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BRIEF COMMUNICATIONS

LAMIACEAE CARBOHYDRATES. III. WATER-SOLUBLE POLYSACCHARIDES FROM *Origanum vulgare*

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We continued our study of carbohydrates from plants of the family Lamiaceae [1] with a preliminary investigation of free carbohydrates and water-soluble polysaccharides from the aerial part of *Origanum vulgare* L.

Raw material (500 g) was treated successively with hexane, $CHCl_3$, and ethanol (80%). The alcohol extract was concentrated to an aqueous residue. Phenolic compounds were removed using lead acetate. The solution was passed successively through columns of polyamide, cation-exchanger KU-2-8 (H^+-form), and anion-exchanger ASD-4p (Cl^--form). The final effluent was concentrated and analyzed by PC (FN-12, 80% isopropanol, descending chromatography, developer diphenylamine phosphate in butanol, 1%) and HPTLC [1]. Glucose, fructose, and saccharose were detected (Table 1). The total content of free carbohydrates (FC) in the aerial part of *O. vulgare* was 6.60% (anthrone method [2]).

After removal of components soluble in alcohol, the raw material was extracted with water at 20 and 100°C. The aqueous extracts were concentrated, dialyzed, and precipitated with ethanol (95%) to afford two fractions of water-soluble polysaccharides (WSPS) $WSPS_C$ and $WSPS_H$ (for extraction at 20 and 100°C, respectively). These were colored amorphous substances that did not give a reaction with iodine and contained significant amounts of N and ash elements (Table 2).

Then fractions $WSPS_C$ and $WSPS_H$ were hydrolyzed (TFA, 2M, 100°C, 6 h). The monosaccharide composition was determined by PC and HPTLC. The dominant monosaccharides of both fractions were Gal, Ara, and Glc in 3.9:2.1:1.0 and 1.3:0.5:1.0 ratios for $WSPS_C$ and $WSPS_H$, respectively (Table 1). The IR spectrum (Perkin—Elmer IR-Fourier spectrometer, film on KRS-5 glass) contained absorption bands for α- and β-bonds.

Demineralization over cation-exchanger KU-2-8 (H^+-form) and deproteination by pronase [3] of $WSPS_C$ and $WSPS_H$ produced fractions $WSPS_{C'}$ and $WSPS_{H'}$ the hydrolysates of which contained Gal, Ara, and Glc in 3.6:1.8:1.0 and 1.2:0.3:1.0 ratios for $WSPS_{C'}$ and $WSPS_{H'}$, respectively (Table 1). Gel chromatography over Sephadex G-100 (1.5 × 50 cm column, 0.3% NaCl eluent) indicated that both fractions were homogeneous. IR spectra of $WSPS_{C'}$ and $WSPS_{H'}$ were typical and contained the following absorption bands (cm^{-1}): $WSPS_{C'}$, 3391, 2921, 1638, 1414, 1146, 1076, 916, 890, 840; $WSPS_{H'}$, 3404, 2934, 1621, 1412, 1134, 1097, 917, 894, 841. Absorption bands of α- and β-bonds were observed in the fingerprint region (Table 2).

TABLE 1. Qualitative and Quantitative Composition of FC and WSPS from *Origanum vulgare*

Fraction	Yield, %	Monosaccharide composition, %						
		Ara	Frc	Gal	Glc	Rha	Sac	Xyl
FC	6.60	-	12.4	-	84.3	-	3.2	-
$WSPS_C$	4.86	29.1	-	53.1	13.7	1.4	-	2.6
$WSPS_{C'}$	0.07	27.4	-	54.7	15.1	0.3	-	2.4
$WSPS_H$	0.63	15.4	-	44.7	34.2	2.1	-	3.5
$WSPS_{H'}$	0.43	12.0	-	43.2	36.9	1.9	-	5.9

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TABLE 2. Physicochemical Properties of WSPS from *Origanum vulgare*

Fraction	$[\alpha]_D^{20}$, ° (*c* 0.7, water)	Content, %			MW, kDa	ν_{max}, cm^{-1}	
		carbohydrates	N	ash		α-bond	β-bond
$WSPS_C$	+17	4.43	12.39	35.94	-	917, 840	893
$WSPS_{C'}$	+54	98.12	-	<0.1	49	916, 840	890
$WSPS_H$	+21	57.13	8.14	14.73	-	918, 840	895
$WSPS_{H'}$	+67	97.93	-	<0.1	60	917, 841	894

TABLE 3. Biological Activity of Polysaccharide Fractions from *Origanum vulgare*

Fraction	Binding of atherogenic lipids, %	IC_{50} with osmotic hemolysis, mg/mL	IC_{50} (DPPH), mg/mL
$WSPS_C$	16.01±0.08	0.1864	-
$WSPS_{C'}$	28.31±0.10	0.1054	0.244±0.009
$WSPS_H$	18.54±0.09	0.2841	-
$WSPS_{H'}$	21.20±0.09	0.1127	0.267±0.011
Heparin	100	-	-
Caffeic acid	-	0.7621	0.084±0.005

Antiatherogenic (binding of atherogenic lipoproteides of blood serum [1]), membrane-stabilizing (osmotic hemolysis [1]), and antiradical (DPPH method [4]) activity was observed for the WSPS from *O. vulgare* using *in vitro* methods (Table 3).

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