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ANALYSIS OF THE CHEMICAL COMPOSITION OF THE ROOTS OF DOG ROSEHIP (ROSA CANINA) GROWING IN KATON-KARAGAY NATIONAL PARK

7M10104 – «Pharmacy»

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NORMATIVE REFERENCES

The following standards are referenced in the master's thesis:

1. State Pharmacopoeia of the Republic of Kazakhstan. Volumes 1, 2, 3. – Almaty: Zhibek Zholy Publishing House, 2008, 2009, 2014. Articles applied in the study: "Roots, Rhizomes, Bulbs, Tubers, and Corms" (general article), 2.2.32 (Moisture Content), 2.4.16 (Total Ash), 2.8.1 (Ash Insoluble in Hydrochloric Acid).

2. GOST 24027.2-80. Medicinal Plant Raw Materials. Methods for Determination of Moisture Content, Ash, Extractable Substances, Tannins, and Essential Oil Content.

3. GOST R ISO 9768-2011. Tea. Determination of Water-Extractable Substances.

4. GOST R ISO 14502-1-2010. Tea. Determination of Total Polyphenol Content. Spectrophotometric Method Using the Folin–Ciocalteu Reagent.

5. GOST 24556-89. Products Processed from Fruits and Vegetables. Methods for Determination of Vitamin C.

6. State Pharmacopoeia of the Russian Federation, 14th Edition.

7. WHO Guidelines on Good Agricultural and Collection Practices for Medicinal Plants.

DEFINITIONS

Medicinal plant raw material – dried or fresh parts of plants containing therapeutically relevant concentrations of pharmacologically active substances and complying with the requirements of the State Pharmacopoeia or applicable regulatory and technical documentation.

Biologically active substances – a collective term for chemical compounds synthesized in plant organisms that, at low molecular concentrations, exert specific effects on the physiological functions of living organisms (humans, animals).

Phytochemical analysis – a system of analytical methods aimed at identifying, isolating, and characterizing groups of chemical compounds in plant raw material both qualitatively and quantitatively.

Macroscopic analysis – a method for determining the external characteristics of medicinal plant raw material (shape, color, odor, taste) by visual inspection with the naked eye or with the aid of a magnifying loupe.

Microscopic analysis – a type of analysis based on the identification of characteristic diagnostic features of plant cells and tissues using a microscope.

Pharmacognostic quality analysis – an investigation conducted to determine the quality, moisture content, ash content, and impurity levels of raw material in compliance with applicable regulatory documents.

ABBREVIATIONS AND SYMBOLS

SP RK – State Pharmacopoeia of the Republic of Kazakhstan
RTD – regulatory and technical documentation
WHO – World Health Organization
GACP – Good Agricultural and Collection Practice
BAS – biologically active substances
BAFS – biologically active food supplement
MPRM – medicinal plant raw material
MP – medicinal preparation
UV – ultraviolet
SRS – state reference standard

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INTRODUCTION

Relevance of the study. At the present stage, one of the priority directions of state policy in the pharmaceutical sector of the Republic of Kazakhstan is the strengthening of public health and the reduction of dependence on imports in domestic pharmaceutical production. In accordance with the priorities of the "National Development Plan of the Republic of Kazakhstan until 2029" and the "Concept for the Development of the Pharmaceutical and Medical Industry for 2025–2029," the expansion of the nomenclature of high-quality, safe, and accessible medicinal products based on domestic plant resources is of strategic importance.

The natural and climatic conditions of the Republic of Kazakhstan provide ample opportunities for the widespread utilization of plant resources rich in biologically active compounds; however, the chemical composition of certain morphological parts of these plants has not been sufficiently studied. In this regard, the investigation of the roots of dog rosehip (*Rosa canina* L.) growing in the high-mountain ecosystems of Katon-Karagay National Park holds scientific and practical significance. Although the fruits of this species – rich in vitamins and antioxidants – constitute pharmacopoeial raw material and are used in medicine as antioxidant, wound-healing, and choleretic agents, data on the chemical composition and pharmacological activity of the root system remain limited. Furthermore, the tendency of biologically active substances in the fruits to undergo oxidative processes during storage reduces their therapeutic efficacy. The components found in the roots, by contrast, are chemically more stable, as the roots are protected from factors that promote the degradation of active substances – such as sunlight, moisture, and temperature fluctuations. According to contemporary scientific research, the antioxidant activity of root extracts is three times higher than that of the fruits (Macit et al., 2023). In addition, the root system accumulates specific phenolic compounds, such as epicatechin, gallic, syringic, *p*-coumaric, and ellagic acids, which are absent in the fruits (Subas et al., 2025).

