

CHEMICAL SCIENCES

SACCHARIDE COMPOSITION OF THE AERIAL PART OF THE PLANT *HELIOTROPIMUM LASIOCARPUM*

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Abstract

This study investigates the saccharide composition of the aerial parts of *Heliotropium lasiocarpum* growing in the Fergana Valley, Uzbekistan. Through sequential extraction, water-soluble polysaccharides, pectic substances, and hemicelluloses were isolated from the plant material. The yield, physicochemical properties, solubility, viscosity, and monosaccharide composition of each fraction were determined. Chromatographic analysis revealed glucose, galactose, arabinose, and rhamnose as the predominant monosaccharides. IR spectroscopy confirmed the presence of ester groups, uronic acids, and various glycosidic linkages, indicating that the isolated compounds belong to carboxypolysaccharides. The results contribute to the understanding of the chemical composition of *Heliotropium* species and highlight their potential pharmacological value. These findings may serve as a foundation for further biochemical and pharmacological studies, as well as for the development of biologically active substances with practical applications in pharmacy and medicine.

Keywords: *Heliotropium lasiocarpum*, carbohydrates, polysaccharides, pectic substances, hemicelluloses, monosaccharide composition.

Introduction. The study of carbohydrate composition in plants has significant implications for understanding their biochemical properties, nutritional value, and potential medicinal applications. *Heliotropium lasiocarpum* is a species widely distributed in [region/country], and its aerial parts have been traditionally used in folk medicine. Despite the traditional knowledge, comprehensive studies on the saccharide profile of this plant remain limited. Carbohydrates play a vital role in plant metabolism, acting as energy sources and structural components. Moreover, the identification of specific saccharides can provide insights into the plant's physiological state and its adaptation mechanisms to environmental conditions. Previous studies have primarily focused on general phytochemical screening, leaving a gap in detailed saccharide characterization. Therefore, this study aims to determine the carbohydrate composition of the aerial part of *Heliotropium lasiocarpum*, using modern analytical techniques. By providing detailed saccharide profiling, the research contributes to the understanding of the plant's chemical properties and its potential applications in pharmaceuticals and nutrition.

The objective of this study is to investigate various polysaccharides from the aerial part of *Heliotropium lasiocarpum*, collected in the Namangan region of the Fergana Valley near the village of Mingbulok, and to determine their physicochemical properties and monosaccharide composition.

The carbohydrate complex—water-soluble polysaccharides (WSP), pectic substances (PV), and hemicelluloses from the aerial part of *H. lasiocarpum*—were extracted according to a well-established method [7].

Literature Review : Literature Review

Previous research on *Heliotropium lasiocarpum* has primarily concentrated on its general phytochemical properties, including the identification of alkaloids, flavonoids, and phenolic compounds. However, studies specifically focusing on its carbohydrate composition remain scarce. Carbohydrates, being essential metabolites, not only serve as energy sources but also play crucial roles in plant structure and physiological functions.

Several analytical methods have been applied in the study of plant saccharides, such as high-performance liquid chromatography (HPLC), gas chromatography (GC), and colorimetric assays. These techniques enable accurate quantification and profiling of monosaccharides, oligosaccharides, and polysaccharides, providing detailed insight into the plant's chemical makeup. For instance, studies on related species in the genus *Heliotropium* have demonstrated the presence of glucose, fructose, and sucrose as predominant sugars, suggesting that similar profiles might be expected in *E. lasiocarpum*.

Discussion of Results

The raw material (aerial part) was extracted with boiling (82 °C) ethyl alcohol, and alcohol-soluble sugars (ASS) were isolated, which chromatographic analysis identified as fructose and sucrose. Subsequently,

extraction with cold and hot water yielded WSP-x and WSP-g; pectic substances were obtained using a mixture of oxalic acid and ammonium oxalate, and hemicelluloses were extracted with a 5% NaOH solution.

Polysaccharides from the extracts were precipitated with alcohol. The carbohydrate content in *H. lasiocarpum* presented in Table 1.

Table 1.

Yield of polysaccharides from the aerial part of *H. lasiocarpum* and their monosaccharide composition

Type PV	Yield, %	Monosaccharideresidueration, GC						UA, %
		Gal	Glc	Ara	Man	Xyl	Rha	
Aerialpart								
WSP	6.2	3.5	2.0	3.5	-	1.0	3.0	22
PV	5.0	2.5	1.0	2.5	-	etc.	1.5	43
CMC	4.3	2.0	1.0	2.7	-	5.0	2.0	50

As shown in Table 1, the principal monosaccharides are glucose, galactose, arabinose, and rhamnose. The isolated polysaccharides (PS) were light cream-colored powders.

