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WATER-SOLUBLE POLYSACCHARIDES OF SEEDS OF THE GENUS *Gleditsia*

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The water-soluble polysaccharides of the seeds of eight species of Gleditsia *have been isolated and studied. They are galactomannans with different viscosities, specific rotations, and ratios of galactose and mannose. A study of their chemical transformations and IR spectral data has shown that the monosaccharide residues are pyranoses linked by β-1—4-glycosidic bonds having in the second component* D-galactose *with the α-configuration of the glycosidic center and the most probable chair conformation of the pyranose ring.*

The genus *Gleditsia* (honey locust) belongs to the family Caesalpiniaceae [1] and numbers 12 species growing wild in North and South America, east Asia, and tropical Africa.

Eight species and forms of honey locust have been introduced into the F. N. Rusanov Botanical Garden, Academy of Sciences of the Republic of Uzbekistan. We have previously reported a study of the carbohydrates and proteins from the seeds of big-spine honey locust (*Gleditsia macracantha*) and have isolated a water-soluble polysaccharide — a galactomannan [2]. With the aim of determining the distribution of galactomannans in the *Gleditsia* genus, we have studied the water-soluble polysaccharides (WSPSs) of eight species.

The WSPSs were isolated as described in [2]. Their yields and characteristics are given in Table 1. The honey locust seeds contained from 6.3 to 21.2% of WSPSs. The polysaccharides formed fibrous powders, white with a cream tinge. On dissolution in water they formed very viscous solutions. The intrinsic viscosities of the various galactomannans were high: from 7.54 to 10.43 liters/mg.

To determine their qualitative and quantitative monosaccharide compositions, the polysaccharides were subjected to complete acid hydrolysis. Galactose and mannose were detected by PC and GLC. Consequently, the polysaccharides of the species that we have studied, like those described previously [2], are galactomannans.

It was established that the ratios of galactose and mannose in the galactomannans differed quantitatively (see Table 1).

In disagreement with literature figures, in the galactomannans isolated from *G. caspia* and *G. triacanthos*, the ratios of galactose and mannose were 1:8.3 and 1:4.6, respectively [3, 4].

The constancy of the ratios of galactose and mannose when the WSPSs were fractionated with heavy-metal salts, such as lead acetate, Fehling's solution, and barium hydroxide confirmed the homogeneity of the galactomannans. Furthermore, the results of precipitation of the WSPSs with these reagents permitted the assumption of the presence of β-1—4-bound mannopyranose residues in the chains [5].

In the analysis of the IR spectra of the GMs under investigation (**1**—**8**) the two most informative frequency ranges were singled out: 700—900 and 3000—3600 cm^{-1}.

The presence in the spectrum of each GM (**1**—**8**) of two absorption bands, at 820 and 880 cm^{-1}, which are observed in the IR spectra of β-mannans [6] and glucomannans [7] and correspond to the deformation vibrations of C—H bonds [8], showed that the mannose fragments in the galactomannans under investigation were pyranoses and were linked by β-1—4-glycosidic bonds. This situation is responsible for the negative angle of rotation of various polymers. However, polysaccharides (**1**—**8**) possess positive angles of rotation, which gives grounds for assuming the existence of the α-configuration of the glycosidic center

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TABLE 1. Yields, Physicochemical Characteristics, and IR Spectral Frequencies in the 700—800 and 3000—3500 cm^{-1} Regions of Galactomannans (GMs) from the Genus *Gleditsia*

GM	Plant	Yield of WSPSs, %	Gal:Man ratio	$[\eta]_{rel}$ (c 0.5%; H_2O)	$[\alpha]_D$, deg. (c 0.1%; H_2O)	$S_{C\text{-}H}$, cm^{-1}	ν_{OH}, cm^{-1}
1	G. japonica	10.0	1:5.0	119.9	+17	767, 799	3393
2	G. caspia	11.5	1:8.3	101.5	+13	769, 799	3336
3	G. triacanthos	18.0	1:4.6	101.5	+7	768, 799	3293
4	G. aquatica	21.2	1:2.6	184.7	+10	768, 799	3293
5	G. delavayl	11.9	1:1.7	121.1	+20	768, 799	3398
6	G. texana	18.0	1:3.8	139.4	+35	768, 799	3394
7	G. sinensis	6.30	1:2.8	92.5	+22	768, 799	3361
8	G. macracantha	14.6	1:5.0	43.4	+35	-	3429

in the second component, *D*-galactose. This experimental fact was confirmed by the presence in the spectrum of each of the polysaccharides under investigation of an absorption band at 720 cm^{-1} corresponding to the pulsation vibrations of the pyranose ring of α-*D*-galactose [9, 10].

