УДК 582.794.1:[543.6+615.322]
DOI 10.31654/2786-8478-2026-BN-2-48-59

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PRIMARY PHYTOCHEMICAL ANALYSIS OF *PRANGOS ACAULIS* (LINDL.) BORNM. AND COMPARATIVE ANALYSIS WITH *PRANGOS FERULACEA* (L.) LINDL.

Prangos is a perennial herb belonging to the Apiaceae family. This genus is primarily distributed in dry and semi-desert climates. Numerous species of the Prangos genus have been studied, and extensive phytochemical analyses have been conducted, revealing that they are rich in biologically active compounds. However, sufficient information regarding Prangos acaulis is lacking. Major classes of phytochemicals with disease-preventive properties include dietary fiber, antioxidants, anticancer agents, detoxifying substances, immune enhancers, and neuropharmacological compounds. The potential richness of the phytochemical composition of this species, considering its pharmacological properties, suggests that it may become a future raw material for medicine, and for this reason, the study of the plant is important. The aim of the research is to perform a phytochemical analysis of P.acaulis and P.ferulacea species using paper chromatography, to conduct preliminary screening for P. acaulis, and to evaluate the obtained results in comparison with P.ferulacea. The percolation method was employed to obtain plant extracts. Both dried and powdered plant samples were accurately weighed to 20 g each. The measured plant material was soaked in 20 ml of 70% ethanol and allowed to stand for 24 hours. After another 24-hour standing period, the liquid was completely filtered and returned to the separating funnel. From the 20 g sample, 80% was collected as the primary extract at a rate of approximately 40 drops per hour and set aside. To conduct the research, the collected plants were dried under identical conditions, ground into powder, extracts were obtained, and the analysis was performed using paper chromatography. Based on the results of the analysis, specific biologically active compounds were identified on the chromatogram, and the Rf values were calculated and compared. The evaluation of the obtained results suggests that the chromatographic profiles of the extracts, in combination with the specific reagents employed, provide insight into the possible functional groups present in the separated compounds. In the present study, a preliminary phytochemical analysis was conducted on 70% ethanol extracts obtained from the leaves and roots of Prangos acaulis *using the percolation method. This similarity suggests that* P. acaulis *may also possess a diverse array of bioactive compounds. Nevertheless, it should be emphasized that further comprehensive and detailed investigations are necessary to accurately identify the specific classes of bioactive substances present and to determine the exact molecular composition of this species.*
Keywords: Prangos acaulis, *Prangos ferulacea, paper chromatography, phytochemical analysis.*

İntroduction. Medicinal plants have played a significant role in the development of human civilization. For thousands of years, they have been used to treat health ailments, enhance flavor and preserve food, and prevent disease epidemics [1]. Medicinal plants play an important role in disease prevention, and their promotion and use align with all existing prevention strategies [2].

Across Europe, over 1,300 species of medicinal plants are utilized, with the vast majority (90%) being sourced from wild populations. In the United States, natural products form the basis for approximately 118 of the top 150 prescription medications. Furthermore, plant-based remedies constitute the primary healthcare method for up to 80% of individuals in developing nations. Similarly, in developed countries, more than a quarter (25%) of prescription drugs are derived from wild plant species [3].

Every year, individuals turn to herbal remedies under the belief that they cause fewer adverse side effects. In the United States, approximately 8% of hospital admissions result from negative responses to synthetic drugs, with at least 100,000 deaths occurring annually due to such toxic effects [4]. More than 50,000 plant species-representing over one-tenth of all known species-are utilized in pharmaceutical and cosmetic products. However, the global distribution of medicinal plants is uneven, and they are primarily harvested from wild populations. In fact, in recent decades, the demand for wild plant resources has risen by 8% to 15% annually in regions such as Europe, North America, and Asia [5].

