# A STUDY OF THE DIOXANE LIGNIN OF THE STEMS OF THE COTTON PLANT OF VARIETY S-4880

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The dioxane lignin (DLA) has been isolated from the ripe stems of a cotton plant of the variety S-4880 B by a modification of Pepper's method. The yield was 16.9% on the Komarov lignin. The semiempirical formula of a phenylpropane structural unit has been calculated on the basis of elementary and functional analyses. It has been established that the amount of the main functional groups in the DLA and in the lignins isolated from the varieties of 108-F and Tashkent-2 basically coincide, but the DLA investigated was appreciably more highly methoxylated. The IR, UV, and PMR spectra of the DLA have been taken. The molecular-weight distribution of the DLA has been investigated and it has been shown that is is polydisperse and has a weight-average molecular weight of 7400. Nitrobenzene oxidation and cleavage with sodium and liquid ammonia of the isolated DLA has shown that guaiacyl structural units predominate in it.

The cotton plant variety S-4880 that we have investigated differs from the 108-F variety and those of the Tashkent series by a high crop yield and resistance to wilt [1]. We have studied ripe stems collected after the harvesting of the cotton crop. The plant was ground and passed through a 0.25-mm sieve. Then it was subjected to 48-h ethanol—benzene (1:2) extraction and extraction with hot water.

Dioxane lignin (DLA) close to the natural lignin was isolated from the ground cotton plant of variety S-4480 by a modification of Pepper's method [1]. The yield of DLA was 16.9% on the Komarov lignin. A light brown powder readily soluble in the usual lignin solvents was obtained. The DLA was purified by two reprecipitations of its aqueous dioxane (1:9) solution in absolute ether. The semiempirical formula of a phenylpropane structural unit was calculated from the elementary and functional analyses with allowance for the amount of carbohydrates present (1.16%):

Molecular weight 207.5:

$$C_9H_{7,81}O_{0,58}(OCH_3)_{1,31}(OH_{ph})_{0,24}(OH_{al})_{0,92}(O_{CO})_{0,56}(O_{ar\text{-}al})_{0,76}(OOH_{COOH})_{0,03}.$$

DLA of the variety 108-F (II):

$$C_9H_{8,02}O_{0,58}(OCH_3)_{1,23}(OH_{ph})_{0,44}(OH_{al})_{0,96}(O_{CO})_{0,22}(O_{ar\text{-}al})_{0,56}(OOH_{COOH})_{0,08}.$$

DLA of the variety Tashkent-1 (I):

$$C_9H_{6,22}O_{1,21}(OCH_3)_{0,82}(OH_{ph})_{0,63}(OH_{al})_{0,88}(O_{CO})_{0,46}(O_{ar\text{-}al})_{0,37}(OOH_{COOH})_{0,17}.$$

A comparison with the composition of the dioxane lignin isolated from ripe stems of the cotton plant of the 108-F and Tashkent-1 varieites showed that the DLA contained a far smaller amount of phenolic OH groups but was more highly methoxylated, while the number of carbonyl groups per structural unit proved to be 2.5 times greater than for variety 108-F and 1.3 times greater than for variety Tashkent-1.

To investigate its molecular-weight distribution (MWD), the DLA was subjected to gel chromatography on Sephadex G-75 using dimethyl sulfoxide as eluent and solvent [3]. The eluogram and the integral and differential curves of molecular weight distribution are given in Figs. 1 and 2.

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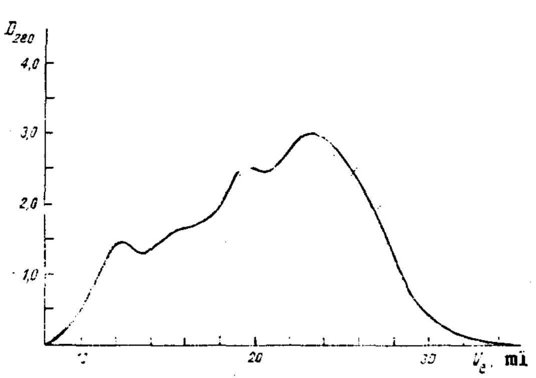

Fig. 1

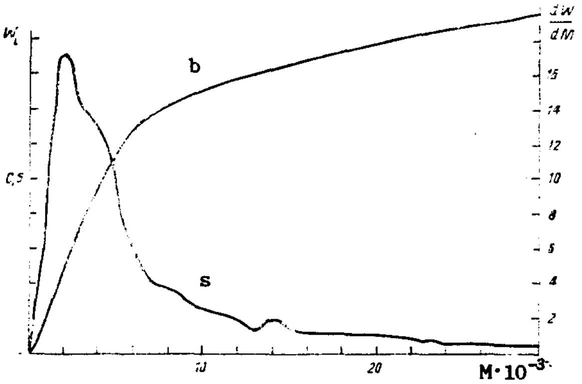

Fig. 2

Fig. 1. Eluogram of the molecular-weight distribution of the DLA of a cotton plant of variety S-4880.