Conducting a chemical analysis of the roots of dog rosehip growing in Katon-Karagay National Park will enable the identification of groups of compounds – including flavonoids, tannins, organic acids, saponins, and others – that exhibit antioxidant, anti-inflammatory, and antimicrobial activity. These data will form the scientific basis for the standardization of dog rosehip's roots as a new medicinal plant raw material and for their introduction into pharmaceutical circulation.

The aim of the research:

To study the chemical composition of the roots of dog rosehip (*Rosa canina* L.) growing in the Katon-Karagay National Park.

Research objectives:

1. To conduct a qualitative analysis of the main groups of biological active substances of the roots of dog rosehip.
2. To perform a quantitative analysis of the identified bioactive substances of the roots of dog rosehip.

3. To compare the results of the analysis with the chemical composition of fruits of dog rosehip.

Research materials:

Dried roots of dog rosehip (*Rosa canina* L.) collected in Katon-Karagai National Park during the autumn of 2025, at the end of the vegetative stage. The medicinal plant raw material was stored in a dry, well-ventilated area protected from light at a temperature of +15 °C to +25 °C and relative humidity not exceeding 60%.

Research methods:

Phytochemical analysis: qualitative (chemical methods) and quantitative (spectrophotometry, titrimetry) determination.

Scientific novelty:

For the first time, a comprehensive phytochemical profile of the roots of dog rosehip (*Rosa canina* L.) growing in the high-altitude conditions of the Katon-Karagay National Park has been established. New comparative data on the distribution of bioactive substances between the roots and fruits of the Kazakhstan population of *Rosa canina* L. have been obtained. The study identifies specific quantitative markers that define the unique phytochemical signature of the East Kazakhstan population, contributing significantly to the global geographical chemotaxonomy of the species.

Practical significance:

The results of the study into the qualitative and quantitative composition of bioactive compounds in roots of *Rosa canina* L. may provide a scientific basis for expanding the range of medicinal plant materials used in pharmaceutical practice and provide a foundation for further research.

Research base:

NJSC “Medical University of Astana”. Department of Pharmaceutical Disciplines.

Main provisions for defense:

1. The anatomical and morphological diagnostic characteristics of *Rosa canina* L. roots grown in the high-altitude conditions of Katon-Karagay National Park have been identified, and the authenticity of the raw material has been confirmed.

2. The quality indicators of the raw material were determined within pharmacopoeial requirements: moisture content – 7.41%, total ash – 3.63%, HCl-insoluble ash – 1.24%, extractive substances – 11.46%.

3. The qualitative phytochemical composition of *Rosa canina* L. roots has been established, including polyphenolic compounds, flavonoids, tannins, catechins, triterpenoids, saponins, coumarins, organic acids, and amino acids.

4. The quantitative content of biologically active substances was determined using spectrophotometric and titrimetric methods: polyphenols – 3.62%, flavonoids – 0.83%, tannins – 0.42%, vitamin C – 0.065%.

5. Comparative analysis of the chemical composition of roots and fruits revealed the accumulation of specific biomarkers in the roots (ellagic acid, gallic acid, and epicatechin) which are absent in the fruits.

6. The phytochemical profile of the Katon-Karagay population was compared with data from Russia, Ukraine, and Turkey, revealing distinct geographical variability.

Volume and structure of the master's thesis:

The master's thesis comprises 66 pages of computer-generated text, supplemented by 19 tables and 12 figures. The structure of the work consists of an introduction, 3 chapters, results, conclusions, and a list of references containing 115 literary sources.

Approbation of the master's thesis was conducted at the Department of Pharmaceutical Sciences, Astana Medical University.

The main principles and results of the master's thesis were presented at international scientific-practical conferences and reflected in published scientific articles:

1. A review article entitled "Prospects of Using the Roots of Dog Rose (*Rosa canina* L.) in Pharmacy" was published in the proceedings of the XII International Scientific and Practical Conference "World Globalization: Fundamental and Applied Aspects" (in Russian).

2. A scientific thesis entitled "Pharmacognostic Characterization of Dog Rosehip (*Rosa canina* L.) Roots from the Katon-Karagay National Park (Kazakhstan)" was published in the proceedings of the International Scientific and Practical Conference "Sander Readings."

3. A scientific thesis entitled "Comparative Chemical Analysis of the Roots and Fruits of Rose (*Rosa canina* L.) from the Katon-Karagay Region" was published in the proceedings of the IX International Scientific and Practical Conference "Abu Ali ibn Sino and Innovations in Modern Pharmaceutics."

4. An article entitled "Quantitative Analysis of Bioactive Compounds in the Roots of *Rosa canina* L. (Katon-Karagay) in Comparison with Published Data" has been submitted to the CQASHE-recommended journal "Kazakhstan Pharmacy", which is included in the list of scientific publications recommended by the Science and Higher Education Quality Assurance Committee of the Ministry of Science and Higher Education of the Republic of Kazakhstan.