Medicinal plants contain various organic compounds that exert specific physiological effects on the human body. These bioactive substances include tannins, alkaloids, carbohydrates, terpenoids, steroids, and flavonoids. Such compounds are produced through the secondary metabolism of living organisms. Secondary metabolites are chemically diverse and often have unclear taxonomic or functional roles, yet they are widely applied in human medicine, veterinary science, agriculture, research, and numerous other fields [6]. Research conducted over the past two to three decades indicates that these phytochemicals play a significant role in preventing chronic diseases, including cancer, diabetes, and coronary heart disease. Major classes of phytochemicals with disease-preventive properties include dietary fiber, antioxidants, anticancer agents, detoxifying substances, immune enhancers, and neuropharmacological compounds [7].

Among these, flavonoids are recognized for their strong antioxidant, anti-inflammatory, antibacterial, and anticarcinogenic activities, as well as their ability to regulate free radicals. Consequently, they are utilized in the treatment of various diseases [8]. Coumarins, another group of bioactive compounds, have been shown to possess anti-inflammatory, anticoagulant, antibacterial, antifungal, antiviral, anticancer, antihypertensive, antituberculosis, antioxidant, and neuroprotective effects [9]. Additionally, terpenoids are reported in the literature to exhibit a range of pharmacological activities [10].

A variety of methods are employed to identify biologically active compounds in plants, with chromatographic analysis being one of the most commonly used techniques. Chromatography operates on the principle of separating molecules within a mixture by applying them to a solid surface or solid phase and then mobilizing them using a liquid or gaseous mobile phase [11]. The fundamental mechanism underlying all chromatographic methods relies on two key components: the stationary phase and the mobile phase [12]. Paper chromatography is a specific analytical technique used to separate colored substances or compounds, in which paper serves as the stationary phase [13].

In this method, the cellulose fibers of the paper function as the stationary phase, while a solvent, referred to as the developing liquid or eluent, acts as the mobile phase [14]. It is important to note that the behavior of the stationary phase may vary depending on the nature and solubility of each compound being analyzed. Despite its utility, paper chromatography has certain limitations. One drawback is that different compounds may exhibit identical retention values when using the same solvent system. Furthermore, the reproducibility of the method can be influenced by factors such as solvent temperature, the quality of the chromatography paper, solvent purity, and the amount of sample applied.

These limitations have contributed to a decline in both the popularity and the range of applications of this method over time [15].

For this reason, paper chromatography was employed for the preliminary screening of the phytochemical composition of selected species within the *Prangos* genus, which serves as the focus of this research. The *Prangos* genus consists of perennial herbs belonging to the Apiaceae family. Within the flora of Azerbaijan, the Apiaceae family comprises 75 genera and 184 species, while in the flora of the Nakhchivan Autonomous Republic, it includes 57 genera and 107 species. The *Prangos* genus itself is represented by five species in this region [16]. Most of these species are aromatic plants characterized by hollow stems, and many are utilized as vegetables or spices [17]. *Prangos ferulacea* (L.) Lindl., commonly known as "common chashir," can reach heights of up to 150 cm. It features light yellow flowers and emits an unpleasant odor. Its umbels consist of 6 to 16 rays, and the length of its oval fruits ranges from 15 to 20 mm. Flowering typically occurs in May and June, with fruit production taking place from June to August [16].

In contrast, *Prangos acaulis*, referred to as "stemless chashir," grows to a height of 20 to 40 cm. Although many species within the genus have been extensively investigated in previous studies [18], research on *P.acaulis* remains quite limited. In the present study, preliminary phytochemical analyses of *P.acaulis* were conducted using paper chromatography, and a comparative analysis with *P. ferulacea* was also carried out.

Materials and Methods. *P. acaulis* specimens were collected in May 2025 from the Ordubad district of the Nakhchivan Autonomous Republic, specifically at the foot of Soyuqdag near the bank of the Diyachay River. *P. ferulacea* samples were collected in May 2025 from the Kalbajar district. Following collection, the plant materials were thoroughly dried in open air, protected from direct sunlight. Once dried, they were stored in dark glass containers placed in a cool, dark location until extract preparation.

