

CHEMICAL COMPOSITION OF DRY EXTRACTS FROM *Alcea rosea*

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Dry extracts from roots and stems of Alcea rosea were prepared. Their chemical composition was studied by characterizing the carbohydrate, protein, and elemental compositions.

Key words: *Alcea rosea* L., dry extract, polysaccharide, mucus, pectinic substances, hemicellulose, proteins, elemental composition, expectorant.

The search for new plant polysaccharides is exceedingly critical because of, on one hand, the need to expand the number of mucous preparations and, on the other, the paucity of known sources.

In this respect, mucus-containing plants of the Malvaceae family are very interesting as sources of carbohydrates especially because some of them are used in medicine. For example, the tincture, liquid extract, tea, and powder of *Althea officinalis* L. root are used as expectorants [1].

The goal of our work was to prepare dry extracts from the aerial part and roots of *Alcea rosea* and to study their chemical composition. The raw material for preparation of the dry extract was stems and roots collected in Syrdarinsk District of Uzbekistan.

A. rosea is a perennial herbaceous plant that is cultivated as a medicinal and decorative. It grows wild in Uzbekistan in warm regions on plains and hills, along the edges of forests, in meadows, and in large river basins [2]. Food dyes are obtained from flower petals of *A. rosea* [3].

Plant material from stems and roots was extracted separately with hot water to isolate biologically active compounds. Then the extract was filtered, condensed in vacuo, and dried in a spray drier by nozzle spraying. The yield of dry extract from stems was 18%; from roots, 22%.

Pectinic substances (PS), the content and monosaccharide composition of which are shown in Table 1, were isolated after extraction with hot water in order to recycle the plant wastes.

Table 1 shows that the PS content in stems was greater than in roots. After reprecipitation by alcohol, demineralization, and drying, PS from stems were a cream-colored powder that was soluble in water to form a colloidal solution with $[\alpha]_D^{20} +170^\circ$ (*c* 0.5, water). This indicated that the glycoside bond had the α -configuration between uronic acid units. The content of uronic acids was 49%. IR spectrum (ν , cm^{-1}): 3600-3200, 2500, 1640, 1350, 1265, 1230, 1100, 830, 710; similar to spectra of PS from higher plants [4]. Pectin consisted of arabinose, glucose, rhamnose, galactose, xylose, and galacturonic acid, which formed the main chain.

Hemicellulose (HC) from stems (26.6%) and roots (14.1%) was a light-brown amorphous powder. Aqueous solutions of HC did not give a reaction with starch. The hydrolysate contained galactose, glucose, xylose, arabinose, rhamnose, and galacturonic acid. Treatment of the plant with base probably extracted PS that were insoluble upon extraction by ammonium oxalate.

Dry extracts were amorphous hygroscopic powders with a yellow tint, without an odor, and with a mild mucous and sweet taste. Dry extracts were analyzed by the literature methods [5].

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TABLE 1. Monosaccharide Content and Composition in PS from Various Organs of *A. rosea*

Organ	Polysaccharide type	Yield, %	Sugar ratio					
			Rham	Ara	Xyl	Glc	Gal	UA
Stems	PS	4.3	4.5	1.0	Tr.	Tr.	4.2	+
Roots	PS	2.3	18.0	3.0	Tr.	Tr.	1.0	+

TABLE 2. Microelement Composition of Dry Extracts from Stems and Roots of *A. rosea*

Element	Amount, mg/kg		Element	Amount, mg/kg	
	stems	roots		stems	roots
Pb	30.1	26.1	Sn	23.1	18.5
Cd	0.67	0.83	Cr	196.0	200.5
Cu	30.0	24.2	Fe	106.0	79.0
Zn	93.6	37.5			

Samples of dry extracts from stems and roots were dissolved in water to form viscous mucous solutions. Treatment with ammonia or NaOH solution gave a yellow color (presence of mucus). The extracts did not contain starch according to a negative test with iodine.

The dry extract of stems and roots contained 16 and 20% water-soluble polysaccharides (mucus, WSPS), respectively. The latter were isolated by dissolution of dry extracts at room temperature followed by precipitation by alcohol.

The alcoholic mother liquor was evaporated to a syrup. Paper chromatography (PC) detected glucose, galactose, arabinose, and oligosaccharides at R_f values of saccharose.

PC and GC of the products from total acid hydrolysis of mucous polysaccharide from dry extracts of stems identified rhamnose, arabinose, glucose, and galactose in a 17.6:2:1:2.8 ratio, respectively, in addition to galacturonic and glucuronic acids. The mucus hydrolysate from dry extracts of roots also contained rhamnose, arabinose, glucose, and galactose in a 20:2:1:1 ratio and the same uronic acids. Rhamnose predominated in mucus samples.

We used atomic absorption spectrometry to study the microelement composition and determine their quantitative content in dry extracts of stems and roots. Table 2 gives the results and shows that the dry extracts contained seven microelements.

The contents of Fe, Zn, and Cr were greatest of all observed microelements. The Cd contents were similar for both extracts.

Thus, according to the results the content of the determined microelements can be an indicator of the species because the plants absorb from the soil those elements that are needed for its metabolism and variations in the quantitative content depend on the habitat and plant variety.

The protein contents in the dry extracts that were determined by the Kjeldahl method [6] were 11.3% for stems and 12.1% for an analogous root sample.

The amino-acid composition of the protein was determined after acid hydrolysis and consisted of 17 amino acids (Table 3).