LITERATURE REVIEW

1.1 General Characteristics of Plants of the Genus *Rosa*

The genus *Rosa* L. – belonging to the family Rosaceae and the order Rosales – is one of the largest and most polymorphic groups in the plant kingdom. The name of the genus was first described by Carl Linnaeus in his work *Species Plantarum*, in which he included 12 species known at the time [1]. The first systematic classification of the genus was proposed by A.P. De Candolle (1818), who divided it into 11 groups: *Synstylees*, *Rubiginees*, *Galicanes*, *Chinoises*, *Cannelles*, *Hebeclades*, *Pimprenelles*, *Yelves*, *Acent-Feuilles*, *Caninae*, and *Eglantiers* [2]. According to current modern botanical sources, based on the APG IV system, 405 confirmed species have been recorded, while 2,595 names remain pending confirmation of their taxonomic status [3]. The number of cultivated varieties, according to some sources, reaches up to 10,000, while other data indicate figures of 25,000 and even 50,000 [4-7].

Species of *Rosa* are predominantly distributed across the temperate and subtropical climatic zones of the Northern Hemisphere. Their principal genetic centers are East Asia (China, Japan), Central Asia, and the Near East [8-11].

In the flora of Kazakhstan, 25 species of the genus *Rosa* are found, four of which are endemic (Ili rosehip (*Rosa iliensis* Chrshan.), Zharkent rosehip (*Rosa dsharkentica* Chrshan.), Pavlov's rosehip (*Rosa pavlovii* Chrshan.), glabrous-leaved rosehip (*Rosa glabrifolia* C.A. Mey. ex Ledeb.) [12], and three (many-flowered rosehip (*R. multiflora*), Chinese rosehip (*R. chinensis*), white rosehip (*R. alba*)) have been introduced from other countries, such as Japan and China [13].

Classification of *Rosa* species by ecological habitat:

- Forest zones: bristly rosehip (*R. acicularis*), Albert's rosehip (*R. alberti*), may rose (*R. majalis*), glabrous-leaved rosehip (*R. glabrifolia*), loose rosehip (*R. laxa*), dog rosehip (*R. canina*);
- Mountain and submontane zones: sharp-thorned rosehip (*R. oxyacantha*), Schrenk's rosehip (*R. schrenkiana*), glabrous-leaved rosehip (*R. glabrifolia*), loose rosehip (*R. laxa*), Begger's rosehip (*R. beggeriana*), Fedchenko's rosehip (*R. fedtschenkoana*), dog rosehip (*R. canina*), Kokand rosehip (*R. kokanica*), burnet rosehip (*R. spinosissima*), corymb rosehip (*R. corymbifera*), broad-thorned rosehip (*R. platyacantha*);
- Riverbanks and water bodies: may rose (*R. majalis*), Pavlov's rosehip (*R. pavlovii*), Begger's rosehip (*R. beggeriana*), Ili rosehip (*R. iliensis*), dog rosehip (*R. canina*), corymb rosehip (*R. corymbifera*);
- Steppes, meadows, sandy and gravelly-stony slopes: glabrous-leaved rosehip (*R. glabrifolia*), Ili rosehip (*R. iliensis*), Zharkent rosehip (*R. dsharkentica*), Hissar rosehip (*R. hissarica*), Samarkand rosehip (*R. samarkandensis*), broad-thorned rosehip (*R. platyacantha*) [14, 15].

According to literary sources, species of the genus *Rosa* L. are evergreen or deciduous shrubs with erect, procumbent, or arcuately curved stems, with or without underground shoots, ranging in height from 0.2 m to 3–5 m (certain liana-

like species reaching up to 10 m). In the first year of growth, rosehip forms vegetative shoots, and from the second year onward the plant begins to flower and fruit. The stems, and in most cases the petioles as well, bear prickles of varying shape and size. The leaves are alternate, imparipinnate, with elliptical or ovate leaflets and serrate margins.

The flowers are bisexual, solitary or gathered into corymb- or panicle-like inflorescences, and most commonly bear petals. The hypanthium is globose, ovoid, or ellipsoid in shape; its upper portion bears a more or less developed nectariferous disc. During the fruiting period, the sepals – pinnately lobed, leaf-like, or with entire margins – may be persistent or deciduous. The number of petals is 4–5; their color is yellow, white, or pink, and red hues also occur. The numerous stamens bear two-locular anthers. The gynoecium of *Rosa* consists of numerous sessile or shortly stipitate carpels located on the lower inner walls of the hypanthium; they are hairy, unilocular, and contain a single anatropous ovule. The stylodium (glabrous or hairy, free or connate) is situated mostly within the hypanthium, but may occasionally protrude beyond the inner circle of stamens. The stigma is capitate in shape. The fruits are globose or ovoid, glabrous, orange-red or red, fleshy, and contain numerous achenes [16-18].