Fig. 2. Integral (a) and differential (b) curves of the molecular-weight distribution of the DLA of a cotton plant of variety S-4880.

The number-average ($\bar{M}_n$), weight-average ($\bar{M}_w$), and mean ($\bar{M}_z$) molecular weights were calculated with the use of the coefficients found previously:

$$\overline{M}_n = 2800;\ \overline{M}_w = 7400;\ \overline{M}_z = 15000,\ \overline{M}_n : \overline{M}_W : \overline{M}_z = 1 : 2{,}64 : 5{,}35.$$

An analysis of the MWD curve showed that the lignin under investigation was polydisperse.

The IR spectrum of the DLA contained all the main absorption bands characteristic for lignins, $\nu_{max}$, cm$^{-1}$: 3440 (OH groups and associated hydrogen bonds); 2940 (stretching vibrations of C–H bonds); 2860, 1465, 1425, 1330 (C–H bonds in $OCH_3$ groups); 1725 (CO bonds); 1600, 1520 (aromatic rings); and 1230 (phenolic OH groups).

The UV spectrum of the DLA taken in methylcellosolve showed a maximum at λ 280 nm corresponding to the absorption of aromatic rings with substituents.

The nature of the substitution of the aromatic rings and of the side chain of the lignin can be calculated from an analysis of the PMR spectrum of the acetylated DLA. The quantitative interpretation of the PMR spectrum was carried out according to [4]. The value of one proton was calculated on the basis of the percentage content of methoxy groups (zone IVa) and their number in the seimempirical formula of a phenylpropane structural unit. The results of the calculation are given below.

| Zone of chemical shifts | Type of protons | ppm | Number of protons |
|---|---|---|---|
| I | Aromatic | 2.0–3.7 | 1.83 |
| II | β-Vinyl and benzyl ether protons of the side chain | 3.7–4.3 | 0.36 |
| III | Coumarane structures | 4.3–4.8 | 0.27 |
| IV | Methoxy protons and protons of $C_3$ side chain | 4.8–7.5 | 6.93 |
| IVa | Methoxy | 6.06–6.6 | 3.93 |
|  | Other (α-, β-, and γ-protons of the side chain) |  | 3.03 |
| V | Aromatic acetoxy groups | 7.5–7.8 | 1.13 |
| VI | Aliphatic acetoxy groups | 7.8–8.5 | 2.56 |
| VII | Highly screened $CH_3$ and $CH_2$ groups | 8.5–9.5 | 1.10 |

According to the PMR spectrum, the number of phenolic OH groups was high and that of aliphatic OH groups low in comparison with the semiempirical formula. This is obviously connected with the poor resolution of the bands in the spectrum.

The total number of protons per $C_9$ in the spectrum was lower than according to the analytical results. The number of free aromatic protons in the DLA investigated was 1.83. We know theoretically that in each PPSU of lignin the number of free aromatic protons is 4 − 1.31 = 2.69. We calculated the deficiency of aromatic protons: 2.69 − 1.83 = 0.86. Knowing that the deficiency of aromatic protons in the DLA of stems of a cotton plant of variety 108-F is 0.43 and for that of Tashkent-1 it is 1.52 [2] it is possible to draw the conclusion that the stem DLA of the S-4880 variety of cotton plant occupies an intermediate position with respect to degree of condensation between the 108-F and Tashkent-1 varieties. It also proved to be poorer in coumarane structures, protons of the side chain, and benzyl alcohol groups.

The structure of the lignin isolated was investigated with the aid of nitrobenzene oxidation (NBO) and by reductive cleavage with sodium in liquid ammonia [5, 6].

On treatment of the products of NBO, the acid fraction was extracted first and then the aldehyde fraction, and they were analyzed separately. For the DLA the yield of products was 36.14%. We give the qualitative and quantitative composition of the products of NBO (in percentages on the DLA): p-hydroxybenzoic acid (0.38), vanillic acid (5.13), p-coumaric acid (0.15), syringic acid (0.72), guaiacol (0.73), vanillin (15.24), and syringaldehyde (10.39). The ratio of p-coumaryl, guaiacyl, and syringyl structural units was 0.02:1:0.63. In the products of NBO isolated from the DLA of a cotton plant of the S-4880 variety guaiacyl structures predominated.