The percolation method was employed to obtain plant extracts. Both dried and powdered plant samples were accurately weighed to 20 g each. The measured plant material was soaked in 20 ml of 70% ethanol and allowed to stand for 24 hours. Subsequently, the soaked material was transferred to a separating funnel, and additional 70% ethanol was added until a mirror layer formed. After another 24-hour standing period, the liquid was completely filtered and returned to the separating funnel. From the 20 g sample, 80% was collected as the primary extract at a rate of approximately 40 drops per hour and set aside. To complete the extraction process, 70% ethanol was continuously added to the separating funnel containing the plant material until the resulting extract became colorless. The extract was then evaporated until a 20% dry residue was achieved and combined with the primary extract. To remove ballast substances, the combined extract was refrigerated for 24 hours, filtered through filter paper, and prepared for subsequent use.

For the chromatographic analysis, Whatman No.1 paper was used as the stationary phase. The following solvent systems were employed as mobile phases: chloroform:methanol:water (7:3:0.4), chloroform:butanol:water (8:1:1), and butanol:acetic acid:water (4:1:5). For each analysis, the paper was cut into pieces measuring 9–13 cm in length and 6–8 cm in width, and a starting line was drawn 2 cm from the bottom edge. Using a micropipette, extracts obtained from the leaves and roots of the plants were applied separately onto the starting line in multiple applications, allowing each spot to dry between applications. After the spots were completely dried, the paper was placed into a chromatography chamber that had been previously saturated with mobile phase vapors.

The development process was allowed to proceed for 45 to 90 minutes, during which the migration of the compounds was observed. Following development, the paper was removed from the chamber, dried, and then exposed to ammonia vapor to enhance the visibility of the separated spots. For the identification of biologically active compounds, chemical reagents such as 1% aluminum chloride ($AlCl_3$) and 1% iron(III) chloride ($FeCl_3$) were used. Additionally, spots that were not visible under normal light were detected using

UV light at a wavelength of 365 nm. Finally, the retention factor (Rf) values of the detected substances were calculated and compared.

Results. In the present study, the chromatographic analysis revealed the presence of several components in the plant samples, and successful separation of these compounds was achieved. The chromatograms obtained from leaf and root extracts of *Prangos acaulis* and *Prangos ferulacea*, developed using different solvent systems, are presented below in Figures 1–3.

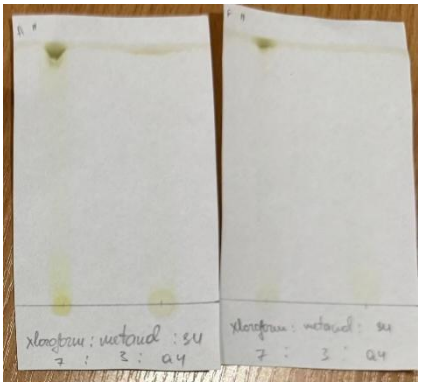

Figure 1. Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of chloroform, methanol, and water in a ratio of 7:3:0.4

In the chromatogram developed using the chloroform:methanol:water (7:3:0.4) solvent system, a green spot with an Rf value of 0.92 was observed in the leaf extract of *Prangos acaulis*, while the yellow component remained almost at the baseline, showing minimal migration. In the root extract of the same species, a yellow spot with an Rf value of 0.06 was detected, indicating very limited movement. For *Prangos ferulacea*, the leaf extract exhibited a green spot with an Rf value of 0.94, with the yellow component again showing negligible migration. In the root extract, a yellow spot with an Rf value of 0.05 was observed, also reflecting very low mobility.

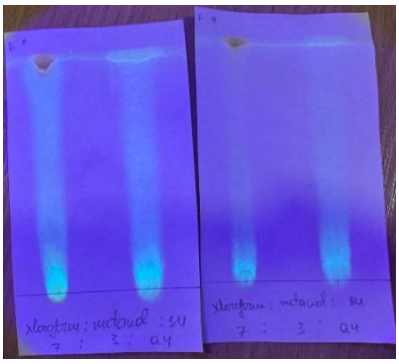

Figure 1 (a). Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of chloroform, methanol, and water in a ratio of 7:3:0.4, observed under UV light

In the chromatogram developed using the chloroform:methanol:water (7:3:0.4) solvent system, previously invisible spots were detected in both the leaf and root extracts of the two plant species. These spots exhibited blue fluorescence and a brown coloration when visualized under UV light at a wavelength of 365 nm.