In the identification of species diversity within the genus *Rosa* in the flora of Kazakhstan, the morphological structure of fruits and roots serves as the principal diagnostic indicator. In this regard, based on a systematization of literary data, the distinguishing characteristics of *Rosa* species are summarized in Tables 1 and 2.

Table 1. Diagnostic characteristics of fruits of *Rosa* species

Species name	**Indumentum**	**Shape**	**Color**
Dog rosehip (*Rosa canina*)	Glabrous	Ovoid, diameter approx. 2 cm (sepals deciduous at maturity)	Red
May rose (*Rosa majalis*)	Glabrous	Globose, diameter approx. 2 cm (sepals persistent)	Red
Bristly rosehip (*Rosa acicularis*)	Upper portion of fruit covered with glandular bristles	Ovoid or ellipsoid, diameter 1.5–2.5 cm	Red
Begger's rosehip (*Rosa beggeriana*)	Glabrous, fine	Globose, diameter 0.5–1 cm	Dark red or black-red
Fedchenko's rosehip (*Rosa fedtschenkoana*)	Glandular-pubescent or glabrous	Large, elongated-ovoid	Deep red
Loose rosehip (*Rosa laxa*)	Glabrous	Elongated-ellipsoid	Red

Table 2. Diagnostic characteristics of roots of *Rosa* species

Species name	Color and surface character of root bark	Wood density and fracture	Microscopic diagnostic features
Dog rosehip (*Rosa canina* L.)	Dark brown or reddish-brown; surface deeply and longitudinally furrowed	Wood very dense and heavy; fracture – wood-like, rough, splintery	Large druses of calcium oxalate accumulated in parenchyma cells
May rose (*Rosa majalis* Herrm.)	Dull brown or dark brown; surface with fine, transversely arranged cracks	Wood yellowish in hue, of medium density; fracture – firmly fibrous	Cells of the bark region contain anthocyanin pigments
Bristly rosehip (*Rosa acicularis* Lindl.)	Grayish-brown; bark thin, easily separated from wood, surface relatively smooth	Wood firm but elastic; fracture – fibrous-wood-like	Starch grains accumulated in wood parenchyma
Begger's rosehip (*Rosa beggeriana* Schrenk.)	Pale gray or pale brown; surface smooth, lustrous	Wood porous and light; fracture – uneven, finely rough	Wood rays slender; arrangement of conducting vessels (tracheae) diffuse
Fedchenko's rosehip (*Rosa fedtschenkoana* Regel.)	Brown-red; root thick, fleshy, bark thick	Wood dense; medullary rays clearly visible to the naked eye	Mucilaginous cells and saponin inclusions occur in bark parenchyma
Loose rosehip (*Rosa laxa* Retz.)	Grayish-yellow or yellowish-brown; surface scaly, thin	Wood loose, pale in color; fracture – fibrous, thread-like, extensible	Characterized by a predominance of wide libriform fibers and parenchymatous tissue

1.2 Chemical Composition and Medical Applications of the Genus *Rosa*

The phytochemical composition of plants of the genus *Rosa* may vary even within a single species depending on geographical location, ecology, soil composition, and other environmental factors. In particular, the content of ascorbic acid and phenolic compounds is subject to considerable variability [19, 20]. The results of a retrospective content analysis of literary sources revealed the complexity of the phytochemical composition of rosehip plants:

1. Polyphenolic compounds: this group is represented by flavonols (quercetin, kaempferol, isoquercitrin – 987.8 mg in terms of quercetin), catechins (epigallocatechin, gallocatechin, epigallocatechin gallate, epicatechin gallate), anthocyanin compounds, and gallic acid (3,000 mg/100 g) [21-24].

2. Vitamin complex: rosehip fruits belong to multivitamin raw materials. The content of ascorbic acid ranges from 0.2% to 6% depending on species characteristics [25]. Rosehip fruits represent the richest source of the L-isomer of vitamin C among fruits, berries, and vegetables (from 300 to 4,000 mg/100 g). Additionally, the raw material contains 0.7–8 mg% carotenoids (lutein, β-carotene, lycopene), as well as vitamins B_1, B_2, PP, K, and pantothenic acid [26].

3. Carotenoids: lycopene and β-carotene participate in biochemical reactions by neutralizing the harmful effects of free radicals. Lycopene is a potent antioxidant and antimicrobial agent capable of preventing degenerative diseases such as cancer of the lung, bladder, cervix, prostate, breast, and skin, as well as atherosclerosis and ischemic heart disease [27-30].

