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PHYTOCHEMICAL CHARACTERIZATION AND COMPARATIVE ANALYSIS OF BIOLOGICALLY ACTIVE COMPOUNDS IN *CIRSIUM VULGARE* AND *CIRSIUM ARVENSE*

Annotation: *Species of the genus Cirsium are considered promising sources of biologically active compounds and attract considerable interest in phytochemical and pharmacognostic research. Among them, Cirsium vulgare and Cirsium arvense are widely distributed in Kazakhstan and have traditionally been used in folk medicine. Previous studies have reported the presence of phenolic compounds, fatty acids, vitamins, and other secondary metabolites in representatives of this genus; however, comparative phytochemical data for these species remain limited.*

The aim of the present study was to perform a comparative analysis of physicochemical characteristics and biologically active compounds in aqueous-ethanolic extracts of Cirsium vulgare and Cirsium arvense. Fatty acid composition was determined by gas chromatography with flame ionization detection, total phenolic compounds were evaluated spectrophotometrically using the Folin-Ciocalteu method, and vitamin C content was analyzed by high-performance liquid chromatography. In addition, moisture content and total ash values of the plant material were determined according to standard analytical procedures.

The obtained results demonstrated noticeable differences in the phytochemical composition of the studied species. In total, twenty-one fatty acids were identified in the extracts. Cirsium arvense was characterized by a higher content of linolenic acid and vitamin C, whereas Cirsium vulgare contained increased amounts of total phenolic compounds together with linoleic, eicosenoic, and palmitic acids. Similar differences in the accumulation of biologically active compounds may be associated with species-specific metabolic features and environmental conditions.

The present study expands current information on the phytochemical composition of local representatives of the genus Cirsium. The obtained results may be considered as preliminary comparative data for further pharmacognostic, phytochemical, and applied investigations related to medicinal plant raw materials and biologically active compounds.

Key words: *cirsium vulgare, cirsium arvense, fatty acids, phenolic compounds, vitamin c, phytochemical analysis.*

Introduction

Kazakhstan is distinguished by its rich flora and the wide distribution of wild medicinal plants, which are considered a source of biologically active compounds. Despite the growing interest in medicinal plant raw materials, the phytochemical composition of many wild plants has not yet been fully studied. The study of local plant resources and the identification of biologically active substances is one of the important directions of pharmaceutical and pharmacognostic research.

Currently, plant-derived compounds are considered promising components of phytopharmaceutical and nutraceutical products. Natural products differ in structural diversity compared to synthetic substances and the presence of biologically important metabolites. In this regard, the study of wild medicinal plants growing in Kazakhstan is of scientific and practical interest [1].

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One of the important stages of phytochemical research is the isolation and analysis of biologically active compounds from plant raw materials. The complexity of plant matrices, the variability of secondary metabolites, and the influence of environmental factors require the use of modern analytical methods. Gas chromatography, high-performance liquid chromatography, and spectrophotometric methods are widely used in the study of fatty acids, phenolic compounds, vitamins, and other metabolites [2].

Plants belonging to the *Cirsium* (*Asteraceae*) family, widespread in Kazakhstan, are of particular interest. Representatives of this family have long been used in folk medicine. However, information on their phytochemical composition is still limited. The literature indicates that some *Cirsium* species contain flavonoids, phenolic compounds, fatty acids, vitamins, and other secondary metabolites. However, comparative studies conducted under the same analytical conditions are lacking [3].

The scientific novelty of the study is associated with the comparative phytochemical analysis of *Cirsium vulgare and Cirsium arvense* plants collected in the Almaty region. For the first time, the physicochemical parameters, fatty acid composition, vitamin C, and total phenolic compounds of these plants were systematized and comparatively studied under a single analytical condition. The aim of the study was to conduct a comparative analysis of the physicochemical parameters and biologically active compounds of *Cirsium vulgare and Cirsium arvense*. The species-specific characteristics of their phytochemical composition were assessed by determining the content of fatty acids, vitamin C, and total phenolic compounds.

Materials and methods

The Study Objects and Raw Material Preparation

The objects of the study were the aerial parts of *Cirsium vulgare* and *Cirsium arvense* collected during the flowering period in the Almaty region of the Republic of Kazakhstan in 2025. Taxonomic identification of the plant material was carried out according to morphological characteristics using standard floristic references.