The dependence of the frequencies of the pulsation vibrations of a pyranose ring, which appear in the 700—800 cm^{-1} region, on the configuration of the C—H groups can be used for the conformational analysis of the pyranose ring [10, 11]. In view of what has been said above, the absorption bands at 870 and 790 cm^{-1} in compounds (**1**—**7**) presuppose the presence of equatorial (E) and axial (A) C—H groups at the C_1—C_4 atoms of the EAAE and AEAA types, which corresponds to the chair conformation (C1) of the pyranose rings in the galactomannans concerned.

It must be mentioned that these frequencies were present in the IR spectra of all the galactomannans with relative viscosity coefficients of 92—184.7, but were absent in the case of the polysaccharide of *G. macracantha*, which has $\eta = 43.4$. This fact is probably explained by the existence of Van-der-Waals interactions in polysaccharides (**1**—**7**), preventing the formation of OH···H intermolecular and intramolecular associations and, for this reason, favoring the formation of free OH and CH_2OH functional groups.

On the basis of a comparison with the IR spectra of *D*-galactose and *D*-mannose in this interval [10], the OH absorption observed in the frequency interval of 3290—3400 cm^{-1} shows the most probable chair conformation (C1) of the pyranose rings in the WSPSs investigated.

EXPERIMENTAL

PC was conducted on Filtrak FN-11, 7, 1 paper in the butanol—pyridine—water (6:4:3) system by the descending method. Aniline hydrogen phthalate was used to indicate the spots. The quantitative compositions of the monosaccharides were determined via the corresponding aldononitrile acetates [12]. The GLC of the derivatives was performed on a Tvset-101 instrument with a flame-ionization detector; steel column (0.3 x 200 cm), 5% of Silicone XE-60 on Chromaton NAW (0.200—0.250 mesh), column temperature 210°C, carrier gas helium at 60 ml/min.

The viscosities of 0.5% solutions of the galactomannans were measured in an Ostwald viscometer with a capillary diameter of 0.73 mm, the time of outflow of water, τ_0, being 49 s.

IR spectra were measured on a Perkin-Elmer system 2000 Fourier IR spectrometer in the form of tablets molded with KBr. The number of scans was 100. Specific rotations were determined on a Zeiss polarimeter in a tube 1 dm long with a volume of 10 ml.

The hydrolysis of the galactomannans was achieved with 2 N H_2SO_4 at 100°C for 8 h.

Isolation of the Galactomannans. Seeds (20—30 g of each species separately) were ground and boiled in chloroform—methanol (1:1) for 1 h. They were then extracted three times with water (in volume ratios of 1:15, 1:10, and 1:5) at room temperature for 4 h each time. The extract was concentrated to 300 ml and precipitated with three volumes of alcohol.

The fibrous precipitates obtained were separated off, washed with alcohol, and dried in vacuum over P_2O_5.

Qualitative Reactions of the Galactomannans. In drops, a 1% solution of lead acetate was added to 10.0 ml of a 0.5% solution of each of the galactomannans (**1—8**), giving a white pulverulent precipitate. The galactomannans were precipitated with barium hydroxide similarly.

Formation of Cu Complexes. Fehling's solution was added gradually, dropwise, to solutions of 0.5 g each of the galactomannans in 100 ml of water. The resulting precipitate was separated off by centrifugation. Copper ions were eliminated by washing with 70 and 7% aqueous solutions of acetic acid, and the residues were dewatered and dried in vacuum over P_2O_5. The yield of purified galactomannan was 0.28 g.

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