
PHYTOCHEMICAL RESEARCH OF THE GENUS „GEUM L.“

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Abstract: The task of modern phytotherapy is to find promising medicinal plants in order to create new herbal medicines. Promising for this group are drugs based on plant material. Obviously, herbal medicinal products, to some extent, can be considered as an alternative to antibiotics, while some forms of bacterial infections caused by multiresistant strains of gram positive and gram negative bacteria resistant to other classes of antimicrobial agents and chemicals used to treat a variety of dosage forms. Among the biologically active substances /BAS/ that accumulate in plants and show pharmacological activity, tannins occupy an important place. Perspective to produce phytopreparations are species of the genus *Geum* L., the family Rosaceae L. There are 8 species naturally distributed in Bulgaria, 2 of which are protected by the Biodiversity Act. The most perspective are *Geum urbanum* L., *Geum rivale* L., *Geum rhodopaeum*. and also, a related species – *Geum montanum* L., which sprout in the southwestern region of Bulgaria. Phytochemical studies of water, alcohol and water extracts of acidified earth hydrochloric acid (leaves) and underground (rhizomes with roots) of *Geum urbanum* and *Geum montanum* for the presence of basic groups of biologically active substances have been found to contain a significant amount of tannins (hydrolyzed and condensed), insignificant The number of anthocyanins, flavonoids and coumarins in the studied vegetative organs. The phytochemical study of water extracts of the above ground and underground organs and ethanol extracts of underground bodies of *Geum urbanum* and *Geum montanum* has established the content of polyphenolic compounds: oxidized phenols and flavonoids. In extracts of the above ground organs of *Geum montanum* the rock content sum of oxidized phenols is 1.2 times higher than that of *Geum urbanum*. In extracts of underground organs of *Geum montanum* the content of sum of oxidized phenols is 1.3 times higher than that of *Geum urbanum*. In extracts of the above ground organs of *Geum montanum*, the mountain content of flavonoids is 1.2 times higher than that of *Geum urbanum*. Medicinal plant raw materials – above ground organs and underground organs *Geum urbanum* and *Geum montanum* are promising for further research of pharmacological activity and the creation on its basis of new medicinal herbal remedies. The obtained results by chromatography in a thin layer of sorbent indicate the presence of mountain flavonoids, in particular routine and quercetin, in the above ground organs (sheet) of *Geum urbanum* and *Geum montanum*.

Keywords: *Geum urbanum* L., *Geum montanum* L., tannins.

1. INTRODUCTION

An important task of modern phytotherapy is the search for promising medicinal plants in order to create new phytopreparations. Among the biologically active substances contained in plants and having pharmacological activity, an important place is occupied by polyphenolic compounds, in particular, tannins. This is largely due to their value for medicine as a source of anti-inflammatory, antibacterial, fungicidal, antioxidant agents.

The genus *Geum* L. includes about 40 species of wild and cultivated perennial herbaceous plants that are distributed around the globe. In the genus *Geum* L., more often one species imperceptibly passes into another, due to the presence of new biomorphological properties and, as a rule, increased plasticity and heterosis. Plants of the genus *Geum* L. belong to the family Rosaceae (Rosaceae Juss.). *Geum* L. is a perennial herbaceous plant with an erect stem, depending on the species height of 20 - 75 cm, with thickened, cylindrical and numerous cordate roots, which have a pleasant aroma of cloves (hence the name "clove root"). The leaves are green, large, pubescent, lyre-pinnate collected in a basal rosette. Stem trifoliolate. The flowers are regular, five-petalled, mostly yellow, pink, red, 0.5 to 4 cm in diameter, solitary or gathered in inflorescence shield or panicle. The plants bloom in May, July, even August. Re-flowering is possible until October. The fruit is a collection of nut-shaped achenes with a long curved hook-shaped nose at the top. Of the genus *Geum* L. in Bulgaria there are four species: *Geum urbanum* L., *Geum rivale* L., *Geum rhodopaeum*. and also, a related species – *Geum montanum* L. Promising for the production of phytopreparations are species of the genus *Geum* L: *Geum urbanum* L., *Geum rivale* L., as well as a related species – *Geum montanum*, which grow in Western and Eastern Bulgaria. According to the literature among the plants of the genus is most widely used in folk medicine *Geum urbanum* L. At the same time, *Geum montanum* L. is the least studied species. Given the prospects of this area, it is important to conduct comparative phytochemical studies, in particular, *Geum urbanum* L. and *Geum montanum* L.