4. Other groups of biologically active substances [BAS]: pectic substances (up to 4%), sugars (up to 18%), macro- and microelements (iron, phosphorus, manganese, magnesium, calcium, etc.), as well as organic acids (up to 4%) [31].

Rosehip oil contains γ- and δ-tocopherols, unsaturated fatty acids (omega-3 – α-linolenic; omega-9 – oleic; omega-6 – linoleic), and carotenoids. The proportion of polyunsaturated fatty acids in the oil reaches up to 80%. The oil of low-vitamin species belonging to section *Caninae* consists of 50% triglycerides of monounsaturated oleic acid, whereas in species of section *Cinnamomeae* the content of polyunsaturated acids is increased [32].

The phytochemical composition of rosehip is variable and changes depending on the age of the plant and the developmental stage of the flower. The distribution of BAS groups among the morphological organs of the plant is as follows:

- Fruits: primarily accumulate polyunsaturated fatty acids (PUFA), flavonoids, triterpenoids, and phytosterols [19, 26, 27]. Galactolipids present in the composition exhibit anti-inflammatory and antitumor activity [33].
- Flower buds (*R. rugosa*): contain acidic polysaccharides with antioxidant and rejuvenating properties, neuroactive depsidic glucosides, flavonoids, and tannins [34, 35].
- Leaves: in the leaves of various *Rosa* species, polyphenols accumulate at levels ranging from 5.70% to 15.20% of dry weight. In particular, it has been demonstrated that the polyphenol content in the leaves of *R. canina* is higher than in its fruits [32, 36].

Compounds isolated from *Rosa* species are used and investigated in official medicine for the treatment of the following diseases:

1. Hepatoprotective and gastroenterological: skin conditions [37] and gastrointestinal tract disorders (including liver diseases) [38-41];
2. Nephroprotective: renal function impairment [42, 43];

3. Anti-inflammatory and endocrine: arthritis [44], diabetes [45], hyperlipidemia [46];

4. Oncological: antitumor activity is associated with the antioxidant properties of phenolic compounds [47, 48]. Isoflavone phytoestrogens present in *R. canina* exhibit activity against breast cancer (MCF-7) [49, 50], while extracts of *R. rugosa* affect the apoptosis of prostate cancer cells [51].

Alcoholic extracts have demonstrated antiviral activity. Furthermore, a methanolic extract of *R. canina* fruits inhibits the conjugation of bacterial plasmids, thereby enabling the overcoming of antibiotic resistance [52].

More than 14 species of rosehip (including *R. alba*, *R. canina*, *R. multiflora*, *R. rugosa*, and others) are used in cosmetic products [53]. For example, an ethanolic extract of *R. multiflora* prevents biochemical damage caused by ultraviolet (UV) radiation leading to photoaging by reducing reactive oxygen species, interleukin (IL)-6, IL-8, and matrix metalloproteinase (MMP)-1 [54]. *R. canina* fruit powder (containing seeds and hypanthium) increases cell viability, reduces wrinkles, moisturizes the skin, and improves its elasticity [55]. An extract of *R. gallica* petals reduces UV radiation-induced MMP-1 expression, which is a principal marker of wrinkle formation [56]. Polyphenols in *R. canina*, *R. gallica*, and *R. rugosa* (quercetin, kaempferol, ellagic acid) inhibit melanin synthesis, thereby producing a skin-lightening effect [57, 58].

Rosehip fruits are used in official medicine as a vitamin preparation in the form of infusions, extracts, syrups, tablets, and dragées. Table 3 presents the chemical composition, medical applications [59-61], and raw material reserves [62–64] of *Rosa* species found in Kazakhstan.

Table 3. Chemical, pharmacological, and resource characteristics of *Rosa* species used in official medicine in Kazakhstan

Species name (raw material)	**Chemical composition**	**Pharmacological activity**	**Raw material reserves**
R. canina L. – dog rosehip (fruits)	Vitamin C, triterpenoids, phenolcarboxylic acids, flavonoids, tannins, carotenoids, anthocyanins, fixed oils, tocopherols	Choleretic, adaptogenic, hemostatic, diuretic, detoxifying, general tonic	Ili, Zhetysu Alatau, East Kazakhstan (South Altai). Cultivated at the Institute of Botany and Phytointroduction of the Republic of Kazakhstan
R. majalis Herrm. – may rose (fruits)	Vitamins C and P, carotenoids, catechins, flavonoids, anthocyanins, leucoanthocyanidins	Polyvitamin, choleretic, adaptogenic, general tonic	Forest-steppe zones of Northern and Eastern Kazakhstan (Irtysh and Ishim river areas)
R. acicularis	Vitamins C, B_2, P,	Anti-	Zhetysu, Ili,

