

The Isolation and Characterization of Lignin of Kenaf Fiber

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ABSTRACT: In this article, milled wood lignin (MWL) was isolated and purified from retted kenaf fiber, the lignin obtained was characterized by elemental analysis, FTIR, ¹H-NMR, and ¹³C-NMR spectroscopy. The C₉ formula is calculated for kenaf fiber MWL as C₉H_{9.32}O_{3.69}(OCH₃)_{1.30}. The spectra of FTIR, ¹H-NMR, and ¹³C-NMR show the kenaf fiber lignin to be of the G/S type with high proportion of syringyl (S) unit. The numbers of phenolic and aliphatic hydroxyl groups in the kenaf fiber MWL are estimated to be 0.14 and 1.31, respectively, per C₉ unit. The OH_{aliph} is 90.3%

in total numbers of hydroxyl groups of kenaf fiber MWL, and the OH_{ph} is 9.7%. It is evident that the β-O-4 structures are mainly linkage in the MWL of kenaf fiber, which contain more *erythro* stereochemistry type in β-O-4 units than *threo* stereochemistry type. In general, the characteristics of lignin of kenaf fiber are similar to that of hardwood. © 2009 Wiley Periodicals, Inc. *J Appl Polym Sci* 114: 1896–1901, 2009

Key words: kenaf; fiber; milled wood lignin; FTIR; ¹H-NMR; ¹³C-NMR

INTRODUCTION

Kenaf is a renewable, cheap, and easily grown annual plant.¹ Traditional products produced from kenaf are low-grade products such as animal bedding, packing bags, and ropes. However, the chemical treated fibers of kenaf have been shown to meet the quality requirements of textile fibers using for high-grade textile products.²

Kenaf fiber is a naturally existing polymeric composite composed of cellulose, pectin, hemicellulose, lignin, and some other extractives.³ All fiber constituents are responsible for most of the physical and chemical properties exhibited by fiber products. Because of its high degree of polymerization and crystallinity, cellulose is responsible for strength in fiber. Pectin is considered to be binder and is rich in the primary wall and middle lamella. Hemicellulose acts as a matrix for the cellulose and acts as a link between the fibrous cellulose and amorphous lignin. Lignin, a polyphenolic compound, acts as a cementing material for fibers.⁴

Retting is the freeing of the bast fibers from non-fiber tissues in the stem. The key step in the production of getting finer and soft fibers from kenaf fiber is degumming, which is the removing of noncellulosic substances from the fiber. Noncellulosic substances, such as hemicellulose, pectin, and lignin, affect the textile processes of spinning, weaving, dyeing, and finishing.⁵ The lignin content of retted kenaf fiber still reaches about 15% or more, which is the main objective component removed in degumming process.³

Lignins from different botanical origins are usually classified according to the abundance of their basic units with guaiacylpropane (G), syringylpropane (S), and 4-hydroxyphenylpropane (H).⁶ The characteristics of lignin obtained depend on both the kind of origins and the type and intensity of the delignification process. The basic phenylpropanes of softwood lignins are almost exclusively guaiacyl units, hardwood lignins contain guaiacyl and syringyl units.⁷

Lignin composition is an important parameter that affects fiber production. In general, the efficiency of production is directly proportional to the amount of syringyl (S) units in lignin. The G units have a free C-5 position available for carbon-carbon interunit bonds, which make them fairly resistant to lignin depolymerization, whereas the S lignin is relatively unbranched and has a lower condensation degree and therefore is easier to delignify.⁸ Therefore, an understanding of the chemical composition of kenaf fiber lignin gives insight into the preparation to get finer

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fibers and application of kenaf fiber as material for clothing and other high-grade products.

The structure of the kenaf's lignin has been previously investigated.^{1,9-15} Ralph⁹ suggested that kenaf is possibly unique in the type and amount of syringyl lignin present. Seca et al.¹⁰ isolated the dioxane and milled wood lignins (MWLs) from the core and dioxane lignin from the bark of kenaf, which showed that large differences in chemical composition exist between *in situ* and isolated lignins of kenaf. That is, both the core and bast *in situ* lignins are rich in the guaiacyl lignin units, whereas the lignins isolated from the core and bast are rich in the syringyl lignin units. Mazumder et al.¹⁴ analyzed the hand-separated bast fibers by pyrolysis-GC, which showed that mature bast lignin has high S/G value. This work reports the isolation of MWL from retted kenaf fiber of China, the MWL lignin was measured by elemental analysis, FTIR, ¹H-NMR, and ¹³C-NMR spectroscopy to get the C₉ formula of lignin and figure out the structure characterization. The results are expected to be helpful for further application of kenaf fiber as materials of higher grade products in textile field.

