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Polyphenolic Compouds of Plant of Lepidium Ruderale Linn. and Their Biololical Activity

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Abstract

This article presents the results of the study of the plant Lepidium ruderale L. A method was developed for the complete extraction of polyphenolic substances from the plant, using the method of high-performance liquid chromatography flavonoids were identified and determined in the plant: kaempferol, quercetin; using mass spectrometry method, isovitexin belonging to the class C of flavonoids was identified; the chelating activity of the plant Lepidium ruderale L. was studied and it was revealed that the plant has a high biological activity; the content of organic acids was determined by capillary electrophoresis.

Keywords: Plants Lepidium Ruderale L., Flavanoids, Mass Spectrometry, High-Performance Liquid Chromatography Method, Astragalin, Luteolin, Kaempferol, Quercetin, Biological Activity, Capillary Electrophoresis Method, Organic Acids.

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Introduction

Lepidium ruderale L., which is known as Ban Helem in the Himalayas, is used as food and medicinal plant. It is used for treatment of cough, asthma, hemorrhoid, skin diseases, and rheumatism by the tribal inhabitants of the central Himalaya (Baoliang et al., 2003).

Recently, plant materials or their extracts as substitute and supportive medicine have been used increasingly, and several antimicrobial agents have been discovered and identified (Qusti et al., 2018). In the current study, *Lepidium ruderale L.* was tested for various biological activities like antifungal, antibacterial and antiviral agent. Benzyl glucosinolates, the principled elements of Lepidium ruderale L., showed anti-amebic activity. Quercetin and its derivatives were discovered in the plant Lepidium ruderale L. Quercetin – a plant flavanol, belonging to the group of catechins, the most abundant element in fruits and vegetables, has a higher antioxidant potential than vitamins C and E and a great biological activity (Jyota et al., 2003; Abu Zaker Khaled, 2001). In Kazakhstan, 20 species of peppergrasses are found (Baitenov, 2001).

Medicinal plants have been anticipated to be one of the most valuable resources in therapeutic practices for human diseases(Sargia et al., 2018). The use of herbal medicine represents a long history of human body (Sayed Ahmad et al., 2018). Historically, herbal medicines and their derivatives have demonstrated an important alternative for the treatment of illnesses (Nadri et al., 2018). Therefore, recently, there have been a lot of studies conducted on examining the contents of various plants. El Abidine Ababsa et al., (2018) in a study examined the chemical characterization and biological study of the species Senecio Cineraria.

The purpose of this study: Studying the polyphenolic compounds of the plant Lepidium ruderale L. and determining their biological activity.

Research object: the plant Lepidium ruderale L. (weedy Lepidium), collected in the Pavlodar region, in the nature

reservation territory of the Bayan-aul in the Republic of Kazakhstan.

In the course of this study, an optimal method was developed for extracting polyphenolic compounds from the overgroud part of Lepidium ruderale L., wherein 3 different methods for extracting substances were utilized: hot water, ethyl alcohol and methanol.

Table 1: The total content of polyphenolic compounds in different extract methods

Plant	Metanol	Etanol	Water
	The total amount of phenolic compounds, mg GAE / 100 g	The total amount of phenolic compounds, mg GAE / 100 g	The total amount of phenolic compounds, mg GAE / 100 g
Lepidium ruderale L.	71,4	51,9	56,3

As can be seen from table 1, the most complete extraction is achieved by using extraction with methanol.

Figure 1 shows the absorption spectra of the obtained extracts from Lepidium ruderale L.

