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VISION

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PHLOMOIDES CANESCENS COMPLEX OF CARBOHYDRATES DISTRIBUTED IN THE FERGHANA VALLEY

Hilolaxon Rustamjonovna Rahimova*;
Rustamjon Komiljono'g'li Gulomov**

*Trainee Researcher,
Tashkent Medical Academy,
UZBEKISTAN

Email id: hilolaxon.rahimzoda.87@bk.ru

**Biology Department,
Namangan State University,
Namangan, UZBEKISTAN
Email id: gulomovr92@mail.ru

ABSTRACT

The carbohydrate composition of Phlomoides canescens grown in Uzbekistan has been studied. The carbohydrate complex was isolated, the monosaccharide composition was established, and the IR spectra were studied (WSPS-C, WSPS-H, PS, HMS-A, HMS-B). The presence of water-soluble polysaccharides was established and it was shown that they are heterogeneous polysaccharides of the arabinogalactan type.

KEYWORDS: *Water-Soluble Polysaccharides, Pectin Substances, Hemicellulose, IR Spectroscopy, Neutral Sugars, Uronic Acids.*

INTRODUCTION

Currently, the world pays great attention to determining the species composition of medicinal plants, studying their biological properties, assessing their cenopopulations, identifying natural resources, scientifically substantiating population changes as a result of external influences and analyzing the causes of decline, as well as improving conservation and reproduction. In recent years, the growing demand for natural medicinal herbs has led to a reduction in plant stocks. Based on the foregoing, in order to identify and assess carbohydrates of the *Phlomoides canescens* species common in the Shohimardon region of the Fergana region, in april-may 2021, field studies were carried out in the villages of Iordon of the Shohimardon region. Distributed areas of the plant were identified and samples were taken from the vegetative and generative parts of the plant to study its composition (1-Figure). To isolate and evaluate the complex of carbohydrates from plant samples, the goal was to isolate them using IR spectra: WSPS-C, WSPS-H, PS, HMS-A, HMS-B methods.

Modern concise classification of plants.

Phlomoides canescens belongs to the genus *Phlomoides* Moench of the Lamiaceae family. Central Asia, the mountainous regions of Iran (Iran and Afghanistan) and the Mediterranean are the main centers of species diversity. In the flora of the Earth there are 150–170 species of this genus, in the flora of Central Asia there are 59 species, including 43 species in the flora of Uzbekistan [1,2].

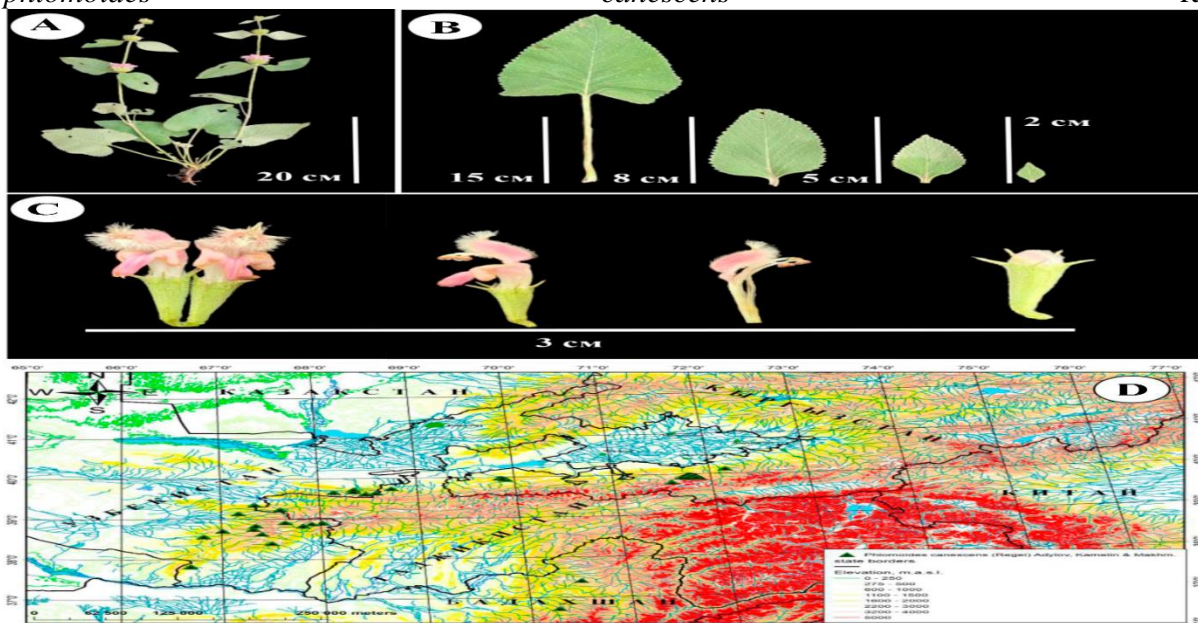
Bibliography. *Phlomoides canescens* (Regel) Adylov, Kamelin & Machmedov in *Opred. Rast. Sred. Azii* 9: 104 (1987). - *Phlomis canescens* Regel in *AHP*. 9 (2): 582 (1886); Popov in *Bull. Univ. As. Centr.* 13: 138 (1926); Knoring in *Fl. URSS*. 21:92 (1954); Vvedensky in *Fl. Uzbekistan*. 5: 355 (1961); Adylov, Kamelin, Makhmedov in *Opr. rast. Wed Az.* 9: 104 (1987) [4,6,7].