METHODS AND MATERIALS

Materials

Kenaf fibers used in this work were supplied by CZJXSI (China Zhejiang Xiaoshan Institute of Cotton and Bast Fiber Crops) and came in straight long fibers. The fibers had been separated from their stalks by water retting in CZJXSI. After the water retting process, the fibers were then cleaned with water and dried under the sunlight before they were delivered to be used in this research. The chemical composition of kenaf fiber was tested according to the GB 5889-86 methods (National standard constituent of bast fiber of China), the result is shown in the Table I, and the fineness of retted kenaf fiber is 109.8 Nm.

Retted kenaf fibers were cut into small pieces and then extracted by benzene and ethanol (V 2 : 1) in a Soxhlet for 6 h, the resultant materials were ground in a planetary ball milling to pass through a 200-mesh sieve.

TABLE I
Chemical Composition of Kenaf Fiber

Composition	Percentage based on dry material (%)
Cellulose	60.8
Hemicellulose	12.65
Lignin	17.82
Pectin	5.07
Water-soluble matter	3.08
Wax	0.58

Extraction of milled wood lignin

Following Björkman,¹⁶ milled fiber meal was soaked in dioxane/water (V 9 : 1) overnight with stirring. The solution was filtered and filtrate was evaporated to dryness under reduced pressure to obtain the coarse MWL. The purification of MWL was applied according to Lundquist.¹⁷ The residue was dispersed in pyridinylamino, acetic acid, and water (V 9 : 1 : 4). Then chloroform was added, the solution was transferred to separatory funnel in which solution was separated into two layers: the bottom layer was chloroform layer and the top layer was brown sediment. In this research, 15 times volume rate of chloroform and pyridinylamino were added to isolate the top layer sediment for three times to collect the chloroform liquid. The collected chloroform liquid was evaporated by rotating evaporimeter at 40°C and vacuum condition. More than 15 times volume of ether was put into the residual liquid, much brown sediment was formed. The sediment and solvent were separated by centrifugation. The steps were repeated till there was no smell of pyridinylamino in the sediment. The brown powder, which was purified MWL, was dried *in vacuo* over P₂O₅.

Acetylation of MWL

The MWL was dissolved in dry pyridine-acetic anhydride.¹⁸ After 72 h of standing in the dark, the solution was poured into cool water (pH = 2) whereupon the acetylated sample was recovered as a precipitate which was dried *in vacuo* over P₂O₅.

Elemental analysis

The element analyses were carried out on an Elemental Analyzer Model Vario EL III. The methoxyl content in lignin was obtained by ¹H-NMR.

Fourier transform infrared spectroscopy

FTIR spectrum of lignin was recorded in KBr discs by use of a Thermo Electron Corporation Nicolet 380 FTIR spectrophotometer.

¹H-NMR

Samples of solutions of 10 mg of acetylated lignin in 0.5 mL of in CDCl₃ were run at 400 MHz on a Bruker AV-400 spectrometer with tetramethylsilane (TMS) as an internal standard. Normalized peak areas were estimated by integration.

¹³C-NMR

The solution of ¹³C-NMR spectrum was obtained on a Bruker AV-400 operating with TMS as an internal

TABLE II
Analytical Compositions, C₉ Formulas

Lignin	Elementary composition (%)					C ₉ formulas
	C	N	H	O	OCH ₃	
MWL of kenaf fiber	56.20	1.013	6.044	36.27	18.31	C ₉ H _{9.32} O _{3.69} (OCH ₃) _{1.30}

standard at 100.6 MHz under total proton decoupled conditions. It was recorded at 25°C from 200 mg acetylated lignin dissolved in 1.0 mL DMSO-*d*₆ after 50,000 scans.

RESULTS AND DISCUSSION

Elemental analysis and C₉ formula

The average C₉ formula was calculated¹⁹ from the elemental analysis and methoxyl content. The results are present in Table II.