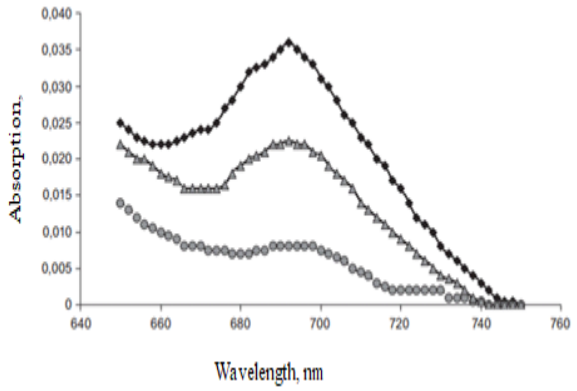


Fig. 1- Absorption spectra of different extracts: 1- spectrum of methanol extract, 2- spectrum of ethanol extract, 3 - spectrum of water extract of a plant

Polyphenolic compounds were separated using high-efficiency liquid chromatography. For the separation efficiency, the corresponding column Zorbax SB C18 (3.5 μm) 3x150 mm was chosen, the mobile phase: methanol - acetic acid solution 0.01% (25: 75); speed of the mobile phase: 1.2 cm3 / min; column temperature: 250 0C; detection under UV, λ = 254 nm, sample volume is 20 mm3. The chromatogram of flavonoids illustrated in Figure 2, the retention time indicated in Table 3.

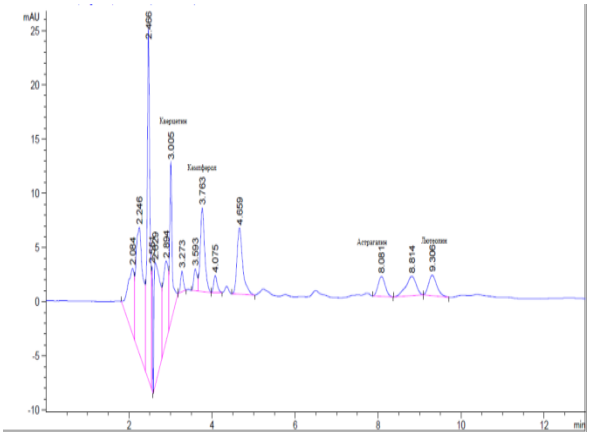
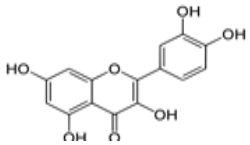
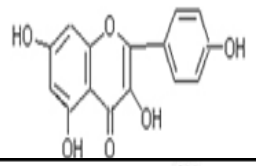
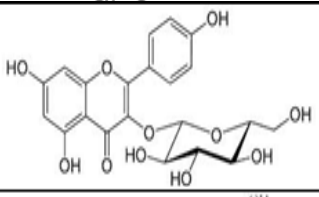
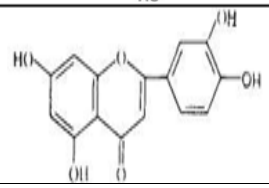


Fig. 2- Chromatogram of flavonoids of the plant Lepidium ruderale L

Table 2: Quantitative content of flavonoids

Plant	Astragalin, g/kg	Luteolin, g/kg	Quercetin, g/kg	Kaempferol, g/kg
Lepidium ruderale	1,03	1,32	3,04	0,63
Total content	6, 02			

Table 3: Identification of flavonoids Lepidium ruderale L.

Substance name	Retention time	Formula
Quercetin	3,005	
Kaempferol	3,763	
Astragalin	8,081	
Luteolin	9,306	

To identify substances not identified by HPLC, mass spectrometry was used. For the determination of flavonoids by mass spectrometry, the method (Sheel Sharma, Nidhi Agarwal, 2011) was used. The results are shown in Figure 3.