Type. TAJIKISTAN: Iskander - Kul, 16.06. [1870] O. Fedchenko (LE, lectotype, designated Rechinger 1982: 315) [5].

Life form and phenology. Hemicryptophytes, polycarpic Blossoms in june - july, bears fruit in july - august. [3].

Habitat. *Juniper* thickets, alpine subalpine meadows, thorny associations, middle and upper mountain belts, 1.600-2.800 m above [5].

Note. This species is close to *phlomoides oreophila*, characterized by leaves with stellate hairs at the top. The density and character of stellate hairs varies within the range of this species, therefore specimens with a low number of stellate hairs on the upper surface of the leaf cannot be distinguished as a separate species of *phlomoides tythaster*. Intermediates to *phlomoides oreophila* are found both at the edge and within the distribution area. They can be obtained as a result of continuous hybridization in areas of contact with *phlomoides oreophila* and the very recent invasion of more eastern and northern species of *phlomoides oreophilainto* the *phlomoides canescens* range



1-Figure. A- Phlomoides canescens species, B, C- Leaves and phenological organs structure, D- GIS distribution map.

The purpose of this work is to study the carbohydrate complex of *Phlomoides canescens*, to establish their monosaccharide composition.

Research methods. Field studies conducted as part of the tour were carried out in april-may 2021 in Shohimardon district of Fergana region (Uzbekistan). The total area of the tour was determined on the basis of international electronic databases Plants of the World Online (www.plantsoftheworldonline.org), Global Biodiversity Information Facility (www.gbif.org). The addresses given in the samples of existing herbarium holdings (LE, TASH, FRU, TAD, MW) were derived from the geographic coordinates of Google Earth, and a GIS map showing the general distribution of the species was prepared using ArcGIS 10.0.

Experimental part.

Inactivation of raw materials. 100 g of dried and crushed raw materials (usimlik nomi) were twice treated with boiling chloroform for 2 hours at a hydromodule of 1: 4 to remove dyes and low molecular weight substances. Then, alcohol-soluble sugars were extracted twice with boiling 82 ° ethanol (1: 4, 1: 3). The alcoholic extracts were separated by filtration, combined and evaporated to a small volume, and analyzed by paper chromatography (PS) in a 6: 4: 3 butanol-n-pyridine-water system. To identify spots, acidic aniline phthalate (1) was used to identify hexose and a 5% alcohol solution of urea (2).

Isolation of WRPS-H. The remainder of the raw material was extracted twice with cold water at room temperature for 1.5 h at a hydromodule of 1: 4, respectively. The extracts were separated by filtration, evaporated to a small volume, and precipitated with a threefold volume of ethyl alcohol. The precipitate that formed was centrifuged (5000 rpm, 10 min), washed, and dehydrated with alcohol. Output WSPS - 1 g.

Then the remainder of the raw material was twice extracted with water at a temperature of 80-85° for 1.5 hours at a hydromodule of 1: 3, 1: 2. The extracts were combined, evaporated and precipitated with alcohol. The formed precipitate was treated as described above. Output WSPS-g 1.2 g.

Isolation of pectin substances (PS). After the isolation of the total WSPS, the meal was twice extracted with an equal mixture of 0.5% solutions of oxalic acid and ammonium oxalate at a temperature of 75° C; the extraction was carried out at a hydromodule of 1: 4, 1: 3. The extract was separated by filtration, dialyzed against running water, evaporated, and precipitated with a threefold volume of alcohol. The precipitate was processed in the same way as described above. Output PS 6.2 g (from air-dry raw materials).

Isolation of HMS-A and HMS-B. After isolation, PS was treated twice with 5% KOH solution at room temperature, for 1.5-2 hours, with a hydromodule of 1: 3. The extracts were separated by filtration, neutralized with CH₃COOH, centrifuged to obtain a HMS-A precipitate with a yield of 1.0 g, the lagging solution was evaporated until thick and precipitated with a three-fold volume of alcohol. The precipitated HMS-B precipitate was separated by centrifugation, washed and dried with alcohol, yield 4.4 g.