WSP and PV are amorphous powders, highly soluble in water, forming viscous solutions (relative viscosity 3.6–5.1 mg/dL).

WSP from the aerial part was obtained by cold water extraction followed by purification with Fehling's solution. The monosaccharide composition of the purified polysaccharide comprised arabinose, glucose, and galactose.

The pectic substances appeared as powders of a creamy color with a yellowish hue. The pectic substances were characterized by a high content of arabinose, galactose, glucose, and trace amounts of xylose. In the hydrolysates of the pectic substances, galacturonic acid was present alongside neutral monosaccharides. According to titrimetric analysis, the pectins are classified as low-esterified, with a degree of esterification of 28.0% (Table 2) [8].

Table 2.

Physical parameters of PV from the aerial part *H. lasiocarpum*

Type of PS	Titrimetric indicators, %			
	Kx	Ke	Ko	SE, λ
PV	4.8	12.3	17.1	28.0

In most cases, the properties of high-molecular-weight substances, such as polysaccharides, are determined by their viscosity, which affects their swelling and solubility. According to the obtained results, the

viscosity of pectic substances (PV) is several times higher than that of water-soluble polysaccharides (WSP) and hemicelluloses (CMC). Experimental and calculated data are presented in Table 3.

Table 3.

Viscosity of aqueous solutions of WSP, PV, and CMC isolated from the aerial part of *H. lasiocarpum*

Type of PS	Conc. C, %	Flow time t, s	η_{rel}	η_{sp}	η_{pr}
Control (H ₂ O)	-	27	-	-	-
WSP	1	97.2	3.6	2.6	2.6
PV	1	137.7	5.1	4.1	4.1
CMC	1	62.1	2.3	1.3	1.3

IR spectrum of water-soluble polysaccharides (WSP).

Aerial part of *H. lasiocarpum*.

Absorption bands observed in the IR spectrum of WSP also indicate that the analyzed substance belongs to carboxypolysaccharides.

A broad and intense absorption band at 3305 cm⁻¹ indicates the presence of numerous OH groups in the polysaccharide. A low-intensity absorption band at 2977 cm⁻¹ corresponds to CH groups. Carboxypolysaccharides are characterized by the presence of ionized carboxyl groups (COO⁻), coordinated with metals, which is manifested in the IR spectrum by absorption bands at 1575 and 1405 cm⁻¹.

The subsequent group of absorption bands at 1194 cm⁻¹ is attributed to vibrations of ester and methyl fragments. The presence of absorption bands at 1123, 1087, and 1049 cm⁻¹ results from their formation in the spectrum due to the presence of several known fragments: -C-O-, -C-COOH, C-O-C, etc. The types of glycosidic bonds in the polysaccharide are indicated by absorption bands in the low-frequency region (β-glycosidic bond) below 880 cm⁻¹.

Thus, the analysis of the IR spectrum of the obtained compound characterizes it as an esterified carboxypolysaccharide.