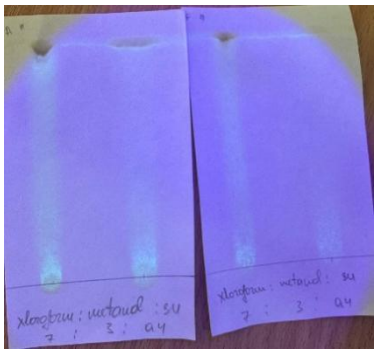

Figure 1 (b). Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of chloroform, methanol, and water in a ratio of 7:3:0.4, treated with $AlCl_3$ and visualized under UV light

After treating the chromatogram of the leaf and root extracts of both plants, developed in the chloroform:methanol:water (7:3:0.4) system, with 1% $AlCl_3$ solution and visualizing under UV light, yellow dotted spots were observed along the migration path of the compounds, in addition to the previously noted blue fluorescence and brown spot.

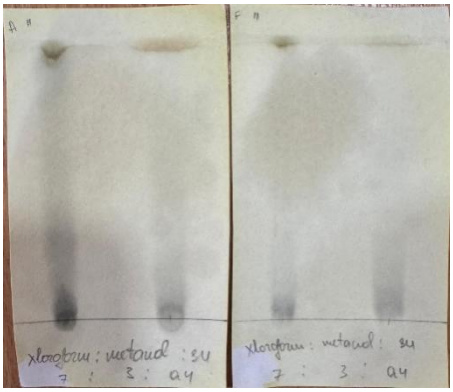

Figure 1 (c). Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of chloroform, methanol, and water in a ratio of 7:3:0.4, treated with $FeCl_3$

Upon treatment of the chromatogram of the leaf and root extracts of both plants, developed in the chloroform:methanol:water (7:3:0.4) system, with 1% $FeCl_3$ solution, blue spots were observed along the migration path of the compounds.

Figure 2. Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of chloroform, butanol, and water in a ratio of 8:1:1

In the chromatogram developed using the chloroform:butanol:water (8:1:1) solvent system, a green spot with an Rf value of 0.85 was observed in the leaf extract of *Prangos acaulis*, while the yellow component remained almost at the baseline, showing negligible migration. In the root extract of the same species, a yellow spot with an Rf value of 0.06 was detected, indicating very limited movement. For *Prangos ferulacea*, the leaf extract exhibited a green spot with an Rf value of 0.45, with the yellow component again showing minimal migration. In the root extract, a yellow spot with an Rf value of 0.7 was observed.

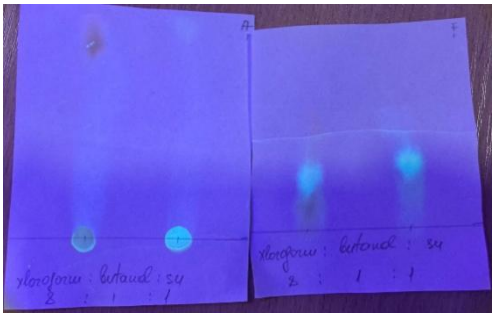

Figure 2 (a). Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of chloroform, butanol, and water in a ratio of 8:1:1, observed under UV light

In the chromatogram developed using the chloroform:butanol:water (8:1:1) solvent system, previously invisible spots were detected in both the leaf and root extracts of the two plant species. These spots exhibited blue fluorescence and a brown coloration when visualized under UV light at a wavelength of 365 nm.

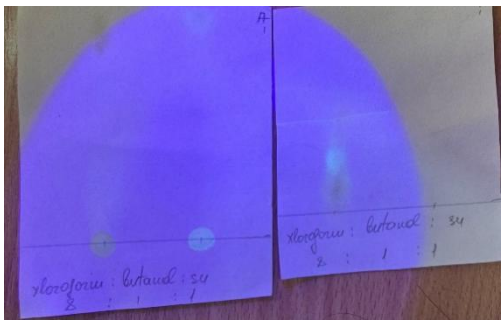

Figure 2 (b). Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of chloroform, butanol, and water in a ratio of 8:1:1, treated with $AlCl_3$ and visualized under UV light

After treating the chromatogram of the leaf and root extracts of both plants, developed in the chloroform:butanol:water (8:1:1) system, with 1% $AlCl_3$ solution and visualizing under UV light, blue fluorescence and brown spots were observed.