The collected raw material was cleaned from mechanical impurities and dried in the shade under natural ventilation at room temperature until reaching an air-dry state. The dried material was ground to obtain a homogeneous powdered mass. Prepared samples were stored in tightly closed containers under dry and dark conditions until further analysis.

Extraction was performed using 70% ethanol as an extractant at a raw material-to-solvent ratio of 1:10.Ultrasonic extraction was carried out for 1 h at room temperature. Extraction was repeated three times to ensure maximum recovery of biologically active compounds. After extraction, the obtained solutions were filtered and concentrated, while the remaining plant residue (meal) was used for further analysis.

Determination of Physicochemical Parameters

Physicochemical characteristics of the plant raw material were evaluated using standard pharmacopoeial and analytical procedures. The investigated parameters included moisture content, total ash, organic acids, saponins, flavonoids, coumarins, and tannins. Moisture content was determined by drying samples to constant weight. Total ash content was evaluated after thermal treatment of accurately weighed samples in a muffle furnace followed by gravimetric analysis. Organic acids were determined by titration using an appropriate indicator system.

Quantitative determination of saponins, flavonoids, coumarins, and tannins was performed spectrophotometrically using corresponding standard compounds and calibration curves. The obtained results were expressed as percentages of absolutely dry plant material [4].

Determination of Fatty Acid Composition

Qualitative and quantitative analysis of fatty acids in hydroalcoholic extracts of Cirsium vulgare and Cirsium arvense was performed by gas chromatography with flame ionization detection (GC-FID) using a GX-1000 “Chromas” gas chromatograph (Russia) according to the methodology of MVI MN 1364-2000. Prior to chromatographic analysis, lipid fractions were isolated from the plant material and subjected to transesterification to obtain fatty acid methyl esters. Chromatographic separation was carried out under programmed temperature conditions using a capillary column and helium as the carrier gas. Identification of individual fatty acids was based on comparison of retention times with those of corresponding standard compounds. Quantitative evaluation of the detected

components was performed by calculating the relative percentage of each fatty acid in relation to the total identified fatty acid content [5].

Determination of Vitamin C

Quantitative determination of vitamin C (ascorbic acid) in the studied plant extracts was carried out using high-performance liquid chromatography (HPLC) on a K2025 chromatograph (China) equipped with a reversed-phase C18 column. Chromatographic analysis was performed according to the standard procedure R.4.1.1672-03 for the determination of water-soluble vitamins. The mobile phase consisted of water and acetonitrile under isocratic elution conditions at a flow rate of 1.0 mL/min. Detection of analytes was performed at wavelengths of 210 and 254 nm. Identification of ascorbic acid was achieved by comparing the retention time of the sample peak with that of the reference standard. Quantitative determination was based on a calibration curve constructed using standard solutions of ascorbic acid, and the results were expressed in mg per 100 g of absolutely dry plant material [6].

Determination of Total Phenolic Content

Total phenolic content in hydroalcoholic extracts was determined spectrophotometrically using the Folin–Ciocalteu method. For analysis, the reaction mixture consisted of the plant extract, Folin–Ciocalteu reagent, and sodium carbonate solution. After incubation under standard laboratory conditions, absorbance was measured at 765 nm using a UV–Vis spectrophotometer. Quantitative determination of phenolic compounds was carried out using a calibration curve prepared with gallic acid standard solutions. The obtained results were expressed as milligrams of gallic acid equivalents per gram of dry plant material (mg GAE/g) [7].

Statistical Analysis

Statistical analysis was performed using Student's t-test. Differences between the compared groups were considered statistically significant at $p < 0.05$. All experiments were carried out in triplicate ($n = 3$), and the results are presented as mean value ± standard deviation [9].

Results and Discussion

Studies of various representatives of the genus *Cirsium* have shown that these plants are rich sources of biologically active compounds, including flavonoids, phenolic compounds, tannins, fatty acids, vitamins, and other secondary metabolites. Previous investigations of species such as *Cirsium japonicum, Cirsium setidens,* and *Cirsium arvense* demonstrated the presence of flavonoids, phenolic acids, tannins, and antioxidant-active compounds associated with the pharmacological potential of these plants.

The objects of the study were the aerial parts of plants of the genus *Cirsium* – *Cirsium vulgare* and *Cirsium arvense*, collected during the flowering phase in the Almaty region of Kazakhstan in 2025. After harvesting, the plant material was dried in shaded conditions at ambient temperature and subsequently milled to a fine powder. Interest in these species is mainly related to their chemical composition, which includes phenols, flavonoids, coumarins, saponins, and polyunsaturated fatty acids responsible for several biological effects. Similar phytochemical trends have previously been reported for other representatives of the *Asteraceae* family (Table 1, 2).