FTIR spectroscopy

To elucidate the structure of MWL isolated from kenaf fiber, FTIR spectrum is recorded. The absorption bands can be assigned empirically to structural groups, based on a multitude of results obtained both from model compounds and lignins.^{4,6,20,21} The assignment is given in Figure 1 and Table III.

The spectrum of kenaf fiber MWL shows hydrogen bonded O—H stretching absorption around 3400 cm⁻¹ and a prominent C—H stretching absorption around 2900 cm⁻¹. In the fingerprint region, between 1800 and 900 cm⁻¹, many sharp and discrete absorption bands because of various functional groups present in kenaf fiber MWL constituents are observed. The absorption at 1738 cm⁻¹ is assigned to the C=O stretching in unconjugated ketones and carbonyl group. Ketone carbonyls conjugated with aromatic rings, substituted in para positions produce a weak absorption¹⁰ at 1667 cm⁻¹. Aromatic skeleton vibrations in lignin are assigned²⁰ at 1593, 1503, and 1422 cm⁻¹; and the C—H deformations (asymmetric

in methyl, methylene, and methoxyl group) appear at 1462 cm⁻¹.²²

The spectrum also shows similar botanical origin to the corresponding lignin. The IR absorption peak intensity of hardwoods is²³ as follows: $I(1455-1465\text{ cm}^{-1}) > I(1500-1515\text{ cm}^{-1})$, $I(1220-1230\text{ cm}^{-1}) > I(1266-1270\text{ cm}^{-1})$, and $I(1121-1125\text{ cm}^{-1}) > I(1030-1040\text{ cm}^{-1})$. The IR absorption peak intensity of softwoods is as follows: $I(1500-1515\text{ cm}^{-1}) > I(1455-1465\text{ cm}^{-1})$, $I(1266-1270\text{ cm}^{-1}) > I(1220-1230\text{ cm}^{-1})$, and $I(1030-1035\text{ cm}^{-1}) > I(1135-1140\text{ cm}^{-1})$. The IR absorption peak intensity of kenaf fiber MWL is as follows: $I(1455-1465\text{ cm}^{-1}) > I(1500-1515\text{ cm}^{-1})$,

TABLE III
FTIR Absorption Peak Location and Assignment of Kenaf Fiber MWL

Wavenumbers/cm ⁻¹	Assignment
3498	O—H stretching in hydroxyl groups
2940	C—H stretching in methyl and methylene groups
2842	—CH ₂ — symmetry stretching in methyl and methylene groups
1738	C=O stretching in unconjugated ketone, carbonyl, and ester groups
1667	C=O stretching in conjugated ketone p-subst. Aryl ketones
1593	Aromatic skeleton vibrations plus C=O stretching; $S > G$; $G_{\text{condensed}} > G_{\text{etherified}}$
1503	Stretching of aromatic skeleton; $G > S$
1462	C—H deformations(asymmetry in methyl group —CH ₃ — and —CH ₂ —)
1422	Aromatic skeletal vibrations combined with C—H in plane deformations
1368	Aliphatic C—H stretching in methyl and phenol OH
1329	S unit plus G unit condensed (G unit bound via position 5)
1229	C—C plus C—O plus C=O stretch; $G_{\text{condensed}} > G_{\text{etherified}}$
1125	Typical of S unit; also secondary alcohol and C=O strct
1040	C—O of primary alcohol, Guaiacyl C—H
836	C—H out of plane in positions 2 and 6 (S units)

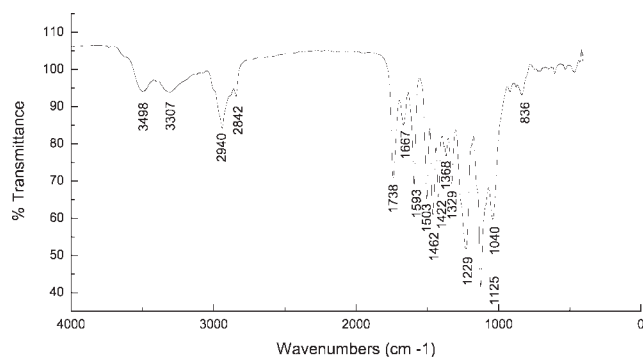


Figure 1 FTIR spectrum of kenaf fiber MWL.