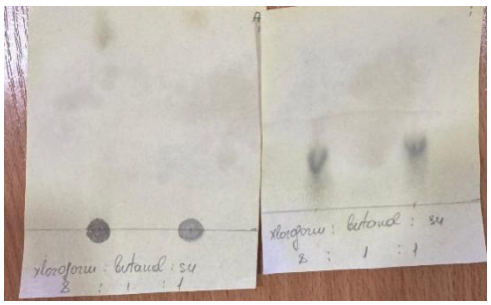

Figure 2 (c). Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of chloroform, butanol, and water in a ratio of 8:1:1, treated with $FeCl_3$

Upon treatment of the chromatogram of the leaf and root extracts of both plants, developed in the chloroform:butanol:water (8:1:1) system, with 1% $FeCl_3$ solution, blue spots were observed along the migration path of the compounds.

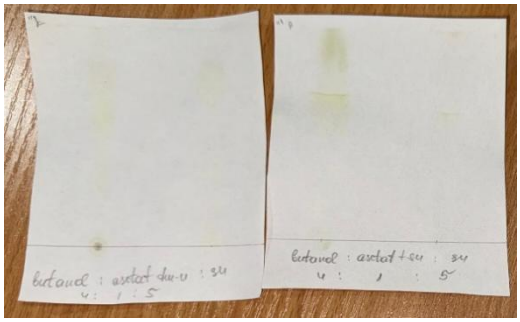

Figure 3. Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of butanol, acetic acid, and water in a ratio of 4:1:5

In the chromatogram developed using the butanol:acetic acid:water (4:1:5) solvent system, a green spot with an Rf value of 0.8 was observed in the leaf extract of *Prangos acaulis*, while the yellow component remained almost at the baseline, showing negligible migration. In the root extract of the same species, a yellow spot with an Rf value of 0.96 was detected. For *Prangos ferulacea*, the leaf extract exhibited a green spot that remained at the origin (Rf = 0) and a yellow spot with an Rf value of 0.8. In the root extract, a yellow spot with an Rf value of 0.8 was also observed.

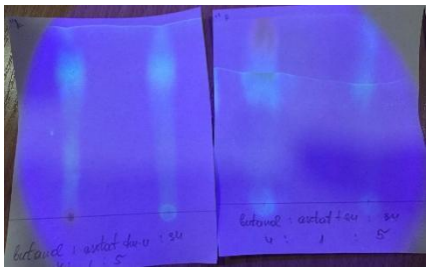

Figure 3 (a). Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of butanol, acetic acid, and water in a ratio of 4:1:5, observed under UV light

In the chromatogram developed using the butanol:acetic acid:water (4:1:5) solvent system, previously invisible spots were detected in both the leaf and root extracts of the two plant species. These spots exhibited blue fluorescence and a brown coloration when visualized under UV light at a wavelength of 365 nm.

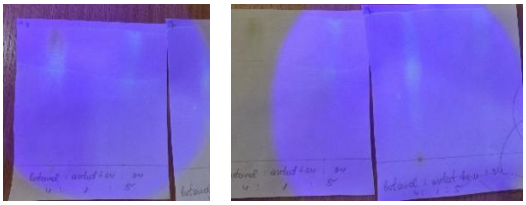

Figure 3 (b). Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of butanol, acetic acid, and water in a ratio of 4:1:5, treated with $AlCl_3$ and visualized under UV light

After treating the chromatogram of the leaf and root extracts of both plants, developed in the butanol:acetic acid:water (4:1:5) system, with 1% $AlCl_3$ solution and visualizing under UV light, blue fluorescence and brown spots were observed.