Lindl. – bristly rosehip (fruits)	flavonoids, tannins, catechins, essential and fixed oils	inflammatory, antiseptic, antibacterial, vasoconstrictive, hemostatic	Kyrgyz Alatau, Western Tarbagatai. Cultivated in botanical gardens (Zhezkazgan, Karaganda)
R. beggeriana Schrenk – Begger's rosehip (fruits)	Vitamins C, E, P, B_2, flavonoids, catechins, tannins, fixed oils	Astringent, polyvitamin, choleretic, general tonic	Southern and South-Eastern Kazakhstan (Tian Shan, Karzhantau). High drought tolerance
R. fedtschenkoana Regel – Fedchenko's rosehip (fruits)	Vitamins C, P, E, tannins, flavonoids	Choleretic, antiseptic, diuretic, antibacterial, general tonic	Mountain zones of Southern Kazakhstan. Cultivated at the Main Botanical Garden
R. laxa Retz. – loose rosehip (fruits)	Vitamins C and P, carotenoids, flavonoids	Vitamin preparation used in anemia, asthenia, hypoacid gastritis, liver diseases, and nephritis	Central and Eastern Kazakhstan (Altai, Tarbagatai, Zhetysu Alatau, Saryarka regions)

1.3 General Information on Dog Rosehip

The name "dog rosehip" (*Rosa canina*) is associated by the Roman encyclopedist Pliny the Elder with the medicinal use of the plant's roots in treating rabies caused by dog bites [65]. According to data from the Oxford Dictionary, the name derives from the ancient Greek word "κυνόροδον" (κύων – dog, ῥόδον – rose) [66]. In the 18th–19th centuries, this name was also justified within the framework of the doctrine of signatures by the resemblance of the prickles to a dog's teeth [67]. In some sources, the name "ternovnik" has been applied to dog rosehip. This word derives from the Proto-Slavic root *tьrnъ* (thorn), which traces back to the Proto-Indo-European roots *(s)ter-n-* ("a plant with a thorny stem") and *ster-* ("to be hard," "to harden") [68].

The oldest dog rosehip bush in the world grows on the grounds of Hildesheim Cathedral in Germany. According to various estimates, the age of this "millennial rose" ranges from 400 to 1,000 years. The plant reaches a height of 13 metres, and the circumference of its trunk at the base measures 50 cm [69]. This fact attests to the high vitality and biological potential of the species *Rosa canina*.

Dog rosehip is a deciduous shrub reaching a height of 1.5 to 2.5 metres. The shoots are thick, arcuately curved, and rarely erect. The bark is green in color, but acquires a pinkish hue on the side directly exposed to sunlight.

The prickles are sparsely arranged, with a short base, laterally compressed, and curved in a sickle-like manner. On the main stems the prickles are predominantly erect, whereas on flowering shoots they are more abundant and always curved in a hook-like manner.

The average length of the mid-stem leaves on flowering shoots is 7–9 cm. The leaves are imparipinnate compound, consisting mainly of seven, and sometimes five or nine leaflets. The leaflets are ovate-elliptical in shape, 2–2.5 cm in length and 1–1.5 cm in width. The leaflet margins are simply serrate, with a shortly acuminate apex. At the base of the petiole, narrow stipules with sharply pointed auricles and glandular-ciliate margins are situated.

The flowers are scentless, solitary or gathered in corymb-like inflorescences of 3–5 at the tips of the shoots. The color of the flowers ranges from white to deep pink, with a diameter of 5–8 cm.

- Sepals: 20–25 mm in length, broadly lanceolate, with pinnately lobed appendages. Their principal diagnostic feature is that they bend backward after flowering and fall off early – before the fruit ripens.
- Disc: flat or conical in shape (4–5 mm in diameter).
- Floral formula**:** $K_{(5)}\ C_5\ A_\infty\ G_\infty$ [70].

The flowering period extends from late May to June (and sometimes to July). The pedicels are 12–18 mm in length and are typically glabrous and eglandular [71-74].

The fruits are cynarhodia, smooth and glossy on the surface, ranging in color from orange-red to red. The length of a ripe fruit is 15–26 mm, and the shape is broadly ovoid or globose. Inside the fruit, numerous hairy, hard achenes are located. The fruits ripen fully in August [75].

The root system consists of rhizomes and lateral roots. The rhizomes are nodose, branched, and reach a length of 20–25 cm. The roots are cylindrical in shape and curved. The outer bark (periderm) is dark brown, reddish-brown, or grayish-brown in color. The surface is deeply longitudinally furrowed and sometimes exfoliates easily. The inner portion of the root (wood) is dense, hard, and yellowish-white or pale brown in color. The fracture is rough, splintery, and fibrous-woody. The odor is faint and characteristic; the taste is astringent, due to the high content of tannins [76].