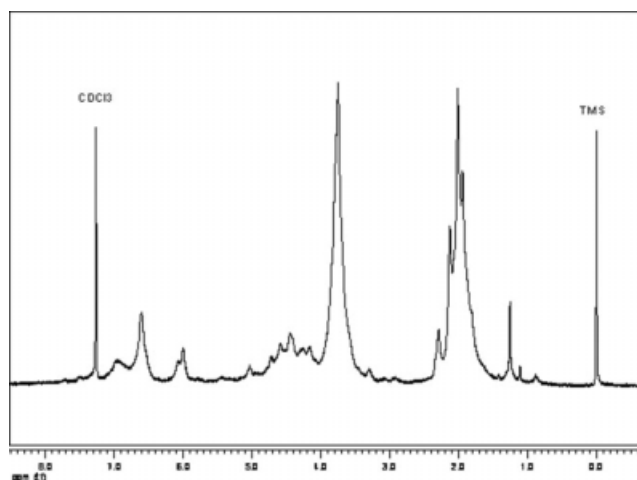


Figure 2 $^1\text{H-NMR}$ spectrum of acetylated kenaf fiber MWL.

$I(1220\text{--}1230\text{ cm}^{-1}) > I(1266\text{--}1270\text{ cm}^{-1})$, and $I(1121\text{--}1125\text{ cm}^{-1}) > I(1030\text{--}1040\text{ cm}^{-1})$, which is coincident with the IR spectra characteristics of the hardwoods.

Proposed by Pandey,⁴ in softwood lignin, the intensity at 1268 cm^{-1} band is much higher than that of 1214 cm^{-1} , and there is no syringyl absorption at 1315 cm^{-1} , whereas the opposite is true for hardwood lignins. That is, a weak 1267 cm^{-1} band, a strong band near 1218 cm^{-1} , and syringyl absorption at around 1315 cm^{-1} . The presence of syringyl unit in hardwood lignin is also evident from the high intensity of band 1462 cm^{-1} . The intensity of band at 1462 cm^{-1} is comparable to that of the 1503 cm^{-1} in hardwood lignin.

The spectrum indicates maximum absorbance at 1125 cm^{-1} , which is typical for hardwood lignin. This characteristic is sensitive for testing the existing of syringyl. In the spectra of softwood lignin, the absorption at 1140 cm^{-1} will be found. When there are syringyl unit existing in the lignin, the absorp-

tion at range $1175\text{--}1065\text{ cm}^{-1}$ will shift to the wave numbers less than 1128 cm^{-1} .^{6,7} Further evidence of the syringyl content of the lignin is afforded by its having a band near 835 cm^{-1} but no bands at 855 or 815 cm^{-1} , according to Obst,²⁴ the latter two are additional guaiacyl bands that are typical of softwoods and are not exhibited by hardwoods, which instead have the 835 cm^{-1} band. The observed absorption near 1045 cm^{-1} would suggest the presence of an approximately constant amount of uncondensed guaiacyl nuclei in lignins.²¹ The absorbance near 1166 cm^{-1} , which is typical for H unit,^{6,25} is not detected in the spectrum of kenaf fiber MWL.

The behavior of the spectrum in the $1065\text{--}1175$ region shows that, in terms of Faix⁶ classification, the kenaf fiber MWL is of type GS. It can in fact be further characterized as of type GS 4, because its absorbance at 1462 cm^{-1} is greater than that of the reference band near 1505 cm^{-1} , absorbance at 1268 cm^{-1} is not obvious, absorbance at 1125 cm^{-1} is high, and there is a well-defined band near 835 cm^{-1} .

$^1\text{H-NMR}$

The integrated NMR spectrum obtained for acetylated fiber MWL sample is shown in Figure 2. Subdivisions of the integration curve are made following Salud and Faix.²⁶ The estimation of the various types of protons per C_9 structural units has been conducted according to Ludwig et al.²⁷ and Isam and Sarkanen.²⁸ Table IV shows the subdivisions and distribution of protons found in the present samples.

