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Constituents of the Seed of *Malva verticillata*. I. Structural Features of the Major Neutral Polysaccharide*

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The seed of *Malva verticillata* L. (Malvaceae) is an Oriental crude drug (Japanese name, Toukishi) used as a diuretic, laxative, and galactopoietic. We have now isolated a pure neutral polysaccharide from the seeds of this plant. Its properties and structural features are reported here.

The seeds were homogenized and extracted with hot water. After addition of ethanol, the resulting precipitate was dissolved in water. After centrifugation, the supernatant was applied to a column of DEAE-Sephadex A-25 (carbonate form). The eluate with water was purified by gel chromatography with Sephadex G-25, then applied to a column of Con A-Sepharose. The eluate with a phosphate buffer was dialyzed, concentrated and purified by gel chromatography with Sephadex G-25, then the eluate obtained was lyophilized.

The polysaccharide gave a single peak on gel chromatography with Cellulofine GCL-2000 m. In addition, it gave a clear band on polyacrylamide gel disk electrophoresis. It had $[\alpha]_D^{24}-13.9°$ (H_2O, $c=0.14$). Gel chromatography with standard pullulans gave a value of 77000 for the molecular weight. The polysaccharide is designated as MVS-I.

As component sugars of MVS-I, L-arabinose, D-galactose, and D-glucose were identified. Quantitative determination showed that the polysaccharide contained 16.1% arabinose, 39.4% galactose, and 44.5% glucose, and that their molar ratio was 3 : 6 : 7. No nitrogen was found in MVS-I. The NMR spectrum and IR spectrum of MVS-I showed no acetyl signal or absorption.

Methylation of the polysaccharide was performed with methylsulfinyl carbanion and methyl iodide in dimethyl sulfoxide. The methylated product was hydrolyzed, and the hydrolyzate was converted into the partially methylated alditol acetates. GLC-MS revealed derivatives of 2,3,5-tri-*O*-methyl-L-arabinose, 2,3-di-*O*-methyl-L-arabinose, 2,4,6-tri-*O*-methyl-D-galactose, 2,3,6-tri-*O*-methyl-D-galactose, 2,4-di-*O*-methyl-D-galactose, 2,3,4,6-tetra-*O*-methyl-D-glucose, and 2,4,6-tri-*O*-methyl-D-glucose as the products in a molar ratio of 3 : 3 : 2 : 6 : 4 : 1 : 13 from MVS-I.

The ^{13}C-NMR spectrum of MVS-I showed five signals due to anomeric carbons at δ 111.889, 110.108, 107.033, 105.845, and 105.225 ppm. The first two signals were assigned to the anomeric carbons of α-L-arabinofuranose and the signals at 107.033 and 105.845 to

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the anomeric carbons of β-D-galactopyranose residues. The last signal was assigned to the anomeric carbons of β-D-glucopyranose.

MVS-I was subjected to periodate oxidation followed by reduction with sodium borohydride. The product from MVS-I contained galactose and glucose in a molar ratio of 6 : 13. None of arabinose was found in the product. Most (93%) of the glucose residues survived after periodate oxidation, while the half of galactose residues were decomposed by this treatment.

Based on the accumulated evidence described above, it can be concluded that the minimal repeating unit of MVS-I is composed of seven kinds of component sugar units as shown in Chart 1.

MVS-I is essentially arabinogalactoglucans. The ratio between arabinose and galactose is 1 : 2 in MVS-I, and the polysaccharide has 1,5-linked α-L-arabinofuranose units and 3,6-branched β-D-galactopyranose residues like typical arabino-3,6-galactans. Further, MVS-I possesses a characteristic glucan structure.

	↓ 6		
(four)[a]	→3 β-D-Gal*p* 1→	(thirteen)[a]	→3 β-D-Glc*p* 1→
(two)[a]	→3 β-D-Gal*p* 1→	(one)[a]	β-D-Glc*p* 1→
(six)[a]	→4 β-D-Gal*p* 1→		
(three)[a]	→5 α-L-Ara*f* 1→		
(three)[a]	α-L-Ara*f* 1→		

Gal*p*, galactopyranose ; Glc*p*, glucopyranose ;
Ara*f*, arabinofuranose

Chart 1. Component Suger Residues in the Minimal Repeating Unit in the Structure of MVS-I

a) Number of residues.