Continuing an investigation of the structure of the dioxane lignin of the stems of the S-4880 cotton plant, we used the method of cleavage by metallic sodium in liquid ammonia. The sum of the cleavage products amounted to 56.3% (18.98% in an ethereal extract at pH 8, and 37.3% in an ethyl acetate extract at pH 2). The following were identified in the ethereal extract:

| Substance | % on the total | % on the DLA |
|---|---|---|
| p-Hydroxyphenylethane | 2,17 | 0,28 |
| p-Hydroxyphenylpropane | 2,02 | 0,23 |
| Guaiacylpropane | 30,39 | 3,96 |
| Vanillin | 17,70 | 2,30 |
| 1-Guaiacylethanol | 1,20 | 0,16 |
| 1-Guaiacylpropan-1-ol | 0,72 | 0,09 |
| 3-Guaiacylpropan-1-ol | 1,20 | 0,16 |
| Syringylpropane | 44,57 | 5,80 |

The ratio of p-coumaryl to guaiacyl to syrigyl units was 0.08:1:0.90.

The qualitative composition of the monomeric cleavage products showed the presence in the side chains of the phenylpropane structural units of the isolated dioxane of hydroxy groups in the α- and γ-positions. The absence of products with β-OH groups showed that the predominating ether bonds in the molecule of the lignin under investigation were β-arylalkyl, decomposing in the course of cleavage. The formation of products with unsubstituted side chains showed that under the action of sodium and liquid ammonia an elimination of aliphatic hydroxy groups took place.

The results of the cleavage by sodium in liquid ammonia confirmed that the main structures in the DLA were guaiacyl and the next in amount were syringyl structures, while there was a very small amount of p-coumaryl structures.

The yield of phenolic products soluble in ethyl acetate at pH 2 was 37.32%. This fraction was characterized by gel filtration of Sephadex LH-20 in the ethanol–water (9:1) system using the distribution coefficients given in [7]. On the basis of the eluograms obtained it was possible to conclude that this combined material included oligomeric, tetrameric, trimeric, dimeric, and monomeric products of the degradation of lignin.

## EXPERIMENTAL

The dioxane lignin was isolated by a modification of Pepper's method. The yield was 16.9% on the Komarov lignin. The determination of the functional groups and the calculation of the semiempirical formula were carried out by standard methods [8]. The results of the analysis were (%): C − 59.74; H − 6.27; $OCH_3$ − 19.79; CO − 7.58; $OH_{tot}$ − 9.41; $OH_{phen}$ − 1.94; $OH_{acid}$ − 0.52.

GLC analysis was performed on a Chrom 4 instrument with a flame-ionization detector. The combined acids were analyzed in the form of their methyl esters under the following conditions: stainless-steel column (0.3 × 120 cm) filled with 5% of SE-30 on Chromaton NAW-DMCS (0.165-0.200 mm), rate of flow of carrier gas (helium) 40 ml/min, temperature 160°C.

The combined aldehydes were analyzed under the following conditions: stainless-steel column (0.3 × 120 cm) filled with 4% of PEGA on Chromaton NAW-DMCS (0.165-0.200 mm), rate of flow of carrier gas (helium) 50 ml/min, temperature 180°C.

The combined phenols were analyzed under the following conditions: stainless-steel column (0.3 × 370 cm) filled with 15% of Apiezon L on Chromaton NAW-DMCS (0.200-0.250 mm), rate of flow of carrier gas (helium) 40 ml/min, temperature 205°C. Identification was carried out from retention times and by spiking with standard samples.

Gel chromatography was performed on Sephadex G-75 using dimethyl sulfoxide as eluent and solvent. The coefficients found in [3] were used, and the number-average, weight-average and mean molecular weights were calculated by standard methods [9].

UV spectra were taken in methylcellosolve on a SF-6 spectrophotometer. $\lambda_{max}$ 280 nm, log ε 3.4900 ($C = 2.263 \cdot 10^{-4}$ M), and IR spectra on a UR-20 instrument (tablets with KBr). The PMR spectrum of the acetylated DLA was recorded on a JNM-4H-100/100 MHz spectrometer at room temperature, C = 10-12% by weight, 10 — HMDS, solvent $CDCl_3$. Calculation was carried out by the method of Alekseev et al. [3].

The nitrobenzene oxidation of DLA was carried out by Leopold's method [5]. The acids were isolated from an ethereal extract with 5% sodium bicarbonate solution, and then the aldehydes and ketones were isolated with a 6% solution of sodium bisulfite. Cleavage with sodium and liquid ammonia was carried out as described in [6].

## SUMMARY

1. Dioxane lignin isolated from a cotton plant of the variety S-4880 and characterized had a number-average molecular weight of 2800, a weight-average molecular weight of 7400, and a mean molecular weight of 15,000. The degree of polydispersity was 2.64.

2. The results of nitrobenzene oxidation and cleavage with sodium in liquid ammonia have shown that the predominating type of structures in the lignin isolated was the guaiacyl type.

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