Complete acid hydrolysis of polysaccharides. Samples of WSPS were hydrolyzed with 1N H₂SO₄ at 100°C, 8 hours, PS, HMS-A and HMS-B 2n H₂ SO₄, 100 ° C, 48 hours. The hydrolysates were neutralized with barium carbonate, deionized with KU-2 (H⁺) cation exchanger, and evaporated. The high-quality monosaccharide composition of PS was studied by BH using known witnesses on Filtrak FN-12 paper, in the system of butanol-pyridine water 6: 4: 3, developer 1.2. GC analysis of the samples was carried out on a Shimadzu GC-2010 chromatograph with a flame ionization detector, a Shimadzu Rxi-624Sil MS quartz capillary column 30mx0.25mmx1.40µm), the mobile phase rate (N₂) 1.5 ml / min, injector temperature 260 ° C, detector temperature 280 ° C and column temperature 230 ° C. The samples were taken in the form of aldonitrile acetates [2]. The IR spectra of the samples were recorded on an IR Fourier spectrometer, System 2000 (Perkin-Elmer) in KBr pellets. Number of scans 100.

RESULTS AND THEIR DISCUSSION

We sequentially isolated various polysaccharides from (nomi plants): alcohol-soluble sugars (ASS), water-soluble polysaccharides (WSPS), pectin substances (PS), hemicelluloses (HMS). SRS according to HR data (system 1, developer 1.2) are represented by glucose and fructose. Water-soluble polysaccharides (WSPS) were isolated in two ways: extraction of raw materials with cold water (WSPS-x), i.e. at room temperature and hot water at a temperature of 80-90 ° C (WSPS-g). Pectin substances (PS) were isolated with a mixture of 0.5% solutions of oxalic acid and ammonium oxalate, hemicellulose (HMS) - with a 5% KOH solution. The yield of polysaccharides and their monosaccharide composition are shown in Table 1.

TABLE 1. CONTENT OF VARIOUS GROUPS OF PHLOMOIDES CANESCENS POLYSACCHARIDES AND THEIR MONOSACCHARIDE COMPOSITION

Carbohydrate type	Output, %	Ratio of monosaccharide residues, GC						
		Rha	Ara	Xyl	Man	Glu	Gal	UAc,%
WSPS-x	1.0	3.0	25	7.0	4,3	10	30	25
WSPS-g	1,2	3.0	27	5	3	12	25	27
PS	6.2	-	16	2	-	7	15	60
HMS-A	1.0	7	30	3	-	-	20	40
HMS-B	4.4	2	25	10	-	-	18	45

As can be seen from Table 1, among the polysaccharides, PS are dominant (6.2%), with the HMS content being 5.4%, and the WSPS content in smaller amounts - 2.2%. WSPS are light beige amorphous powders, readily soluble in water. Monosaccharide compositions of WSPS did not differ sharply qualitatively, but the difference was in the quantitative ratio. The main monosaccharides of WSPS-x are Gal, Ara and Glu, and in WSPS-r- Ara, Gal; other monosaccharides are present in smaller amounts. Aqueous solutions of WSPS give a negative reaction to starch. The ratio of monosaccharides suggests that heterogeneous polysaccharides with a predominant content of glucans constitute the basis of WSPS-x, while both glucans and galactoarabinans may be present in WSPS-g.

In the IR spectra of WSPS-x and WSPS-g, the main absorption bands were found: 3339-3418 cm^{-1} (OH-groups), 1599-1613 cm^{-1} (HOH), 1415-1418 (C = O), 1074-916, 1018-911 cm^{-1} (α -glycosidic bond). Pectin substances are an amorphous cream-colored powder that partially dissolves in water to form a viscous solution.

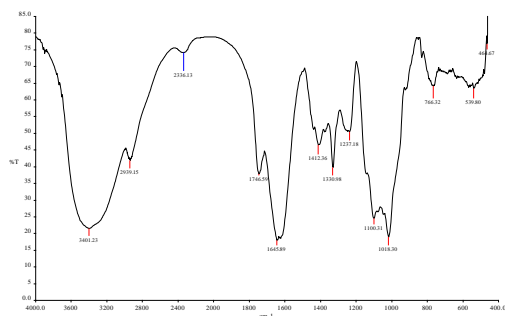
In the IR spectra of PS (Fig. 2), absorption bands were found: 3434 cm^{-1} (OH-groups), 1748 cm^{-1} (C = O free and COO- esterified carboxyl groups), 1434 cm^{-1} (vibrations of ionized carboxyl), 1365 cm^{-1} (-OCH₃), 827 cm^{-1} (α -glycosidic bond) [3,4].

