SUMMARY

1. The dioxane lignin of ripe cotton-plant stems has been separated into six fractions of different molecular weights which were fairly homogeneous and differed considerably in their molecular weights.

2. It follows from the semiempirical formulas that in all the fractions guaiacyl structural units predominated. The chemical nonidentity of the fractions is shown by the different amounts of functional groups in the phenylpropane structural units and by the relative optical densities of the main bands in the IR spectra of the fractions.

3. The low-molecular-weight fraction differed markedly from the others by a higher content of carbohydrates bound to the lignin and by a greater degree of oxidation.

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DIOXANE LIGNINS OF KENAF BAST AND TOW

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The present paper gives the results of a comparative study of the dioxane lignins isolated from the outer (bast) and inner (tow) parts of the stems of kenaf of the cultivated variety Uzbekskii 15-74 gathered on the territory of the Sverdlov kolkhoz [collective farm], Tashkent Oblast'. Kenaf, like the cotton plant, belongs to the family Malvaceae.

The dioxane lignins from the bast (DLALK) and from the tow (DLAKK) were isolated by a method described previously [1]. The isolated dioxane lignins consisted of brown amorphous powders soluble in the same solvents as the DLAs of Althea [2] and the cotton plant [3]. After purification by Bjorkman's method [4], they contained 3.24% (DLAKK) and 3.88% (DLALK) of bound carbohydrates [5].

Below we give the elementary and functional analyses of the dioxane lignins obtained (%):

Elementary composition and amounts of functional groups	DLAKK	DLALK
C	60.18	59.60
H	6.14	6.02
O	33.68	34.38
OCH_3	20.31	19.78
OH_{tot}	10.44	10.78
OH_{alip}	8.17	7.99
OH_{ph}	2.27	2.79
CO	2.44	2.35
COOH	0.48	0.56

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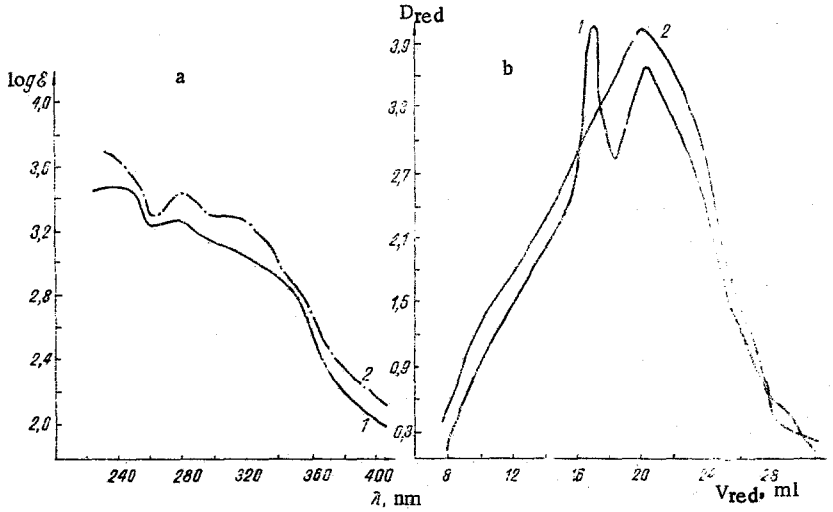

Fig. 1. UV spectra (a) and gel chromatograms (b) of kenaf dioxane lignins: 1) DLALK; 2) DLAKK.

These figures show that in their elementary compositions and amounts of functional groups the DLALK and DLAKK differed little from one another. At the same time, they differed considerably in their content of OCH_3 groups from the DLA of ripe cotton-plant stems [6]. For the DLALK and DLAKK we calculated the semiempirical formulas per elementary C_9 unit taking the presence of carbohydrates into account.

DLAKK, mol. wt. of a structural unit 205.6

$$C_9H_{7.14}\ O_{0.67}\ (OCH_3)_{1.39}\ (OH_{ph})_{0.27}\ (OH_{alip})_{0.92}\ (O_{ar\text{-}al})_{0.73}\ (O_{CO})_{0.18}\ (OOH_{COOH})_{0.022}$$

DLALK, mol. wt. of a structural unit 207.0

$$C_9H_{6.97}\ O_{0.73}\ (OCH_3)_{1.37}\ (OH_{ph})_{0.34}\ (OH_{alip})_{0.88}\ (O_{ar\text{-}alk})_{0.66}\ (O_{CO})_{0.18}\ (OOH_{COOH})_{0.026}$$