1.4 Use of Dog Rosehip in Folk Medicine

The medicinal properties of dog rosehip have been known since ancient times. Abu Ali ibn Sina (Avicenna) used rosehip oil for the treatment of gum inflammation and ocular inflammatory diseases [77]. In medieval Europe, an infusion of rosehip petals in wine was used for gastrointestinal disorders and gynecological pathologies. Furthermore, a mixture of rosehip fruits with honey

was used as an antipyretic agent, and a mixture with vinegar was used for washing purulent wounds [78].

Historical sources attest that during the Russo-Turkish War, infusions of rosehip fruits and petals were used for saturating wound dressings and washing wounds, as well as for internal use in cases of general weakness and malaria [79]. Fruits have long been regarded as the principal remedy for the treatment of hypo- and avitaminosis C.

Applications of fruits in folk medicine:

- Cardiovascular system: used as a primary agent in the prevention and treatment of anemia, atherosclerosis, thrombosis, hypertension, and ischemic diseases.
- Hemostatic effect: demonstrates high efficacy in nasal, pulmonary, and uterine hemorrhage, as well as in hemorrhagic diathesis and hemophilia.
- Infectious and viral diseases: enhances the body's resistance in acute respiratory viral infections, influenza, and conditions of general weakness associated with vitamin C deficiency (hypo- and avitaminosis).
- Metabolic and systemic disorders: used in digestive diseases (hepatitis, cholecystitis), urogenital disorders (cystitis, nephritis), diabetes mellitus, and detoxification of the body [80-84].

The use of dog rosehip roots (*Rosa canina* L.) is extensively described in ethnomedicinal sources of Eastern Europe, Central Asia, the Caucasus, and Siberia [85]. According to practitioners of traditional medicine, collection of root raw material in autumn, after the end of the vegetation period, ensures maximum accumulation of biologically active substances. In folk practice, roots are used primarily in the form of decoctions and infusions.

Principal applications of root decoctions:

- Urinary system: normalizes metabolism, exhibits pronounced choleretic, diuretic, and bactericidal activity. Possesses the capacity to expel sand and small stones (calculi) from the urinary tract.
- Gastrointestinal tract: used as an astringent agent in diarrhea and spastic conditions of the intestine, owing to the high content of tannins.
- Musculoskeletal system: used for its anti-inflammatory and metabolic-regulating effects in rheumatism and gout [76, 86].

1.5 Chemical Composition of Dog Rosehip

The chemical composition of freshly harvested rosehip fruits is characterized by the following indicators: moisture content ranges from 71.93 to 82.14%, and the fruit pulp from 54.50 to 54.76%. The total sugar content of the fruit pulp amounts to 23.93–23.97%, including invert sugars (18.56–18.64%) and sucrose (5.09–5.14%). Total acidity (expressed in terms of malic acid) is 2.84–2.86%, ascorbic acid (vitamin C) 3.79–3.84%, and citric acid 1.58–1.64%. Additionally, the raw material contains pectic substances (14.10–14.65%), crude fiber (12.52–12.67%), and crude ash (6.43–6.45%) [87-90].

On an absolutely dry raw material basis, the ascorbic acid content reaches up to 1,352 mg, and carotenoids up to 0.07%. The fruits are rich in flavonoids, catechins, anthocyanin compounds, vitamins B, K, and P, pectins, and organic acids (malic, citric). The micro- and macroelement composition is represented by sodium, potassium, magnesium, iron, and phosphorus. The achenes (seeds) of rosehip contain a fixed oil consisting of saturated and unsaturated fatty acids (linoleic, linolenic, palmitic, oleic, and others). Na, K, Ca, Mg, Cu, Mn, and Zn elements have also been found in the seeds. The flavonoid isoquercitrin and its derivatives have been isolated from the fruits. The mass fraction of moisture in dried fruits is approximately 12%. The characteristic sour taste of the fruits is attributable to the presence of malic and citric acids [91-94].

The roots of *Rosa canina* L. accumulate organic acids, polysaccharides, hydrolyzable tannins, saponins, amino acids, and phenolic compounds. According to data from recent studies, ascorbic acid accounts for 0.26±0.004%. The total content of organic acids is 8.06±0.047%, among which tartaric, citric, malic, oxalic, and succinic acids have been identified [95].

The content of water-soluble polysaccharides was determined at 5.29%, pectic substances at 8.1%, and hemicellulose at 2.8%. The content of tannins and readily oxidizable substances amounts to 10.77±0.03%. The content of triterpenoid saponins is 4.66±0.02%. A phytochemical feature of the raw material is the first-time isolation of specific compounds of the 19α-hydroxyursane type: euscaphic, tormentic, and 2-oxopomolic acids, along with their glycosides – rosamultin and kaji-ichigoside F1 [96].