The monosaccharide composition is represented by neutral and acidic monosaccharides. The largest amounts are found for arabinose and uronic acids, the content of the latter according to the carbazole method is 72% [5]. According to the IR spectrum, PS is an esterified polysaccharide. To determine the degree of esterification, titrimetric analysis was carried out, the results of which revealed the content of carboxyl and esterified groups: Kc (free carboxyl groups) - 8.1%, Ke (esterified carboxyl groups) - 9.0%. The data obtained correspond to the degree of esterification - 52.6%, which makes it possible to classify the studied PS as highly esterified pectins [6].

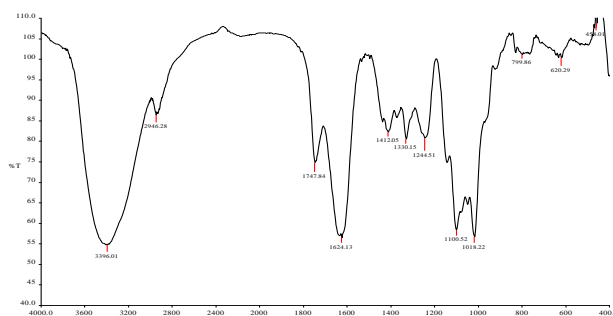
HMS-amorphous powder of light cream color, partially soluble in water, completely in dilute alkali solutions.

In the IR spectra of HMS, absorption bands were found: 3640 cm^{-1} (OH-groups), 1742 cm^{-1} (C = O), 1078 cm^{-1} (pyranose ring), 1588 cm^{-1} (COO⁻), 850 cm^{-1} (α -glycosidic bond) (Fig. 2).

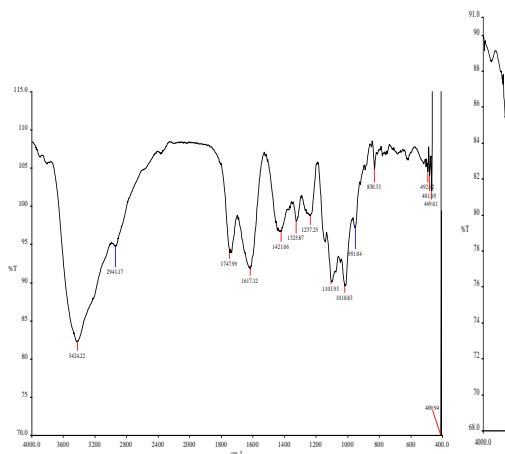
Based on the results obtained, it can be seen that the WSPS of *Phlomoïdes canescens* consist mainly of the following polysaccharides: arabinogalactans.



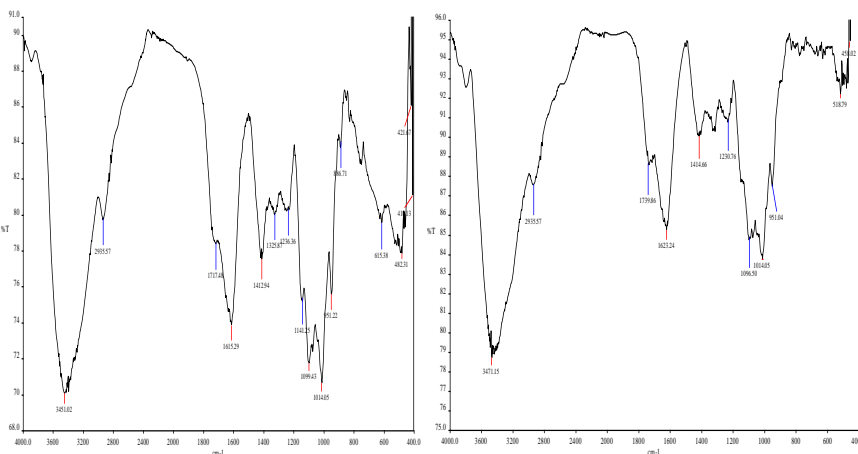
2-Figure. IR spectra - WSPS -x



3-Figure. IR spectra - WSPS -g



4-Figure. IR spectra - P5-Figure. IR spectra - HMS-A6-Figure. IR spectra - HMS-B



Conclusion

Alcohol-soluble sugars, water-soluble polysaccharides, highly esterified pectin substances and hemicelluloses were isolated from *Phlomis canescens*. Their qualitative and quantitative characteristics are given. The isolated polysaccharides were analyzed by IR spectroscopy.

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