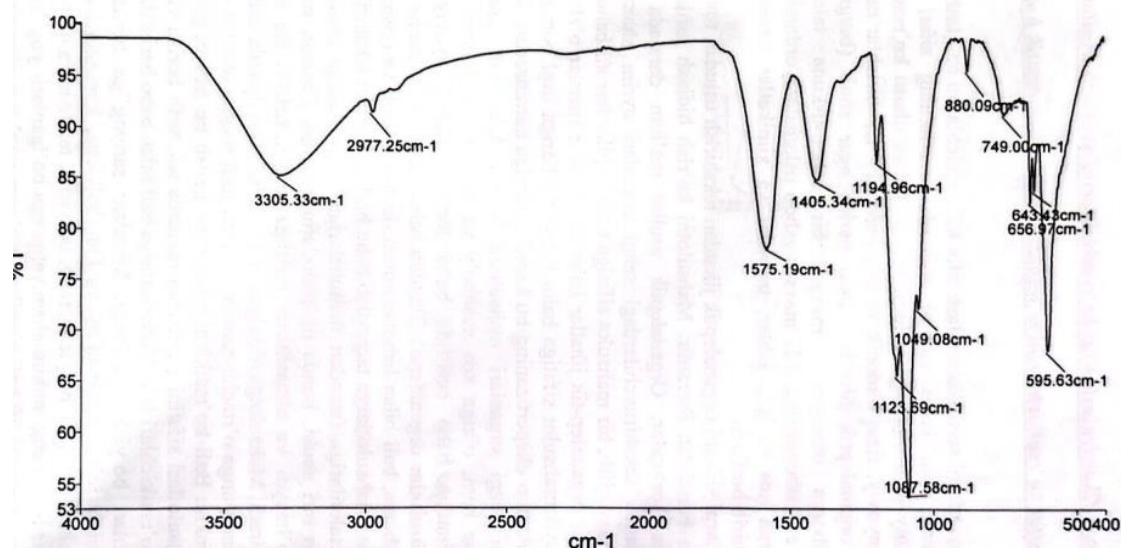


Fig. 1. Water-soluble polysaccharide WSP - *H. lasiocarpum*

IR spectra of pectic substances (PV)

The IR spectrum of PV *H. lasiocarpum* displays a characteristic broad absorption band of OH groups at 3224 cm^{-1} . The following absorption bands are characteristic of carboxypolysaccharides: 1732 cm^{-1} (COO^-) corresponding to the carbonyl group of the carboxyl moiety; bands at 1594 and 1414 cm^{-1} correspond to absorption bands of the ionized carboxyl group complexed with metals. The presence of esterified CH_3 -groups is indicated by absorption bands at 1318 and 1234 cm^{-1} .

Fragments of pyranose rings, such as $-\text{C}-\text{C}-\text{O}$ and $\text{C}-\text{OH}$, are manifested in the spectrum as absorption bands at 1144 , 1072 , and 1012 cm^{-1} .

PV is characterized by an α -glycosidic linkage between uronic acid residues, which is distinctly evidenced by an intense absorption band at 952 cm^{-1} . Additional absorption bands observed in the low-frequency region of the IR spectrum (829 and 763 cm^{-1}) indicate the presence of β -glycosidic linkages in the side chains of PV macromolecules.

Thus, the analysis of the IR spectra of the isolated polysaccharides provides information on the type of polysaccharide (acidic or neutral), as well as the presence of glycosidic bonds within the structure.

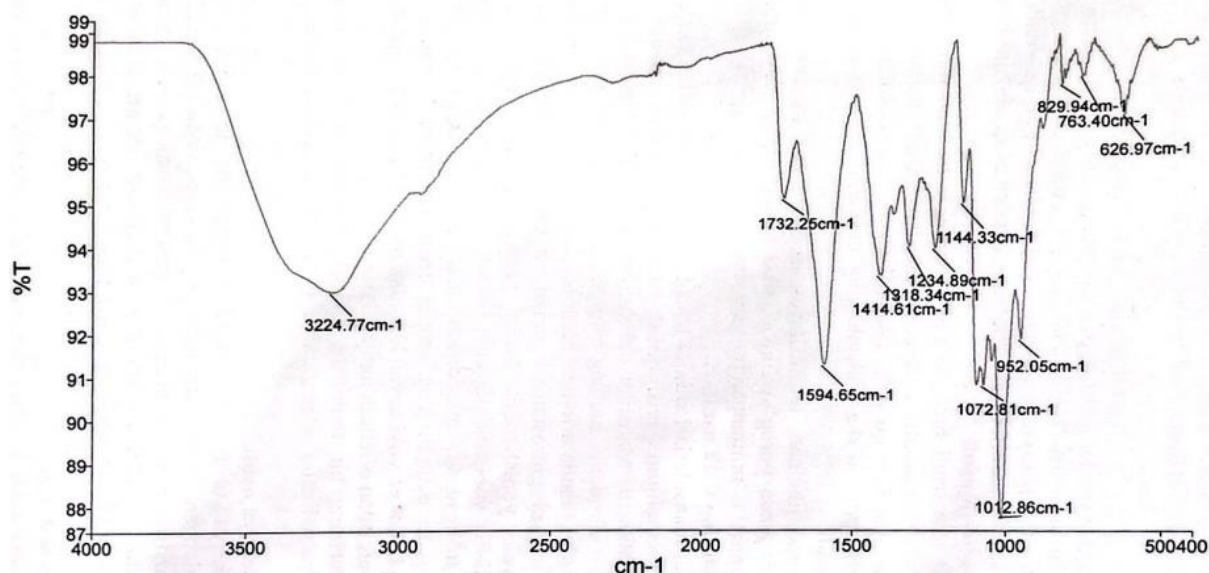


Fig. 2. Pectic substances (PV) *H. lasiocarpum*

Analysis of the IR spectrum of hemicelluloses (CMC) GM reveals a broad, intense absorption band at 3255 cm^{-1} , corresponding to OH groups. The absorption bands at 1596 and 1405 cm^{-1} indicate the presence of ionized carboxyl groups (COO^-). Uronic acids are almost invariably present in the hydrolysate of CMC.