DLAK [1] (dioxane lignin from the whole kenaf stem)

$$C_9H_{6.60}\ O_{0.90}\ (OCH_3)_{1.34}\ (OH_{ph})_{0.25}\ (OH_{alip})_{1.01}\ (O_{ar\text{-}al})_{0.75}\ (O_{CO})_{0.17}\ (OOH_{COOH})_{0.02}$$

The DLA of ripe cotton-plant stems [3], molecular weight of a structural unit 199.6

$$C_9H_{6.85}\ O_{0.96}\ (OCH_3)_{1.00}\ (OH_{ph})_{0.30}\ (OH_{alip})_{0.87}\ (OH_{phA})_{0.1}\ (O_{ar\text{-}al})_{0.60}\ O(_{CO})_{0.21}\ (OOH_{COOH})_{0.045}$$

Thus, the differences between DLAKK and DLALK were insignificant. At the same time, they differed considerably from the DLA of ripe cotton-plant stems. The amounts of methoxy, hydroxy, carbonyl, and carboxy groups, and also the amount of alkyl–aryl ether bonds, were different. The kenaf lignins were more highly methoxylated than the lignins not only of the cotton plant but also of Althea [2]. They contained a larger amount of alkyl–aryl bonds, which shows their lower degree of condensation. In comparison with the DLA of ripe cotton-plant stems, DLAKK and DLALK contained smaller amounts of carbonyl and carboxy groups.

The UV absorption curves of the kenaf lignins that we obtained (Fig. 1a) have two maxima: $\lambda_{max} = 280$ and 300-360 nm (in the form of a shoulder). The maximum at 280 nm characterizes the absorption of the aromatic ring. The band at 300-360 nm is connected with the absorption of a benzene ring bearing substituents with conjugated double bonds and CO groups.

The UV absorption maxima of the dioxane lignins were as follows:

	λ_{max}	ε_{280}	OCH_3/C_9
DLAKK	280	2500	1.39
DLALK	280	2200	1.37
DLAK [1]	280	2600	1.34
DLA A. rosea [2]	280	2400	1.15
DLA of ripe cotton-plant stems [3]	280–282	2500	1.00

Thus, the molar extinctions of the kenaf DLAs are in the range of 2200-2500. They are composed of the extinctions of each phenylpropane structural unit (PPSU) by the law of additivity. The molar extinctions of kenaf

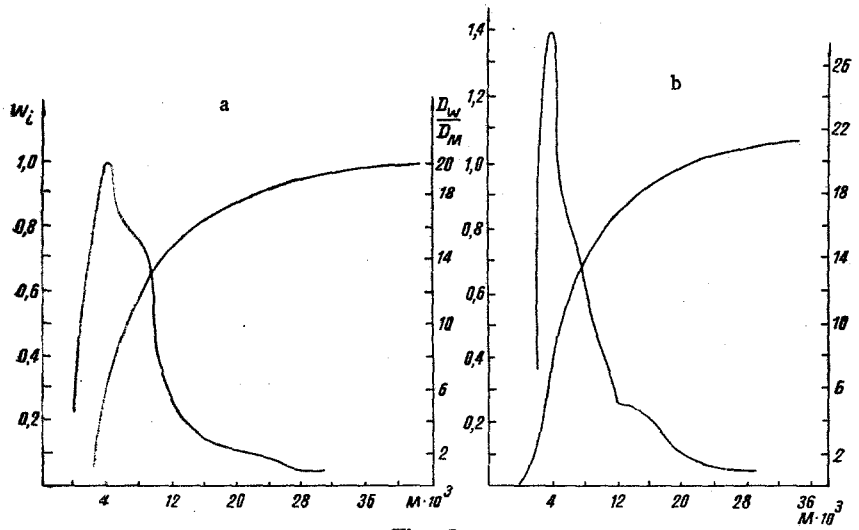

Fig. 2

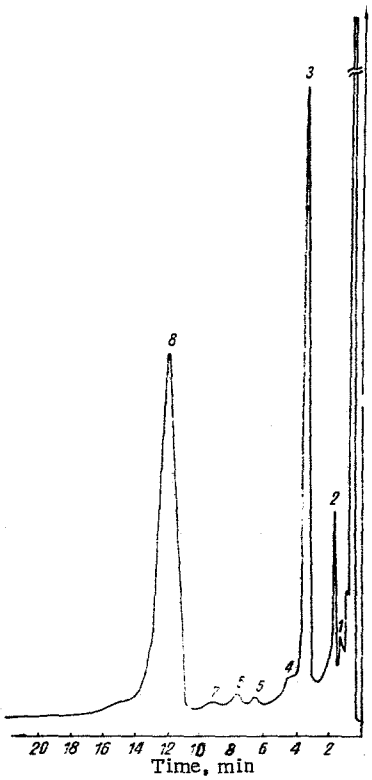

Fig. 3

Fig. 2. Differential and integral curves of kenaf DLAs: a) DLALK; b) DLAKK.