Table 1 – Physicochemical Parameters of *Cirsium* Species

Parameter	*Cirsium vulgare* (%)	*Cirsium arvense* (%)
Moisture content	5.97 ± 0.11	7.65 ± 0.14
Total ash	9.00 ± 0.09	10.00 ± 0.12

Table 2 – Content of Biologically Active Compounds in Medicinal Plant Raw Material of *Cirsium vulgare* and *Cirsium arvense*

Parameter	*Cirsium vulgare* (%)	*Cirsium arvense* (%)
Free organic acids	0.39 ± 0.02	0.36 ± 0.01
Saponins	1.51 ± 0.05	2.51 ± 0.08
Coumarins	0.25 ± 0.01	0.57 ± 0.02
Flavonoids	5.27 ± 0.14	4.06 ± 0.10
Tannins	2.92 ± 0.09	4.45 ± 0.12

Data are presented as mean ± SD (n = 3).

Statistical analysis was performed using Student's t-test, and differences between species were considered statistically significant at $p < 0.05$.

Data are presented as mean ± SD (n = 3).

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The increased moisture and ash values recorded for *Cirsium arvense* suggest differences between the species in water retention and mineral accumulation, which play an important role in plant metabolism. At the same time, the content of organic acids in both species was found to be similar, indicating a comparable contribution of these compounds to the antioxidant properties of the obtained extracts.

A higher content of saponins, coumarins, and tannins was observed in *Cirsium arvense*. These groups of compounds are commonly discussed in the literature in connection with potential immunomodulatory, anticoagulant, and antibacterial effects. In contrast, *Cirsium vulgare* demonstrated a higher level of flavonoids, which may contribute to antioxidant and anti-inflammatory properties.

Overall, the revealed differences reflect species-specific features in the accumulation of biologically active compounds. These findings are important for the rational selection of plant raw materials intended for pharmaceutical use [8-9].

Fatty Acid Profile

Quantitative determination of fatty acids was carried out by gas chromatography using a flame ionization detector. The analysis was performed on a GX-1000 "Chromas" chromatograph in accordance with the MVI MN 1364-2000 protocol. In total, twenty-one individual fatty acids were identified in the studied samples, and the obtained data are summarized in Table 3.

Table 3 – Fatty acid composition (% of total) in *Cirsium* extracts

Compound	Name	*Cirsium vulgare (%)*	*Cirsium arvense (%)*
C12:0	Lauric acid	ND	0.143 ± 0.01
C13:0	Tridecanoic acid	ND	0.205 ± 0.01
C14:0	Myristic acid	1.108 ± 0.03	0.330 ± 0.02
C15:0	Pentadecanoic acid	ND	0.126 ± 0.01
C15:1	Pentadecenoic acid	ND	0.134 ± 0.01
C16:0	Palmitic acid	18.712 ± 0.42	10.016 ± 0.31
C16:1	Palmitoleic acid	ND	0.606 ± 0.03
C18:0	Stearic acid	2.682 ± 0.08	2.112 ± 0.07
C18:1	Oleic acid	4.605 ± 0.12	2.564 ± 0.09
C18:2n6c	Linoleic acid	29.710 ± 0.55	13.849 ± 0.47
C18:3n6	γ-Linolenic acid	ND	0.280 ± 0.02
C18:3n3	α-Linolenic acid	5.913 ± 0.19	32.338 ± 0.74
C20:0	Arachidic acid	6.963 ± 0.21	0.240 ± 0.01
C20:1	Eicosenoic acid	21.409 ± 0.51	3.938 ± 0.14
C20:2	Eicosadienoic acid	4.436 ± 0.15	0.805 ± 0.03
C20:3n3	Eicosatrienoic acid	4.461 ± 0.13	ND
C21:0	Heneicosanoic acid	ND	1.858 ± 0.06
C22:0	Behenic acid	ND	25.775 ± 0.63
C22:6n3	Docosahexaenoic acid	ND	0.573 ± 0.03
C23:0	Tricosanoic acid	ND	0.699 ± 0.03
C24:0	Lignoceric acid	ND	3.048 ± 0.10

ND – not detected.

Data are presented as mean ± SD (n = 3).

Statistical analysis was performed using Student's t-test, and differences between species were considered statistically significant at $p < 0.05$.