2. MATERIALS AND METHODS

The aim of the study was a comparative phytochemical study of extracts of medicinal plant raw materials - aboveground and underground organs of *Geum urbanum* L. and *Geum montanum* L. for the content of polyphenolic compounds - the sum of oxidisable phenols (quantification) and flavonoids (identification and quantification).

The objects of study were - medicinal plant raw materials - fresh and dried aboveground (leaves) and underground (rhizomes with roots) organs of *Geum urbanum* L., collected in August-September in the Sofia and Burgas region and *Geum montanum* L., collected in the same period at an altitude of 1,600 m above sea level on Mount Vitosh, Sofia (Bulgaria), as well as decoctions (aqueous extracts) and ethanolic (70%) extracts underground (rhizomes with roots of mountain gravity and urban gravity) and aboveground organs (leaves) of *Geum urbanum* L. and *Geum montanum* L. Detection of the main groups of biologically active substances of medicinal plant raw materials by means of qualitative reactions is carried out by generally accepted methods of obtaining aqueous, aqueous-alcoholic and hydrochloric acid extracts. Quantitative determination of the raw materials of the medicinal plants polyphenolic compounds included: determination of the content of the sum of oxidisable phenols - in aqueous extracts and determination of the content of flavonoids - in ethanolic extracts. Quantitative determination of the content of the sum of oxidized phenols in the raw materials of the medicinal plants - titrimetric method.

3. RESULTS

The content of the sum of oxidisable phenols X,%, in terms of absolutely dry raw materials is calculated by the formula:

$$X = \frac{(V - V_1) \cdot K \cdot 250 \cdot 100 \cdot 100}{m \cdot 25(100 - W)}$$

where: V is the volume of potassium permanganate solution (0.02 mol / l) used for the titration of the extract, ml; V₁ is the volume of potassium permanganate solution (0.02 mol / l) used for titration in the control experiment, ml; K is the amount of organic phenols corresponding to 1 ml of potassium permanganate solution (0.02 mol / l) (in terms of tannin): K = 0.00582; m - mass of raw materials, g; W - weight loss during drying of raw materials,%; 250 - total volume of extract, ml; 25 - volume of extract taken for titration, ml

Quantitative content of flavonoids was carried out by measuring the optical density of the formed colored complex with a solution of aluminum chloride by spectrophotometric method. The content of the sum of flavonoids in terms of quercetin and absolutely dry raw material in percent (X) is calculated by the formula:

$$X = \frac{D \cdot 25 \cdot 100 \cdot 100}{764,6 \cdot m \cdot 2(100 - W)}$$

where: D is the optical density of the test solution; 764.6 - relative absorption of the quercetin complex with aluminum chloride at a wavelength of 428 nm; m - mass of raw materials, g; W is the weight loss during drying of raw materials, %

Identification of flavonoids in medicinal plant raw materials was performed by chromatography in a thin layer of sorbent:

- chromatography was performed on plates "Sorbfil PTSH-AF-A-UV";
- the analyzed substances were applied to the plate in the form of ethanolic solutions;
- alcoholic solutions of rutin and quercetin were used as reference samples;
- the applied sample volume was: 20 µl of test solutions and 10 µl of comparison solutions;
- the mobile phase was prepared by mixing the solvents immediately before use. The chromatographic chamber was pre-saturated with solvent vapor for 30 minutes. Ethyl acetate-acetic acid-water solvent system was used as the mobile phase (80:10:10); mobile phase front: 10 cm from the start line; drying: in a fume hood at room temperature to remove traces of solvents; manifestation: after treatment with 2% alcohol solution of aluminum chloride and its subsequent drying, the plate is viewed in UV light at a wavelength of 254 nm.

The results of the phytochemical study are presented in table 1.

Phytochemical study of aqueous, aqueous-alcoholic and aqueous extracts acidified with hydrochloric acid, leaves and rhizomes with roots of urban and mountain gravity for the presence of major groups of biologically active substances revealed the presence of tannins in all studied organs, anthocyanins, flavonoids and flavonoids. No alkaloids, anthracene derivatives and cardiac glycosides were detected.

The results of determining the content of oxidisable phenols in terms of absolutely dry raw materials in the aboveground and underground bodies of urban and mountain gravity are shown in table 2, which indicate that their content is different in both aboveground and underground bodies and depends on the species.