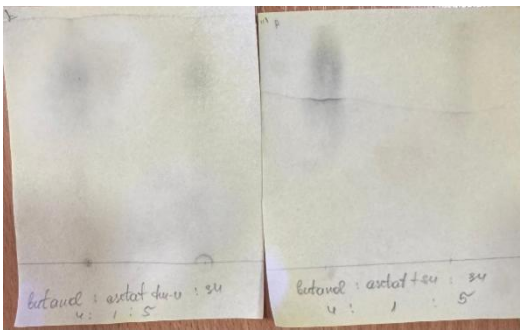

Figure 3 (c). Chromatogram of leaf and root extracts obtained from *Prangos acaulis* and *Prangos ferulacea*, developed using a solvent system consisting of butanol, acetic acid, and water in a ratio of 4:1:5, treated with $FeCl_3$

Upon treatment of the chromatogram of the leaf and root extracts of both plants, developed in the butanol:acetic acid:water (4:1:5) system, with 1% $FeCl_3$ solution, blue spots were observed along the migration path of the compounds.

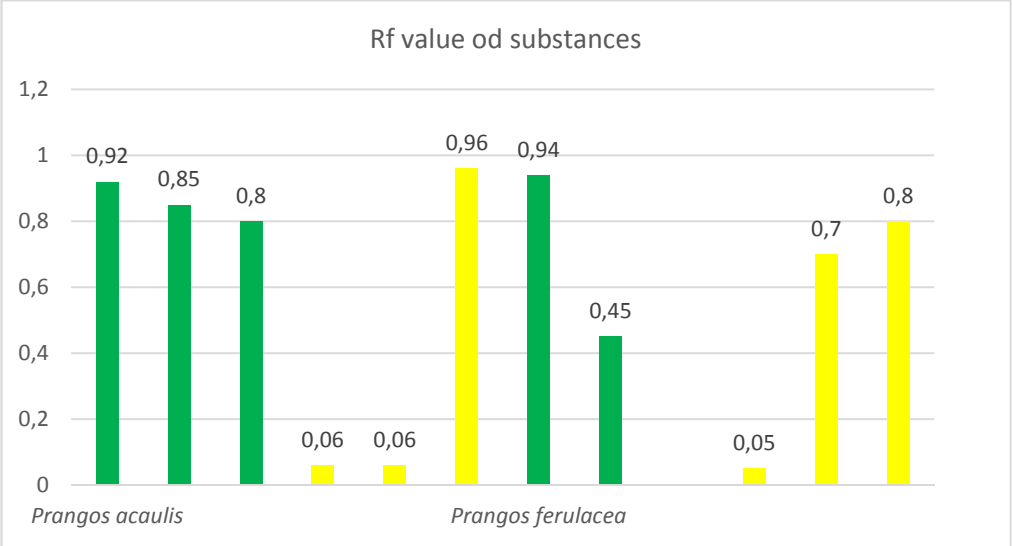

Diagram 1. Rf values calculated from the chromatograms of extracts obtained from *Prangos acaulis* and *Prangos ferulacea*

Order of the solvent systems: chloroform:methanol:water – chloroform:butanol:water – butanol:acetic acid:water

Discussion. The evaluation of the obtained results suggests that the chromatographic profiles of the extracts, in combination with the specific reagents employed, provide insight into the possible functional groups present in the separated compounds. Examination of the chromatograms obtained from different solvent systems, as presented in Figures 1, 2, and 3, indicates that the spots observed in both studied plant species may be associated with photosynthetic pigments such as chlorophyll a, chlorophyll b, and carotenoids.

As previously noted, the chromatograms shown in Figures 1a, 2a, and 3a were visualized under UV light at a wavelength of 365 nm. Across all three solvent systems, spots

that were not visible to the naked eye exhibited blue fluorescence and brown coloration under UV illumination. Based on available literature, such fluorescence characteristics suggest the potential presence of coumarins, flavonoids, and other phenolic compounds [19].

In Figures 1b, 2b, and 3b, the chromatograms were treated with $AlCl_3$ and subsequently visualized under UV light. A closer examination of Figure 1b reveals the presence of yellow dotted spots along the migration path of the compounds. Based on findings from previous studies indicating that such spots are characteristic of flavonoids [20], it can be inferred that the investigated plant species may also be rich in flavonoid compounds. It is worth noting, however, that similar spots were not observed in the chromatograms presented in Figures 2b and 3b, which may suggest that these compounds did not dissolve effectively in the respective solvent systems used.