Cirsium arvense was found to contain relatively high levels of docosahexaenoic acid (C22:6n3). Since docosahexaenoic acid is commonly found in marine and microbial organisms, its

detection in terrestrial plant samples may be due to the specific analytical conditions, overlapping chromatographic peaks, or environmental factors. The results should be considered preliminary and require further confirmation using reference standards or by additional chromatographic studies. The relatively high content of behenic acid observed in the studied samples may be associated with species-specific peculiarities of lipid metabolism, ecological growing conditions, climatic factors, or the developmental stage of the plants at the time of collection.

There are differences in the fatty acid composition between *Cirsium vulgare* and *Cirsium arvense*. Linoleic, eicosenoic, and palmitic acids predominate in *Cirsium vulgare. Cirsium arvense* had higher levels of linolenic acid and long-chain saturated fatty acids. Similar fatty acids have been described in other members of the *Cirsium* family (*Asteraceae*), and differences in their quantitative composition may be related to environmental conditions, plant growth period, and extraction parameters. Polyunsaturated fatty acids identified in the studied extracts are important structural components of plant cell membranes. According to literature, linoleic and linolenic acids are associated with antioxidant and protective properties of lipid fractions of plant origin. The high content of linolenic acid in *Cirsium arvense* may indicate specific features of lipid metabolism of this species. The results should be considered primarily as a phytochemical characterization of the studied species. The identified fatty acid profile indicates the potential biological significance of these compounds and demonstrates the prospects for further pharmacological and biochemical studies on *Cirsium* extracts [10].

Vitamin C content

The content of ascorbic acid was quantified by high-performance liquid chromatography using a C18 column. Detection was carried out at wavelengths of 210 and 254 nm with a K2025 chromatographic system (China), in accordance with method R.4.1.1672-03 (Table 4).

Table 4 – Vitamin C content (mg/100 g of dry plant material)

Thistle species	Vitamin C (mg/100 g DW)
Cirsium vulgare	0.915 ± 0.03
Cirsium arvense	3.099 ± 0.08

Data are presented as mean ± SD (n = 3).

Statistical analysis was performed using Student's t-test, and differences between species were considered statistically significant at $p < 0.05$.

The obtained results demonstrated differences in the content of ascorbic acid between the studied *Cirsium* species. A higher concentration of vitamin C was detected in *Cirsium arvense* compared to *Cirsium vulgare.* Similar variations in ascorbic acid accumulation have previously been described for medicinal plants depending on species characteristics, environmental conditions, and vegetation stage.

Ascorbic acid is known as one of the important water-soluble antioxidants present in plant tissues. In addition to its protective role against oxidative processes, vitamin C may contribute to stabilization of other biologically active compounds, including phenolic substances and flavonoids. The increased content of ascorbic acid identified in *Cirsium arvense* may reflect specific metabolic features of this species.

At the same time, the obtained data should be considered primarily as comparative phytochemical characteristics of the studied plant materials. The relatively high vitamin C content observed in *Cirsium arvense* indicates the prospects for further investigation of this species as a potential source of biologically active compounds for phytopharmaceutical and nutraceutical applications [11].

The obtained HPLC chromatogram contains a clearly defined ascorbic acid peak, enabling reliable determination of vitamin C content in the samples (Figure 1).

Phenolic Compounds Content

The determination of total phenolic compounds in 70% ethanolic extracts was carried out using the Folin-Ciocalteu spectrophotometric method (Table 5).

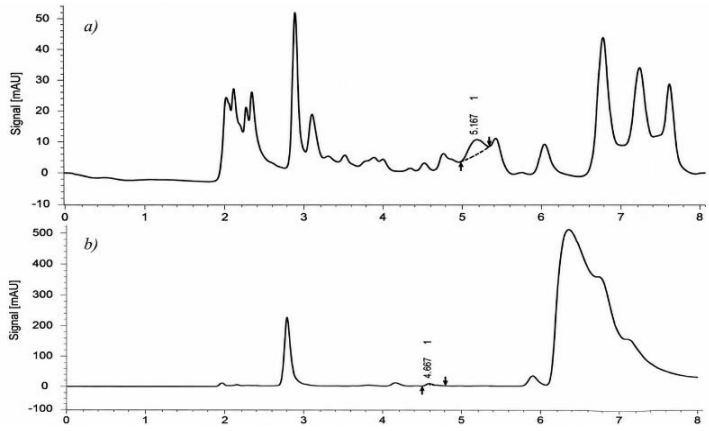


Figure 1 – HPLC chromatograms of vitamin C in extracts of:
a) Cirsium arvense; b) Cirsium vulgare

Table 5 – Phenolic Compounds (mg GAE/g)

Thistle species	Phenolic compounds (mg GAE/g DW)
Cirsium vulgare	30.11 ± 0.62
Cirsium arvense	25.32 ± 0.54

Data are presented as mean ± SD (n = 3).