Fig. 3. Chromatogram of the products of the nitrobenzene oxidation of DLAKK: 1) ferulic acid; 2) p-coumaric acid; 3) vanillin; 4) acetovanillone; 5) p-hydroxybenzaldehyde; 7) p-hydroxyacetophenone; 8) syringaldehyde.

DLAs with a far higher value of the OCH_3/C_9 ratio than for Althea and the cotton plant are fairly high if the extinction of syringyl structures is taken into account ($\varepsilon_{280} = 1150$) [7]. We have previously reported the detection of dehydrodivanillin in the products of the oxidation of fractions of kenaf DLA. The high values of the molar extinctions of DLALK and DLAKK can possibly be explained partly, as well, by the presence of structures of the diphenyl type (ε_{280} for diphenylpropane is 6000).

The IR spectra of the lignins isolated show bands due to a benzene ring with substituents (1510, 1600, and 1470 cm^{-1}) and to hydroxy (3450 cm^{-1}), carbonyl (1720 cm^{-1}), and ether (1280 in the form of a shoulder, 1230, and 1040 cm^{-1}) groups. The fairly strong band at 1330 cm^{-1} corresponds to the absorption of C–H bonds in OCH_3 groups. Lindberg [9] considers that the absorption at 1330 cm^{-1} is due to syringyl nuclei. The kenaf lignins, like Althea lignins [2], showed a weak absorption band at 915 cm^{-1}. It is considered [10] that this absorption is also due to syringyl nuclei. Thus, we see that the spectra of DLAKK and DLALK are similar.

For a comparative study of the molecular weights (MWs) and polydispersities of the lignins we employed gel chromatography on Sephadex G-75, using the factors found previously [11]. The molecular-weight distribution (MWD) of the lignins was found from the gel chromatograms obtained in the elution of the samples through a column of gel calibrated for MWs (Figs. 1b and 2). The MWs and polydispersities of the dioxane lignin were:

	$\overline{M}_w$	$\overline{M}_n$	$\overline{Mz}$	$\overline{M}_w/\overline{M}_n$
DLAKK	10600	3700	22400	2.87
DLALK	8600	4200	19700	2.04
DLAK [1]	6500	2500	11100	2.57
DLA of A. rhyticarpa [2]	12000	5900	17300	2.05
DLA of ripe cotton-plant stems [3]	15200	5800		2 60

It can be seen from these figures and the gel chromatograms that the dioxane lignins of kenaf stems, Althea [2], and ripe cotton-plant stems [3] were polydisperse, but the low-molecular-weight fraction predominated in the kenaf dioxane lignins. The results show that the DLALK had a lower polydispersity than the DLAKK.