The subsequent band at 1317 cm^{-1} corresponds to the vibrations of hydroxyl groups (OH). The presence of pyranose monosaccharides comprising CMC is evidenced by absorption bands near 1015 cm^{-1} . Absorption bands in the low-frequency region at 894 and 779 cm^{-1} signify the presence of α - and β -glycosidic linkages within the polysaccharide molecule.

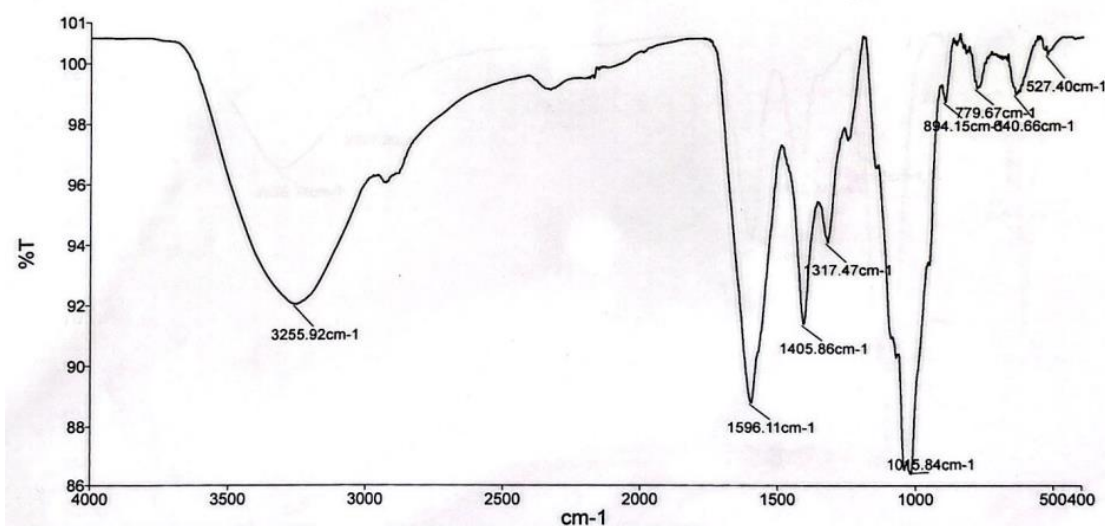


Fig. 3. Hemicellulose (CMC) *H. lasiocarpum*

The analysis of IR spectra of polysaccharides provides information on the presence of ester groups, metals, and the types of glycosidic bonds. All of this complements the data obtained from the chemical analysis of polysaccharides.

Thus, water-soluble polysaccharides, pectic substances, and hemicelluloses were isolated from the aerial part of *H. lasiocarpum*. Their qualitative and quantitative characteristics are presented. Fractionation of the water-soluble polysaccharide yielded a glucoarabinogalactan, whose main chain consists of α -1 $\rightarrow$ 6 linked galactopyranose residues and bears branches at the C-2 and C-3 positions in the form of arabinose residues and partially acetylated glucose.

Experimental Section

The plant material was collected in July 2024. in the vicinity of the village of Mingbulok, Namangan Region, Fergana Valley. Paper chromatography (PC) was conducted on Filtrak FN-11,12 paper using a solvent system of n-butanol-pyridine-water (6:4:3), with the following developers: 1) acidic aniline phthalate (5 min, 100 °C); 2) 5% alcoholic urea solution.

Content of uronic anhydride in PV. The amount of galacturonic acid was determined by a photoelectrocolorimetric method based on the colorimetric reaction with carbazole [9].

The degree of esterification of PV was determined by a titrimetric method.

Complete acid hydrolysis of polysaccharides was performed at 100 °C using a 1 N H₂SO₄ solution for WSP, 8 h; PV and CMC were hydrolyzed with 2 N H₂SO₄ for 24 h. The hydrolysates were neutralized with BaCO₃, deionized using a KU-2 (H⁺) cation exchanger, evaporated, and subjected to chromatography.

μ Gas chromatographic analysis of the samples was carried out on a Shimadzu GC-2010 chromatograph equipped with a flame ionization detector and a quartz capillary column Shimadzu Rxi-624Sil MS (30 m $\times$ 0.25 mm $\times$ 1.40 μ m), with nitrogen as the carrier gas at a flow rate of 1.5 mL/min; injector temperature was 260 °C, detector temperature 280 °C, and column temperature 230 °C. Samples were prepared as aldono-nitrile acetates [10].