Statistical analysis was performed using Student's t-test, and differences between species were considered statistically significant at $p < 0.05$.

Phenolic compounds are among the major groups of biologically active substances found in medicinal plants. The obtained results demonstrated that the total phenolic content in *Cirsium vulgare* was higher than in *Cirsium arvense*. Similar differences in phenolic accumulation have previously been described for representatives of the *Asteraceae* family and may be associated with species characteristics, environmental conditions, and physiological stage of plant development.

Phenolic compounds identified in the studied extracts are known as important components of plant protective systems. According to literature data, these substances may contribute to antioxidant and membrane-protective properties of plant extracts. The increased phenolic content detected in *Cirsium vulgare* may indicate certain features of secondary metabolism in this species.

At the same time, the obtained data should be considered primarily as comparative phytochemical characteristics of the studied plants. The results demonstrate the potential of further investigation of *Cirsium* species as potential sources of phenolic compounds and other biologically active substances for pharmacognostic and phytochemical studies[12].

In the present study, comparative phytochemical differences between *Cirsium vulgare* and *Cirsium arvense* were demonstrated. The obtained results confirmed species-specific features in the accumulation of fatty acids, phenolic compounds, flavonoids, tannins, coumarins, saponins, and vitamin C, which is generally consistent with literature data for other representatives of the genus Cirsium and the *Asteraceae* family.

Conclusions

A comparative phytochemical study of *Cirsium vulgare* and *Cirsium arvense* plants collected in the Almaty region of Kazakhstan showed differences in physicochemical parameters and the content of biologically active compounds. The results confirmed the presence of species-specific features in the accumulation of fatty acids, vitamin C and phenolic compounds.

While *Cirsium arvense* contained a higher content of ascorbic acid and linolenic acid, *Cirsium vulgare* was characterized by a higher content of total phenolic compounds, as well as linoleic, eicosene and palmitic acids. Such differences in phytochemical composition may be associated with the characteristics of plant metabolism, environmental conditions and the physiological development stage of the plant.

The identified compounds are known to be important components of plant biochemical systems and may contribute to the biological significance of the studied extracts. Since the study is limited only to comparative phytochemical characterization, conclusions regarding potential

biological activity should be considered preliminary and require additional pharmacological and biochemical studies.

The scientific novelty of the study is characterized by a comparative analysis of *Cirsium vulgare* and *Cirsium arvense* plants growing in Kazakhstan under uniform analytical conditions. For the first time, comparative data on the physicochemical parameters, fatty acid composition, vitamin C content and total phenolic compounds of these local plant raw materials were systematized and analyzed. The results complement the current knowledge about representatives of the *Cirsium* genus and indicate the prospects for future pharmacognostic, phytochemical and applied research aimed at finding plant sources of biologically active compounds.

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***CIRSIUM VULGARE ЖӘНЕ CIRSIUM ARVENSE ӨСІМДІКТЕРІНДЕГІ БИОЛОГИЯЛЫҚ БЕЛСЕНДІ ҚОСЫЛЫСТАРДЫҢ ФИТОХИМИЯЛЫҚ СИПАТТАМАСЫ ЖӘНЕ САЛЫСТЫРМАЛЫ ТАЛДАУЫ**

Cirsium туысына жататын өсімдіктер биологиялық белсенді қосылыстардың перспективті көздері ретінде қарастырылып, фитохимиялық және фармакогностикалық зерттеулерде үлкен қызығушылық тудырады. Олардың ішінде Cirsium vulgare және Cirsium arvense Қазақстан аумағында кең таралған және халық медицинасында дәстүрлі түрде қолданылып келеді. Бұрын жүргізілген зерттеулерде бұл туыс өкілдерінің құрамында фенолды қосылыстар, май қышқылдары, дәрумендер және басқа да екіншілік метаболиттер бар екендігі көрсетілген, алайда аталған түрлер бойынша салыстырмалы фитохимиялық мәліметтер әлі де шектеулі.