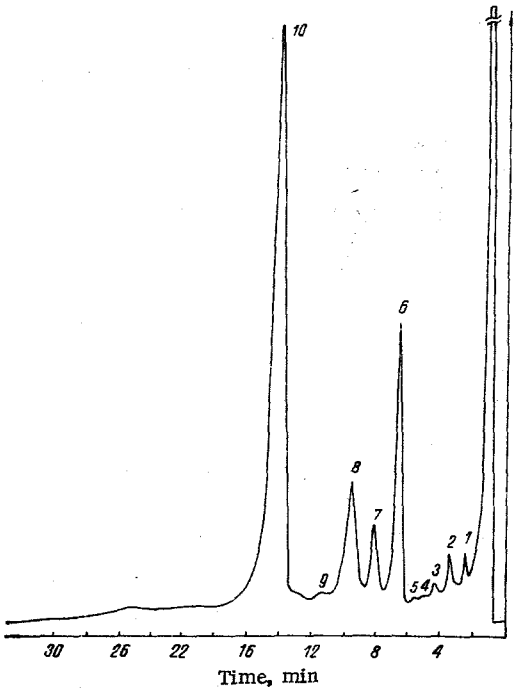

Fig. 4. Chromatogram of the products of the decomposition of kenaf tow with metallic sodium in liquid ammonia: 1) guaiacol; 2) p-hydroxyphenylpropane; 3) vanillyl alcohol; 4) 4-hydroxy-3-methoxyphenylethane; 5) 1-(4-hydroxy-3-methoxyphenyl)ethanol; 6) 4-hydroxy-3-methoxyphenylpropane; 7) vanillin; 8) 1-(4-hydroxy-3-methoxyphenyl)-propan-1-ol; 9) 3-(4-hydroxyphenyl)propan-1-ol; 10) 4-hydroxy-3,5-dimethoxyphenylpropane.

From the values of $\overline{M}_z$ it is possible to calculate on an average of how many phenylpropane structural units the macromolecules of the kenaf dioxane lignins consist. Thus, it has been calculated that the DLAKK consists of 109 and the DLALK of 95 PPSUs.

To study the structures of the lignins isolated, in addition to physicochemical methods we used alkaline nitrobenzene oxidation [12] and reductive degradation with metallic sodium in liquid ammonia [13].

The products of nitrobenzene oxidation (NBO) of the kenaf lignins were investigated by the GLC method (Fig. 3). We give the results of their quantitative chromatographic analysis (% on the initial lignin):

Substance	DLAKK	DLALK	DLAK [1]
p-Hydroxybenzaldehyde	0.66	3.06	3.55
p-Hydroxyacetophenone	—	—	0.17
p-Coumaric acid	1.57	0.30	—
p-Hydroxybenzoic acid*	—	0.54	
Guaiacol	0.13	—	—
Ferulic acid	0.18	0.61	0.90
Vanillin	12.57	13.45	14.37
Acetovanillone	0.54	0.12	—
Vanillic acid*	1.46	0.91	
Syringaldehyde	28.85	27.12	36.19
Syringic acid*	7.12	3.43	
Unidentified	1.59	0.86	
p-Coumaryl: guaiacyl: syringyl ratio	0.15:1:2.4	0.26:1:2.02	0.26:1:2.5

The Althea [2] and cotton-plant [3] lignins contain all three types of structural units: p-coumaryl, guaiacyl, and syringyl. The differences between the DLAKK and DLALK consist in the fact that the latter contained a larger amount of p-coumaryl and guaiacyl structures. However, it must be mentioned that the amount of p-coumaryl structures in both lignins was very small (5-7 times less than the amount of guaiacyl structures). Both in the DLAKK and in the DLALK there were two or more syringyl units for each guaiacyl structural unit. This is obviously connected with the fact that the syringyl structures have fewer possibilities for condensation. In other words, since syringyl structures predominated among the oxidation products, there were more "uncondensed" syringyl structures in the kenaf lignins than structures of the guaiacyl series.

A comparison of the products of the nitrobenzene oxidation of the DLAs of kenaf bast and tow with the DLA of the whole plant shows that the latter contained the same compounds. The differences between them appear mainly in the amounts of these compounds.

*Determined in the form of methyl ethers of methyl esters, but not determined in the DLAK.

Among the products of the nitrobenzene oxidation of kenaf DLA, in addition to aromatic aldehydes and ketones, there are aromatic acids which, as is well known [14, 15], are also formed by this reaction and which we did not succeed in detecting under the given conditions of analysis because of their low volatility. To increase their volatility we subsequently methylated [16] the mixture of oxidation products and then studied them by gas–liquid chromatography. The alkaline (pH 11-12) and acid (pH 1-2) fractions were investigated separately. The formers consisted of aldehydes and ketones while aromatic acids were found in the latter, together with aldehydes.

If the yield of vanillic acid is added to the yield of vanillin and other products of the guaiacyl series (it is the total yield of the latter that gives an idea of the amount of uncondensed guaiacyl units), it is found that the total yield in DLALK was somewhat higher than in DLAKK. Consequently, the guaiacyl units were condensed differently in these lignins. They were less condensed in DLALK, and obviously this contained fewer structures with a diphenyl bond and also diaryl structures. The decomposition with metallic sodium in liquid ammonia of both natural and isolated lignins leads to the cleavage of alkyl–aryl ether bonds [17] with the liberation of the "uncondensed" structural elements. We subjected the DLALK and DLAKK, and also the natural lignins of the bast and tow, to this treatment. The yields of phenols from a single treatment of kenaf dioxane lignins were 11.69 and 6.17% of the weight of the initial lignin for the DLAKK and DLALK, respectively. Consequently, there were somewhat more "uncondensed" aromatic units in the DLAKK than in the DLALK.