All essential amino acids such as valine, threonine, methionine, isoleucine, leucine, lysine, phenylalanine, histidine, and arginine, which are not synthesized in children, were observed in the amino-acid composition. Samples had balanced essential amino acids. The amino-acid composition of protein from the samples had predominantly asparagine, glutamine, and leucine. The high lysine content indicated that proteins from the studied dry extracts of *A. rosea* had high food value. The total amounts of amino acids in the samples were similar.

Thus, the study of the chemical composition of dry extracts of *A. rosea* stems and roots found monosaccharides, oligosaccharides, mucus, microelements, and proteins. Data for the dry extract enabled stems and roots to be standardized for content of mucous polysaccharides. The combined components suggest that the dry extracts can be used as medicinal preparations to treat inflammatory diseases, catarrh of upper respiratory tracts, bronchial asthma, etc.

TABLE 3. Amino-Acid Composition of Proteins of Dry Extracts from Stems and Roots of *A. rosea*

Amino acid	Content, wt. %		Amino acid	Content, wt. %	
	stems	roots		stems	roots
Asp	5.7	8.2	Met	-	0.6
Thr	3.0	3.4	Ile	0.2	1.4
Ser	2.1	2.2	Ze	8.8	5.5
Glu	11.3	11.5	Tyr	2.3	2.6
Pro	3.2	3.0	Phe	3.5	3.7
Gly	1.1	1.2	His	1.8	1.9
Ala	3.4	3.2	Lys	7.7	6.8
Cys	2.7	-	Arg	3.4	3.5
Val	3.5	3.2	Without tryptophan	Σ63.7%	Σ61.9%

Pharmacological investigations of the dry extracts were performed at the Department of Pharmacology, Tashkent Pharmaceutical Institute, under the direction of Prof. Kh. U. Aliev.

The extract obtained from roots is called Alceum and is approved as 0.2-g tablets for medical use as an expectorant [7].

The dry extract of stems was sent for preclinical pharmacological investigations as an anti-ulcer agent to treat stomach and small-intestine ulcers.

EXPERIMENTAL

Solutions were evaporated in vacuo at $45 \pm 5^\circ\text{C}$. Descending paper chromatography used FN 1 and 11 paper. Chromatography used solvent systems (volume ratio) butan-1-ol:pyridine:water (6:4:3). Sugar was developed by anilinium biphthalate solution and urea (5%) for 10 min at 105°C . Sugars were identified using markers.

GC (gas chromatography) was performed on a Chrom-5 instrument with a flame-ionization detector and a stainless steel column (200×0.01 mm), 5% Silicone XE-60 on Chromaton NAW-0.200-0.255 mesh, 210°C , N_2 carrier gas, 60 mL/min, using aldonitrile acetates. The aldonitrile acetates were prepared as before [8].

IR spectra were recorded on a Perkin—Elmer Model 2000 IR-Fourier spectrometer in KBr disks (5 mg compound per 200 mg KBr).

The content of uronic anhydride was determined by the literature method [9].

Macro- and microelement compositions of dry extracts were determined on a Perkin—Elmer (USA) Analyst 200 atomic absorption spectrometer. Table 2 gives the composition.

Amino-acid composition of protein was determined after acid hydrolysis (5.7 N HCl for 24 h at 110°C) on a T-339 amino-acid analyzer (Czech Rep.). Table 3 gives the results.

Preparation of *A. rosea* Dry Extract. Ground raw material (2.5 kg stems) was loaded into a 30-L extractor, treated with water (25 L), heated to 50°C , and soaked for 30 min, and extracted for 30 min. The extraction was repeated twice under analogous conditions. All three extracts were combined, filtered through cloth on a suction filter, and evaporated in a rotary evaporator at $40\text{--}50^\circ\text{C}$. The volume of the condensed extract was 20% of the initial volume.

The condensed extract was dried in an Angidro (Denmark) spray drier. The extract was loaded into the drying chamber by a Zalimp (Poland) peristaltic pump at flow rate 6 L/h. The temperature of the heat-transfer fluid at the entrance was 180°C ; at the exit, 95°C . The pressure of the heat-transfer fluid was 0.15 MPa.

The yield of dry extract from stems was 0.45 kg (18%).

Ground roots (2.5 kg) treated analogously as above afforded dry extract with a yield of 0.55 kg (22%).

PS and HC were isolated successively from one portion of raw material pulp after isolation of WSPS using a mixture of oxalic-acid solution (1%) and ammonium oxalate (1:1) at 70°C and base (10% aqueous). Two extractions were usually performed. A ratio of 1:10 was used for the first; 1:5, the second. Polysaccharides from the acidic and basic extracts were precipitated by alcohol (1:3).

The isolated polysaccharides were rinsed with 80° and then 96° alcohol and dried to afford the corresponding PS and HC. Table 1 gives the yields.

Total acid hydrolysis of polysaccharides was carried out using H₂SO₄ (2 N) at 100°C for 8-16 h. The hydrolysates were treated with BaCO₃ until neutral, filtered, evaporated, and analyzed by PC and GC.

Determination of WSPS Content (Mucus) in Dry Extract. Dry extract (5 g) of stems was dissolved in water (150 mL) at room temperature and centrifuged. Polysaccharide (mucus) was precipitated by alcohol (1:3). The precipitate was washed with alcohol and acetone and dried. Yield, 16%.

Polysaccharide (mucus) was prepared analogously from dry extract of roots in 20% yield.

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