In Figures 1c, 2c, and 3c, the chromatograms were treated with $FeCl_3$. Following this treatment, the spots exhibited a blue coloration. This color change is indicative of the possible presence of phenolic compounds within the plant extracts [21].

As previously noted, while information regarding the phytochemical composition of *P. acaulis* remains limited, the chemical profile of *P. ferulacea* has been extensively documented in the literature [22, 23]. Given the well-established bioactive properties and rich phytochemical content of *P. ferulacea*, a comparison of the chromatograms obtained from both species in this study reveals overlapping spots. This similarity suggests that *P. acaulis* may also possess a diverse array of bioactive compounds. Nevertheless, it should be emphasized that further comprehensive and detailed investigations are necessary to accurately identify the specific classes of bioactive substances present and to determine the exact molecular composition of this species.

Conclusion. In the present study, a preliminary phytochemical analysis was conducted on 70% ethanol extracts obtained from the leaves and roots of *Prangos acaulis* using the percolation method. The analysis was carried out by means of paper chromatography, and the results were compared with those of *Prangos ferulacea*. The chromatographic profiles obtained indicate that both plant species may be rich in biologically active compounds, including phenols, flavonoids, and coumarins.

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ПОПЕРЕДНІЙ ФІТОХІМІЧНИЙ АНАЛІЗ PRANGOS ACAULIS (LINDL.) BORNM. ТА ПОРІВНЯЛЬНИЙ АНАЛІЗ ІЗ PRANGOS FERULACEA (L.) LINDL.

Прангос безлокий (Prangos acaulis) – це багаторічна трав'яниста рослина родини Зонтичних (Apiaceae). Цей рід поширений переважно в сухому та напівпустельному кліматі. Було вивчено численні види роду Прангос, проведено численні фітохімічні аналізи, які показали, що вони багаті на біологічно активні сполуки. Однак достатньої інформації щодо Прангоса безлокатого (Prangos acaulis) бракує. Основні класи фітохімічних речовин з профілактичними властивостями включають харчові волокна, антиоксиданти, протиракові засоби, детоксикуючі речовини, імуностимулятори та нейрофармакологічні сполуки. Потенційне багатство фітохімічного складу цього виду, враховуючи його фармакологічні властивості, свідчить про те, що він може стати майбутньою сировиною для медицини, і з цієї причини вивчення рослини є важливим. Метою дослідження є проведення фітохімічного аналізу видів P.acaulis та P.ferulacea за допомогою паперової хроматографії, проведення попереднього скринінгу на P. acaulis та оцінка отриманих результатів у порівнянні з P.ferulacea. Для отримання рослинних екстрактів використовували метод перколяції. Як висушені, так і порошкоподібні зразки рослин точно зважували до 20 г кожен. Виміряний рослинний матеріал замочували у 20 мл 70% етанолу та давали настоятися 24 години. Після ще одного 24-годинного періоду відстоювання рідину повністю фільтрували та повертали у ділильну лійку. Зі зразка масою 20 г 80% збирали у вигляді первинного екстракту зі швидкістю приблизно 40 крапель на годину та відставляли. Для проведення дослідження зібрані рослини сушили за ідентичних умов, подрібнювали на порошок, отримували екстракти та проводили аналіз за допомогою паперової хроматографії.

Оцінка отриманих результатів свідчить про те, що хроматографічні профілі екстрактів у поєднанні зі специфічними реагентами, що використовуються, дають уявлення про можливі функціональні групи, присутні в розділених сполуках. У цьому дослідженні було проведено попередній фітохімічний аналіз 70% етанольних екстрактів, отриманих з листя та коренів Prangos acaulis, за допомогою методу перколяції. Ця подібність свідчить про те, що P. acaulis також може мати різноманітний спектр біологічно активних сполук. Тим не менш, слід наголосити, що необхідні подальші комплексні та детальні дослідження для точної ідентифікації конкретних класів біологічно активних речовин, що присутні, та визначення точного молекулярного складу цього виду.

Ключові слова: Prangos acaulis, Prangos ferulacea, паперова хроматографія, фітохімічний аналіз.

Стаття до редакції надійшла 01.04.2026 року
Рецензія на статтю надійшла 13.04.2026 року