To investigate the complex mixture of phenols isolated after the separation of the kenaf lignin we used gas–liquid chromatography (Fig. 4). The peaks were identified from their retention times and by the addition of standard substances.

The amounts of phenolic substances in the decomposition products of the natural lignins and the kenaf DLAs were as follows:

	Stems, % on the Komarov lignin		Dioxane lignin, % on the initial lignin	
	two	best	DLAKK	DLALK
4-Hydroxyphenylpropane	0,11	—	0,15	0,41
3-(4-Hydroxyphenyl)propan-1-ol	0,14	0,33	—	—
Guaiacol	0,05	0,01	0,15	—
1-(4-Hydroxy-3-methoxyphenyl)propane-1,3-diol	—	0,03	0,15	—
Vanillyl alcohol	0,05	0,02	—	—
4-Hydroxy-3-methoxyphenylethane	0,03	0,06	0,22	—
1-(4-Hydroxy-3-methoxyphenyl)ethanol	0,03	—	—	—
4-Hydroxy-3-methoxyphenylpropane	1,63	1,21	1,95	1,94
Vanillin	0,60	1,25	0,49	1,28
1-(4-Hydroxy-3-methoxyphenyl) propan-1-ol	1,11	2,06	0,07	1,15
3-(4-Hydroxy-3-methoxyphenyl) propan-1-ol	—	—	1,01	—
4-Hydroxy-3,5-dimethoxyphenyl)propane	7,74	8,97	3,35	3,91
1-(4-Hydroxy-3,5-dimethoxyphenyl)propan-1-ol	0,22	1,62	1,19	2,16
Unidentified	0,02	1,98	3,06	—
p-Coumaryl : guaiacyl : syringyl ratio	0,07:1:2,27	0,07:1:2,27	0,04:1:1,22	0,09:1:1,35.

It follows from these figures that, as in the case of nitrobenzene oxidation, syringyl units predominated in the mixture. This once again confirms the smaller degree of condensation of the syringyl structures than of the guaiacyl structures in the macromolecules of the lignins studied.

A comparison of the products of the cleavage of natural lignins of the bast and tow shows that they differ insignificantly in qualitative and quantitative composition (number of peaks). Nevertheless, in the natural bast lignin there was a predominance of guaiacyl and syringyl units with free OH groups in the side chains in the α positions to the aromatic nucleus (the amount of 1-guaiacylpropan-1-ol in the bast lignin was twice as great as in the tow lignin, and the amount of 1-syringylpropan-1-ol was seven times as great).

So far as concerns the dioxane lignins, considerable differences in the qualitative compositions of the phenolic fractions were observed here. Ten compounds were identified in the decomposition products of DLAKK which were mainly derivatives of guaiacyl- and syringylpropanes. In the alkaline fraction of DLALK, however, only six compounds were observed, among which, just as in DLAKK, p-coumaryl derivatives were poorly represented. Ethane structures were completely absent from the DLALK. However, as also in the case of the natural lignins, a larger amount of α-alcohols of the guaiacyl and syringyl series was found in the products of the decomposition of DLALK as compared with DLAKK.

The fact that in natural kenaf lignins there were more than two syringyl structural units for each guaiacyl unit and there were fewer in the dioxane lignins can apparently be explained by condensation processes taking

place during the isolation of the lignin. On the other hand, in the oxidation of the same kenaf DLA preparations there were two syringyl structures to each guaiacyl structure. One must not forget, however, that probably only the peripheral parts of the macromolecule undergo oxidation, while decomposition with metallic sodium in liquid ammonia affects the whole molecule. From this it may be concluded that the side chains within the molecule and in its peripheral part are different. The former of those mentioned above are more condensed (approximately twofold) than the latter.

Thus, on the basis of what has been said it may be concluded that DLALK and DLAKK differed from one another not only by the ratio of the three structural units making up their macromolecules but also by the structure of the side chains of the PPSUs.

EXPERIMENTAL

The isolation of kenaf dioxane lignins was carried out by a method described previously [1]. The functional groups of these lignins were determined by standard methods [18].

The UV spectra were recorded on a SF-4 spectrophotometer with dioxane—water (9:1) as solvent. The IR spectra were taken on a UR-20 instrument in tablets with KBr.