Hydroxyl group

The numbers of hydroxyl groups are estimated from the acetate signals by considering per C_9 structure unit and the elemental analyses as well as the number of free aliphatic and phenolic hydroxyl groups. On

TABLE IV
Assignments of Signals and Protons per C_9 Structural Unit in the $^1\text{H-NMR}$ Spectra of Acetylated Lignins of Kenaf Fiber

Range number	Range (ppm)	ppm of the maximum	Main assignments	Protons per C_9 unit
	7.30	7.265	CDCl_3	
1	7.25–6.80	6.979	Aromatic protons in guaiacyl units	0.56
2	6.80–6.25	6.610	Aromatic protons in syringyl units	0.83
3	6.25–5.75	6.010	H_α of $\beta\text{-O-4}$ and $\beta\text{-1}$ structures	0.47
4	5.75–5.20	5.448	H_α of $\beta\text{-5}$ structures	0.25
5	5.20–4.90	5.041	H of xylan residue	0.28
6	4.90–4.30	4.441	H_α and H_β of $\beta\text{-O-4}$ structures	1.10
7	4.30–4.00	4.173	H_α of $\beta\text{-}\beta$ structures and H of xylan residue	0.73
8	4.00–2.50	3.751	H of methoxyl groups	3.81
9	2.50–2.22	2.288	H of aromatic acetates	0.42
10	2.22–1.60	2.015	H of aliphatic acetates	3.84
11	1.60–1.30	1.257	Hydrocarbon protons	0.65

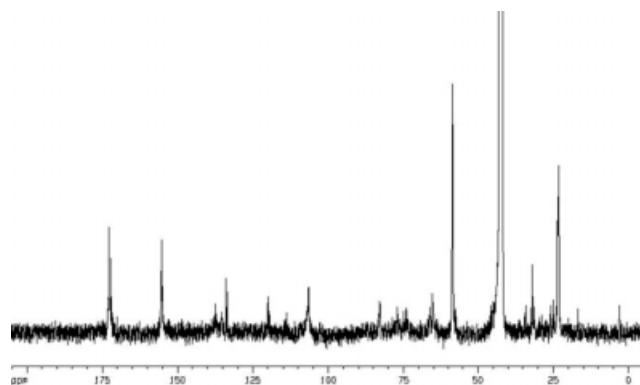


Figure 3 ^{13}C -NMR spectrum of kenaf fiber MWL.

this basis, the empirical formula is $\text{C}_9\text{H}_{9.32}\text{O}_{3.69}(\text{OCH}_3)_{1.30}$, considering both the aliphatic and phenolic hydroxyl groups per C_9 unit (Table III), the mole ratio is $\text{OA}_C/\text{OCH}_3 = (0.42 + 3.84)/3.81 = 1.12$. Thus, the total OA_C/C_9 ratio = $(1.30\text{OCH}_3/\text{C}_9) \times (1.12 \text{OA}_C/1 \text{OCH}_3) = 1.45$; the number of aliphatic $\text{OA}_C/\text{C}_9 = (1.30 \text{OCH}_3/\text{C}_9) \times (3.84 \text{A}_C/3.81\text{OCH}_3) = 1.31$, and the number of aromatic $\text{OA}_C/\text{C}_9 = (1.30 \text{OCH}_3/\text{C}_9) \times (0.42 \text{A}_C/3.81 \text{OCH}_3) = 0.14$. Therefore, the numbers of aliphatic and the free phenolic hydroxyls are estimated to be 131 and 14, respectively, per 100 C_9 units. And the OH_{aliph} is 90.3% in total numbers of hydroxyl groups of kenaf fiber MWL, and the OH_{ph} is 9.7%.

Aromatic protons

Kenaf fiber lignin spectrum exhibits two peaks in the aromatic proton region, which correspond to guaiacyl units (δ 6.979) and syringyl units (δ 6.61). NMR integration shows that kenaf fiber lignin contained $0.56 + 0.83 = 1.39$ aromatic proton per C_9 units.

β -O-4 Structure

The aryl glycerol β -O-4 aryl ether linkage constitutes the main intermonomeric connection in lignin.²⁹ NMR spectrum of kenaf fiber lignin shows that the structural element might contain both *erythro* and *threo* configurations because of the presence of proton at the $\text{C}-\alpha$ position of the side chain. The *erythro* protons (H_α) have stronger peak at δ 6.01 than the peak for *threo* form at δ 6.09. It has been reported³⁰ that the angiosperms lignin contains more *erythro* form in β -O-4 units than *threo* form.

