromolecules* 38 (2005) 6031–6037. DOI: 10.1021/ma047970h

[16]. S. Tsushima, K. Teranishi, S. Hirai, *Energy* 30 (2005) 235–245. DOI: 10.1016/j.energy.2004.04.013

[17]. S. Kang, C.J. Zhang, G.Y. Xiao, D. Yan, G. Sun, *J. Membr. Sci.* 334 (2009) 91–100. DOI: 10.1016/j.memsci.2009.02.021

[18]. J.W. Traer, J.F. Britten, G.R. Goward, *J. Phys. Chem. B*. 111 (2007) 5602–5609. DOI: 10.1021/jp071471b

[19]. D.S. Kosyakov, E.V. Ipatova, S.M. Krutov, N.V. Ul'yanovskii, I.I. Pikovskoi, *J. Anal. Chem.* 72 (2017) 1396–1403. DOI: 10.1134/S1061934817140064

[20]. S.L. Shestakov, A.A. Pavlov, A.V. Maksimychev, A.V. Chernyak, V.I. Volkov, S.V. Timofeev, *Russ. J. Phys. Chem. B* [Original: Khimicheskaya Fizika 29 (2010) 60–68] 4 (2010) 1005–1013. DOI: 10.1134/S1990793110060199

[21]. E.G. Hugo, V. Kotlyar, A. Nudelman, *J. Org. Chem.* 62 (1997) 7512–7515. DOI: 10.1021/jo971176v

[22]. Blümich B. Essential NMR. For scientists and engineers. – M.: Technosphera, 2011. 256 p. (in Russian).

[23]. E. Pretsch, P. Bühlmann, C. Affolter. Structure Determination of Organic Compounds. Berlin: Springer-Verlag, 2000. 436 p. DOI: 10.1007/978-3-662-04201-4

[24]. S. Lequin, D. Chassagne, T. Karbowiak, R. Gougeon, L. Brachais, J.-P. Bellat, *J. Agric. Food Chem.* 58 (2010) 3438–3445. DOI: 10.1021/jf9039364