Gel chromatography was performed on an analytical column of Sephadex G-75, using DMSO as solvent and eluent.

Alkaline nitrobenzene oxidation was carried out by the method described previously [19]. The total yields were 0.2304 g (44.62%) and 0.2142 g (42.81% on the Komarov lignin) for DLAKK and DLALK, respectively. Chromatographic analysis was performed under the conditions used previously [8].

Method of Successive Methylation. The total products from the oxidation of kenaf (0.2039 g) were treated with 0.13 g of caustic soda in 3.3 ml of H_2O and 0.12 ml of dimethyl sulfate. This mixture was boiled on the water bath for half an hour and, after cooling, it was extracted with ether. The ethereal extract of the reaction mixture, which had pH 11-12, was dried over anhydrous $MgSO_4$ and the residue after the elimination of the ether was dissolved in ethanol and was investigated by the GLC method. The aqueous layer was acidified with 5% H_2SO_4 to pH 1-2 and was reextracted with ether. The ethereal extract, after being dried with anhydrous $MgSO_4$, was evaporated to a volume of 6-8 ml and was treated with 10 ml of diazomethane. After standing for 40 minutes, the solvent was distilled off to dryness, and the dry residue was dissolved in ethanol and studied by gas—liquid chromatography.

GLC Conditions. The following stationary liquid phases were tested for the separation of the mixtures of methyl ethers and esters of the products of oxidation with nitrobenzene in an alkaline medium: Apiezon L, SE-30, and UZh F (universal liquid phase). The best results were obtained on working with SE-30.

The products were analyzed on a Khrom-4 chromatography with a flame-ionization detector in a stainless-steel column (120 × 0.3 cm) filled with 5% of SE-30 on Chromaton NAW at a thermostat temperature of 130°C and an evaporator temperature of 180°C. Helium was used as the carrier gas.

Reductive degradation with metallic sodium in liquid ammonia and the study of the reaction products by the GLC method were performed as described previously [2]. The lignin samples amounted to 0.5-0.6 g. The peaks were evaluated quantitatively by the area-normalization method [20].

SUMMARY

1. Dioxane lignins have been isolated from the outer (bast) and inner (tow) parts of kenaf of the variety Uzbekskii 15-74, and the semiempirical formulas of the phenylpropane structural units of these dioxanes have been calculated on the basis of elementary and functional analyses.

2. It has been shown by measurements of molecular-weight distribution that the DLAKK was more polydisperse than the DLALK.

3. A study of the products of alkaline nitrobenzene oxidation and of decomposition by metallic sodium in liquid ammonia has shown that the DLAKK and DLALK were constructed of p-coumaryl, guaiacyl, and syringyl structural units, with the latter predominating.

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NITROBENZENE OXIDATION OF FRACTIONS OF COTTON-PLANT DIOXANE LIGNIN

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The dioxane lignin (DLA) of the ripe stems of the cotton plant of variety 108-F has previously been fractionated according to molecular weights by successive precipitation in the form of six fractions. It has been shown that the fractions differ not only in their molecular weights but also in the amounts of functional groups per C_9 unit and, consequently, in chemical structure [1]. To study their structures, the DLAs of the six fractions and the initial unfractionated lignin were oxidized with nitrobenzene in an alkaline medium [2]. The oxidation products were analyzed quantitatively in a gas–liquid chromatograph. They were identified from their retention times and by the addition of standard substances, and were determined quantitatively by the area-normalization method [3]. Below we give the yields of oxidation products in molar percentages [(% in the mixture/mol. wt.) · 10^2]:

	Fraction						Initial data
	I	II	III	IV	V	VI	
p-Coumaric acid	0.32	0.26	0.43	1.16	0.39	0.30	0.21
p-Hydroxybenzaldehyde	1.22	0.60	0.82	0.78	1.61	1.67	0.36
p-Hydroxyacetophenone	0.44	0.70	0.94	1.48	1.72		0.93
Total p-coumaryl structures	1.98	1.56	2.19	3.42	3.72	1.97	1.41
Ferulic acid			0.38		0.31	0.51	0.32
Vanillin	29.53	26.71	28.12	32.67	32.25	30.46	30.99
Acetovanillone		0.44	0.64				0.81
Acetoguaiacone				0.74		1.68	
Total guaiacyl structures	29.53	27.18	29.14	33.41	32.56	32.65	32.12
Syringaldehyde	28.62	30.98	28.12	24.28	23.98	22.40	26.19

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