Table 1. Results of general phytochemical analysis of the above-ground and underground organs

Researched biologically active substances	Medicinal plant raw materials			
	<i>Geum urbanum</i>		<i>Geum montanum</i>	
	extract of aboveground organs	extract of underground organs	extract of aboveground organs	extract of underground organs
Alkaloids	–	–	–	–
Anthocyanins	±	±	±	±
Tannins	+	+	+	+
Coumarins	±	±	±	±
Saponins	–	–	–	–
Cardiac glycosides	–	–	–	–
Flavonoids	+	±	+	±

(+) - positive reaction; (±) - positive reaction (substances detected in small quantities); (-) - negative reaction (substances not detected)

The quantitative content of sum of oxidized phenols and flavonoids in the above ground and underground organs of *Geum urbanum* L. and *Geum montanum* L.

Indicator	<i>Geum urbanum</i>		<i>Geum montanum</i>	
	extract of aboveground organs	extract of underground organs	extract of aboveground organs	extract of underground organs
Quantitative content of oxidisable phenols, %	11,6 ±0,4	25,2 ±0,5	14,1 ±0,3	30,8 ±0,5
Flavonoids, %	2,8 ±0,2	–	3,4 ±0,2	–

4. DISCUSSION

The obtained results will allow to standardize medicinal plant raw materials with its subsequent use in the creation of phytomedicines.

5. CONCLUSIONS

A comparative phytochemical study found that the content of oxidized phenols in the extract of aboveground organs of *Geum urbanum* L. is 11.6%, while their content in the extract of *Geum montanum* L. - 14.1%, which is 1.2 times more. The content of oxidized phenols in the extract of underground organs of *Geum urbanum* L. is 25.2%, and their content in the extract of *Geum montanum* L. is 30.8%, which is almost 1.3 more. Studies of the quantitative content of flavonoids in ethanolic extracts of aboveground organs showed that their content in *Geum urbanum* L. is 2.8%, and in *Geum montanum* L. extract - 3.4%, which is almost 1.2 more.

Chromatography in a thin layer of sorbent showed that in the lower part of the chromatogram in UV light at a length of 254 nm of the studied samples - alcohol solutions of aboveground organs of *Geum urbanum* L. and *Geum montanum* L. revealed a yellow fluorescent zone at the level of rutin - $R_f = 0.21 - 0.24$. At the top of the chromatogram of the studied samples - ethanolic solutions of aboveground organs of *Geum urbanum* L. and *Geum montanum* L., a yellow-green fluorescent zone was detected at the level of the quercetin zone - $R_f = 0.82 - 0.85$. The data obtained by thin layer chromatography indicate the presence of flavonoids, in particular rutin and quercetin, in

the aboveground part of *Geum urbanum* L. and *Geum montanum* L. Thus, the most valuable tannin-containing raw materials for harvesting are rhizomes with roots of both types of *Geum*.

Phytochemical studies of aqueous, aqueous-alcoholic and aqueous extracts acidified with hydrochloric acid aboveground (leaves) and underground (rhizomes with roots) organs of *Geum urbanum* L. and *Geum montanum* L. for the presence of the main groups of biologically active substances revealed a significant amount of tannins (hydrolyzed and condensed). Phytochemical study of aqueous extracts of aboveground and underground organs and ethanolic extracts of underground organs of *Geum urbanum* L. and *Geum montanum* L. established the content of polyphenolic compounds: quantitative content of the sum of oxidisable phenols (tannins), identification and quantitative content of flavonoids. The maximum amount of oxidized phenols (tannins) was found in the underground organs of both *Geum montanum* L. and *Geum urbanum* L., respectively, 31.6% and 24.8%, less - in the aboveground part, respectively, 14.9% and 12.5%. In the underground and aboveground organs of *Geum montanum* L. by 19 - 27% higher content of the sum of oxidisable phenols (tannins) in relation to *Geum urbanum* L., obviously, due to the strict conditions of growth of *Geum montanum* L. The quantitative content of flavonoids in the aboveground part of *Geum urbanum* L. - 2.6%, while in *Geum montanum* L. - 3.1% of flavonoids was determined by spectrophotometric method. The results of identification by thin layer chromatography indicate the presence of flavonoids, in particular rutin and quercetin, in the aboveground organs (leaves) of *Geum urbanum* L. and *Geum montanum* L. Medicinal plant raw materials - aboveground and underground organs of *Geum urbanum* L. and *Geum montanum* L. are promising for further study of pharmacological activity and the creation of new herbal medicines on its basis.

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