β -5 Structure

There is no noticeable signal in range number 4 [Table IV and Fig. 3] between ppm δ 5.24 and δ 5.75 indicating low amount of phenylcoumaran units in the lignin structure.²²

β -1 and β - β Structure

Signals arising from the carbon atoms in β -1 and β - β units can be detected in kenaf MWL.

^{13}C -NMR

The ^{13}C -NMR spectrum obtained for the acetylated kenaf fiber lignins is shown in Figure 3, and their main signals are listed in Table V together with the assignments made following Nimz et al.,³¹ Delange et al.,³² and Scalbert et al.³³

In the aromatic region (104–165 ppm) of the spectrum, the syringyl and guaiacyl residues are indicated by signals at 155.2, 137.3, 133.9, 133.5, 106.4 ppm (S), 133.9, 133.5, 120, 119.7, ppm (G), respectively. For hardwood lignins, the most intense signals in this region are the C3/C5 and C2/C6 signals of syringyl units. This appearance in that of the kenaf MWL confirms that it is of GS (hardwood) type.

The strong resonance at 58.6 ppm corresponds to OCH_3 in syringyl and guaiacyl units.³⁴ The intensive signals assign to $-\text{methyl}$, and $-\text{methylene}$ groups in *n*-propyl side chains appeared in the spectrum between 14.0 and 33.8.²²

Apart from the methoxyl signal, the aliphatic region of lignin spectrum (50–90 ppm) can include signals for C_α , C_β , and C_γ in β -O-4 structures. In the spectrum obtained in this work, C_α appears at 74.1 ppm, C_β appears at 82.7 ppm, and C_γ appears at 65.1 ppm, respectively. The signal for C_γ in β -1 carbon–carbon bond can be seen at 77 ppm. These signals indicate that kenaf fiber lignin is composed mainly of ether bonds together with small amounts of carbon–carbon linkages.

TABLE V
Main ^{13}C -NMR Signals of Acetylated MWL
of Kenaf Fiber

δ (ppm)	Assignment
172.7	CO in acetates of primary alcohols
172.2	CO in acetates of primary alcohols
155.2	Se 3/5
137.3	Se 4
133.9	Ge 1, Se 1
133.5	Ge 1, Se 1
120	Ge 6
119.7	Ge 6, Ge 5
106.4	S 2/6
82.7	C_β in β -O-4
77	C_α in β -1
74.1	C_α in β -O-4
65.1	C_γ in β -1, α -O-4 and β -O-4
58.6	OCH_3
57.7	C_β in β - β
32.3	$-\text{CH}_2-$
31.5	$-\text{CH}_2-$
24.9	$-\text{CH}_3$ in acetyl

S, syringyl; G, guaiacyl; a, acetylated; e, etherified.

CONCLUSIONS

The average per C₉ unit formulae of kenaf fiber MWL can be expressed as C₉H_{9.32}O_{3.69} (OCH₃)_{1.30}, as calculated from determinations of the C, O, H, and OMe content of the lignins.

FTIR spectrum shows that the kenaf fiber MWL has more syringyl and fewer guaiacyl units.

From ¹H-NMR spectrum, the numbers of phenolic and aliphatic hydroxyl groups in the kenaf fiber MWL are estimated to be 0.14 and 1.31, respectively, per 100 C₉ units. Among total hydroxyl groups, 90.3% are OH_{aliph}, whereas 9.7% are OH_{ph}. The aryl glycerol β-O-4 aryl ether linkage constitutes the main intermolecular connection in kenaf fiber MWL, which contained more *erythro* form in β-O-4 units than *threo* form. β-1 and β-β linkage could be detected, whereas noticeable β-5 linkage cannot be detected in kenaf MWL. The ¹³C-NMR spectrum indicates that kenaf fiber lignin is composed mainly of ether bonds and small amounts of carbon-carbon linkages as well.

In general, the results of this study show that the kenaf fiber lignin is similar to hardwood lignin of type GS. In FTIR, it can be described as GS 4 type in detailed. From FTIR, it can be described as GS 4 type in detailed.