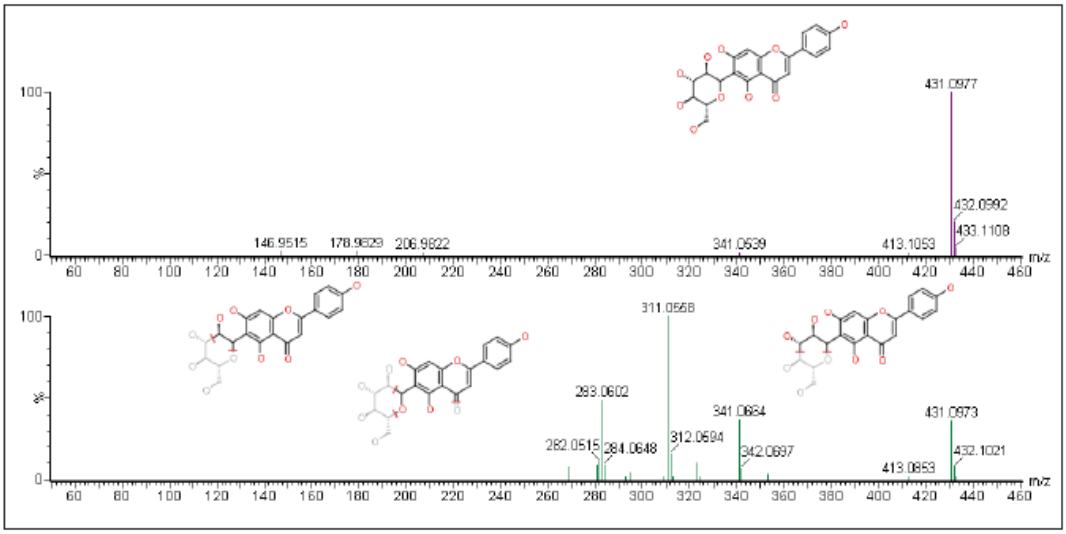
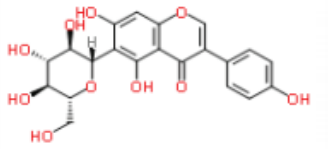


Fig. 3 - Isovitexin mass spectra

Conditions for mass spectrometry analysis: ionization - negative, cell temperature - 1200C, source temperature - 5000C, column temperature - 400C, sample injected volume - 5 ml, analysis time -10 min (table 4).

Table 4: Identification of flavanoids Lepidium ruderale L.

Substance name	retention time	formula
Isovitexin	4,07	

A study of the chelating properties of weedy Lepidium was conducted and it was found that different extracts from the plant exhibit different chelating ability, the data are shown in Table 5.

Table 5: Chelating activity of various extracts of the overground part of weedy Lepidium

Plant	Extract	Concentration, µg/mg	Chelating activity (%)
Lepedium ruderale L.	Methanol	50	33,2
		100	45,6
		250	62,3
		500	77,7
	Ethanol	50	15,2
		100	25,9
		250	33,9
		500	51,5
	Water	50	23,1
		100	33,4
		250	46,7
		500	61,5

It was found that the methanolic extract of the plant has the greatest chelating activity - 77.7%, and aqueous extract has a little less - 61.5%.

Capillary electrophoresis was used to determine the organic acids and 4 organic acids were detected in the extract, which were separated on a 55 cm long capillary with an inner diameter of 0.25 µm, the results of the study are shown in Table 6.

Table 6- Organic acid content

Substance	Contents, g/kg
Tartaric acid	0,4
Malic acid	0,7
Citric acid	1,2
Succinic acid	0,9
Total content of organic acids- 3,2	

Findings: in the plant Lepidium ruderale L. during the study:

- a method was developed for the complete extraction of polyphenolic substances from a plant, and in this way it was determined that, extraction with methanol is 1.5–2 times more efficient than extraction with ethyl alcohol;
- using the high-performance liquid chromatography method in the plant, 6.02 g / kg of flavonoids were identified and determined including: astragaline - 1.03 g / kg; luteolin-1.32 g / kg; kaempferol-0.63 g / kg and quercetin-3.04 g / kg;
- by using mass spectrometry, Isovitexin belonging to the class C of flavonoids was identified;
- the chelating activity of the plant Lepidium ruderale L. was studied, and the plant was found to have a high biological activity of 77.7% in methanolic extract and 61.5% in an aqueous extract;
- by using capillary electrophoresis the organic acid content of 3.2 g / kg was determine: tartaric acid - 0.4 g / kg; malic

acid — 0.7 g / kg; citric acid - 1.2g / kg; succinic acid - 0.9 g / kg.

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