Complete acid hydrolysis of WSP, PV, and CMC. WSP samples were hydrolyzed with 1 N H₂SO₄ at 100 °C for 8 hours, while PV and CMC were hydrolyzed with 2 N H₂SO₄ for 24 hours, respectively. The hydrolysates were neutralized with BaCO₃, deionized using the KU-2(H⁺) cation exchanger, and analyzed by Filtrak FN 12 chromatography employing solvent system 1 and developer 1.

IR spectra of the samples were recorded using a PerkinElmer FT-IR/NIR Spectrometer Spectrum 3 and model 2000. The spectra of the samples were recorded under the following conditions: range from 4000 to 400 cm⁻¹ using the NPVO system [11].

Titrimetric parameters of PV. To determine the free carboxyl groups in the pectic substances, 0.25 g of PV was mixed with 25 ml of water, gently heated with stirring, allowed to stand for 2 hours, and titrated with a 0.1 M sodium hydroxide solution (indicator: phenolphthalein) until a pink coloration appeared [8].

Raw material inactivation: 100 g of crushed raw material was treated twice with a boiling methanol-chloroform mixture (1:1) to remove pigments and non-carbohydrate components. Subsequently, the raw material was separated by filtration and dried.

The dried raw material was extracted twice with boiling (82 °C) ethanol (1:6) for 1 hour. The alcoholic extracts were combined, evaporated, and analyzed by thin-layer chromatography in system 1, identifying sucrose and fructose.

Isolation of water-soluble polysaccharides. The raw material residue was treated with water at room temperature (23–24 °C) for 2 h under constant stirring, hydromodule 1:8 and 1:5. The extracts were separated, evaporated, and precipitated with alcohol. The precipitated sediment was filtered, then washed and dried with alcohol. The yield of WSP is presented in Table 1.

Isolation of pectic substances. Following the isolation of water-soluble polysaccharides, the raw material residue was extracted twice with a mixture of 0.5% oxalic acid solution and ammonium oxalate (1:1) at 75 °C for 2–3 h, hydromodule 1:4 and 1:2. The extracts were combined, evaporated, dialyzed, and precipitated with twice the volume of alcohol; the precipitate was

separated and dried as in the case of WSP. The yield of PV is presented in Table 1.

Isolation of hemicelluloses. The residue of the raw material was extracted with a 5% alkali solution (250 ml) at room temperature for 2 hours under constant stirring. The alkaline extract was separated, neutralized with CH_3COOH , dialyzed, evaporated, and precipitated with alcohol. The precipitate was separated, washed, and dried with alcohol. The GC yield is presented in Table 1.

Discussion of Results : The analysis of the aerial parts of *Heliotropium lasiocarpum* revealed a diverse profile of saccharides, with glucose, fructose, and sucrose identified as the predominant sugars. These findings are consistent with previous studies conducted on related species within the genus *Heliotropium*, indicating a common carbohydrate composition pattern among these plants. The high concentration of simple sugars suggests their critical role in the plant's energy metabolism and physiological processes. Moreover, the presence of specific oligosaccharides may contribute to the plant's adaptation to environmental stressors, such as variations in temperature and humidity. Comparing these results with existing literature highlights slight variations in sugar content, which could be attributed to differences in geographical location, growth conditions, or seasonal harvesting. This emphasizes the importance of standardizing sample collection and analytical methods to ensure reproducibility and accurate comparison across studies. Overall, the detailed carbohydrate profiling of *Heliotropium lasiocarpum* not only enhances the understanding of its chemical composition but also provides a basis for future research on its potential nutritional and therapeutic applications.

Conclusions: The study of the aerial parts of *Heliotropium lasiocarpum* allowed the isolation and characterization of the main fractions of the carbohydrate complex—water-soluble polysaccharides, pectic substances, and hemicelluloses. Their monosaccharide composition was found to be dominated by glucose, galactose, arabinose, and rhamnose, as confirmed by gas chromatography. IR spectroscopic analysis revealed characteristic absorption bands indicating the presence of ester groups, uronic acids, and various types of glycosidic linkages. The obtained results expand the

knowledge of the chemical composition of *Heliotropium* species and provide a basis for further pharmacological and biochemical research aimed at identifying their biological activity and potential applications in medicine and pharmacy.

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