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References

- Nishimura, N.; Izumi, A.; Kuroda, K. I. *Ind Crops Prod* 2002, 15, 115.
- Wang, J. H.; Ramaswamy, G. N. *Text Res J* 2003, 73, 339.
- Shao, K. *Chemistry in Textile Processing*; China Textile Press: Beijing, 1996.
- Pandey, K. K. *J Polym Sci* 1999, 71, 1969.
- Song, K. H.; Obendorf, S. K. *Text Res J* 2006, 76, 751.
- Faix, O. *Holzforschung* 1991, 45, 21.
- Vazquez, G.; Antorrena, G.; Gonzalez, J.; Freire, S. *Holzforschung* 1975, 51, 158.
- Del, R.J. C.; Gutiérrez, A.; Rodríguez, I. M.; Ibarra, D.; Martínez, Á. T. *J Anal Appl Pyrolysis* 2007, 79, 39.
- Ralph, J. D. *J Nat Prod* 1996, 59, 341.
- Seca, A. M. L.; Cavaleiro, J. A. S.; Domingues, F. M. J.; Evtuguin, A. J. D.; Neto, C. P. *J Agric Food Chem* 1998, 46, 3100.
- Neto, C. P.; Seca, A.; Fradinho, D.; Coimbra, M. A.; Domingues, F.; Evtuguin, D.; Silvestre, A.; Cavaleiro, J. A. S. *Ind Crops Prod* 1996, 5, 189.
- Herbert Morrison, W., III; Akin, D. E.; Archibald, D. D.; Dodd, R. B.; Raymer, P. L. *Ind Crops Prod* 1999, 10, 21.
- Kuroda, K.-I.; Izumi, A.; Mazumder, B. B.; Ohtani, Y.; Same-shima, K. *J Anal Appl Pyrolysis* 2002, 64, 453.
- Mazumder, B. B.; Nakagawa-izumi, A.; Kuroda, K.-I.; Ohtani, Y.; Sameshima, K. *Ind Crops Prod* 2005, 21, 17.
- Kuroda, K.-I.; Nakagawa-izumi, A.; Mazumder, B. B.; Ohtani, Y.; Sameshima, K. *Ind Crops Prod* 2005, 22, 223.
- Björkman, A. *Sven Papperstidn* 1956, 59, 477.
- Lundquist, K.; Ohlsson, B.; Simonson, R. *Sven Papperstidn* 1977, 80, 143.
- Li, J. *Wood Spectroscopy*; Science Press: Beijing, 2003.
- Li, Y. M.; Chen, Z. H. *Trans Chin P P* 1999, 1, 20.
- Sarkanen, K. V.; Chang, H.-M.; Ericsson, B. *Tappi* 1967, 50, 572.
- Sarkanen, K. V.; Chang, H.-M.; Allan, G. G. *Tappi* 1967, 50, 587.
- Sun, R.-C.; Mott, L.; Bolton, J. *Wood Fiber Sci* 1998, 30, 301.
- Qin, T. F. *Sci Silve Sin* 1999, 35, 69.
- Obst, J. R. *Holzforschung* 1982, 36, 16.
- Guo, J. B.; Tao, Z. Y.; Luo, X. G. *J Instrum Anal* 2005, 24, 77.
- Salud, E. C.; Faix, O. *Holzforschung* 1980, 34, 113.
- Ludwig, C. H.; Nist, B. J.; McCarthy, J. L. *J Am Soc* 1964, 86, 1196.
- Isam, A.; Sarkanen, K. V. *Holzforschung* 1993, 47, 123.
- Sarwar Jahan, M.; Nasima Chowdhury, D. A.; Khalidul Islam, M.; Iqbal Moeiz, S. M. *Biores Technol* 2007, 98, 465.
- Akiyama, T.; Matsumoto, Y.; Okuyama, T.; Meshitsuka, G. *Phytochemistry* 2003, 64, 1157.
- Nimz, H. H.; Robert, D.; Faix, O.; Nemr, M. *Holzforschung* 1981, 35, 16.
- Delange, M.; Trummer, J. A.; Roberts, R. S.; Muzzy, J. D.; Gelbaum, L. T. *Holzforschung* 1991, 45, 361.
- Scalbert, A.; Monties, B.; Guitter, E.; Lallemand, J. Y. *Holzfor-schung* 1986, 40, 119.
- Wei, H. L.; Shi, S. L.; Pei, J. C. *Trans Chin P P* 2004, 19, 109.