Зерттеудің мақсаты Cirsium vulgare және Cirsium arvense өсімдіктерінің су-этанолды экстракттарындағы физика-химиялық көрсеткіштер мен биологиялық белсенді қосылыстарға салыстырмалы талдау жүргізу болды. Май қышқылдарының құрамы жалынды-ионизациялық детекторы бар газды хроматография әдісімен анықталды, фенолды қосылыстардың жалпы мөлшері Фолин-Чокальтеу спектрофотометриялық әдісі арқылы бағаланды, ал С дәруменінің мөлшері жоғары тиімді сұйықтық хроматографиясы әдісімен зерттелді. Стандартты аналитикалық әдістерге сәйкес өсімдік шикізатының ылғалдылығы мен жалпы күл мөлшері анықталды.

Зерттелген түрлердің фитохимиялық құрамында айқын айырмашылықтар бар екенін көрсетті. Экстракттар құрамынан жалпы жиырма бір май қышқылы анықталды. Cirsium arvense құрамында линолен қышқылы мен С дәруменінің мөлшері жоғары болса, Cirsium vulgare құрамында жалпы фенолды қосылыстардың, сондай-ақ линол, эйкозен және пальмитин қышқылдарының мөлшері басым болды. Биологиялық белсенді қосылыстардың жиналуындағы анықталған айырмашылықтар метаболизмнің түрлік ерекшеліктерімен және қоршаған орта жағдайларымен байланысты болуы мүмкін.Зерттеу Cirsium туысының жергілікті өкілдерінің фитохимиялық құрамы туралы мәліметтерді толықтырады. Нәтижелер дәрілік өсімдік шикізаты мен биологиялық белсенді қосылыстарға байланысты болашақ фармакогностикалық, фитохимиялық және қолданбалы зерттеулер үшін алдын ала салыстырмалы деректер ретінде қарастырылуы мүмкін.

Түйін сөздер: *cirsium vulgare, cirsium arvense, май қышқылдары, фенолды қосылыстар, дәрумен с, фитохимиялық анализ.*

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ФИТОХИМИЧЕСКОЕ ИССЛЕДОВАНИЕ И СРАВНИТЕЛЬНЫЙ АНАЛИЗ БИОЛОГИЧЕСКИ АКТИВНЫХ СОЕДИНЕНИЙ В *CIRSIUM VULGARE* И *CIRSIUM ARVENSE*

Представители рода Cirsium рассматриваются как перспективные источники биологически активных соединений и вызывают значительный интерес в фитохимических и фармакогностических исследованиях. Среди них Cirsium vulgare и Cirsium arvense широко распространены на территории Казахстана и традиционно используются в народной медицине. Ранее проведенные исследования показали наличие у представителей данного рода фенольных соединений, жирных кислот, витаминов и других вторичных метаболитов, однако сравнительные фитохимические данные для этих видов остаются ограниченными.

Целью настоящего исследования являлся сравнительный анализ физико-химических характеристик и биологически активных соединений в водно-этанольных экстрактах Cirsium vulgare и Cirsium arvense. Жирнокислотный состав определяли методом газовой хроматографии с пламенно-ионизационным детектированием, общее содержание фенольных соединений оценивали спектрофотометрическим методом Фолина-Чокальтеу, а содержание витамина С анализировали методом высокоэффективной жидкостной хроматографии. Определены показатели влажности и общей золы растительного сырья согласно стандартным аналитическим методикам.

Результаты показали заметные различия в фитохимическом составе исследованных видов. Всего в экстрактах была идентифицирована двадцать одна жирная кислота. Для Cirsium arvense было характерно более высокое содержание линоленовой кислоты и витамина С, тогда как Cirsium vulgare содержал повышенные количества общих фенольных соединений, а также линолевой, эйкозеновой и пальмитиновой кислот. Выявленные различия в накоплении биологически активных соединений могут быть связаны с видоспецифическими особенностями метаболизма и условиями окружающей среды.

Проведенное исследование расширяет существующие сведения о фитохимическом составе местных представителей рода Cirsium. Полученные результаты могут рассматриваться как предварительные сравнительные данные для дальнейших фармакогностических, фитохимических и прикладных исследований, связанных с лекарственным растительным сырьем и биологически активными соединениями.

Ключевые слова: *cirsium vulgare, cirsium arvense, жирные кислоты, фенольные соединения, витамин с, фитохимический анализ .*

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