Investigation of the amino acid composition identified 15 amino acids (aspartic acid, threonine, serine, glutamic acid, glycine, alanine, valine, methionine, isoleucine, leucine, tyrosine, phenylalanine, histidine, lysine, arginine), seven of which are essential amino acids [97, 98].

Investigation of the elemental composition revealed 30 macro- and microelements, including calcium, magnesium, iron, silicon, zinc, manganese, and others. Among conditionally toxic elements, silver (0.00005 mg% per ash), tin (0.0006 mg%), zirconium (0.002 mg%), and gallium (0.001 mg%) were detected.

Three flavonoids were identified in the rhizomes: two glycosides (rutin and quercetin-3-D-glucoside) and one aglycone (apigenin). A complex of phenolic compounds was found in root extracts, including procyanidin B1, epicatechin, chlorogenic acid, procyanidin B2, salicylic acid, gallic acid, and catechin. Their total content amounts to 12.73% [99, 100]. Six compounds of catechin nature were identified in the raw material of *Rosa canina* L., with a total content of 4.89%. This species is distinguished by the accumulation of the maximum concentration of epigallocatechin among all catechins studied (approximately 3% of the total catechin content) [101, 102].

According to recent data, root extracts have been found to contain epicatechin, gallic, syringic, *p*-coumaric, and ellagic acids – compounds not detected in the fruit [103]. Moreover, the presence of 19α-hydroxyursane-type triterpenoids, including euscaphic acid and tormentic acid, has been confirmed [104].

Based on a systematization of data from the scientific literature, the distribution of groups of biologically active substances among the various organs of dog rosehip is summarized in Table 4.

Table 4. Distribution of groups of biologically active substances by organ in dog rosehip

BAS groups	**Fruits**	**Seeds / achenes**	**Leaves**	**Flowers / petals**	**Roots / rhizomes**
Vitamin C (ascorbic acid)	+	–	–	–	+
Flavonoids	+	–	+	+	+
Catechins (flavan-3-ols)	+	–	–	–	+
Phenolic acids	+	–	+	–	+
Tannins	+	–	+	+	+
Carotenoids	+	–	–	–	–
Pectins / polysaccharides	+	–	–	–	+
Organic acids	+	–	–	–	+
Triterpenoid saponins	–	–	–	–	+
Tocopherols	–	+	–	–	–

1.6 Pharmacological Activity and Medical Applications of Dog Rosehip

The therapeutic efficacy of dog rosehip is determined by the complex of biologically active substances (BAS) it contains. Isoflavonoids and kaempferol exhibit diuretic activity; rutin and compounds with vitamin P activity demonstrate angioprotective and hemostatic effects. Tannins present in the fruits and other morphological parts of the plant possess bactericidal, anti-inflammatory, and astringent properties.

Since the content of ascorbic acid in the fruits of dog rosehip is comparatively low (0.24-0.85%), this species belongs to the "low-vitamin" category. The principal pharmacological value of the raw material lies in its choleretic and metabolism-regulating effects. These properties form the basis for the production of the preparations "Choloshas" and "Polyphytochole," used in the treatment of cholecystitis, hepatitis, and biliary tract diseases. Furthermore, rosehip oil (*Oleum Rosae*), owing to the fatty acids and carotenoids it contains, is widely used in dermatological and gastroenterological practice as a regenerating and reparative agent. In the segment of biologically active food supplements, rosehip fruits constitute an important component. Rosehip included in multi-component herbal teas and complex preparations (for example, "Hepatocrit") produces a synergistic effect in combination with other medicinal plants (chamomile, peppermint, licorice), thereby enhancing the therapeutic outcome [105-109].

The characteristics and scope of application of the principal medicinal preparations and biologically active food supplements authorized in official medicine are systematized in Table 5.

Table 5. Characteristics of medicinal preparations and biologically active food supplements based on dog rosehip

Name	**Composition**	**Dosage form**	**Indications for use**
Medicinal preparations			
Rosehip fruits	Rosehip fruits (whole and powdered)	Packets, paper bags, polypropylene bags, paper pouches	Polyvitamin preparation of plant origin
Rosehip fruit syrup	Rosehip fruits (aqueous extract), sugar, ascorbic acid, citric acid	Jars, dark glass vials, bottles, containers	Polyvitamin preparation of plant origin
Rosehip fruit syrup "Astpharm"	Rosehip fruits (dry extract, infusion), ascorbic acid, sugar, citric acid	Vials	Enhancement of nonspecific resistance of the body, stimulation of tissue regeneration, reduction of vascular permeability, normalization of carbohydrate metabolism, choleretic effect
Rosehip fruit oil	Rosehip fruits (oil)	Vials	Plant-origin stimulator for internal use and for topical and external application for tissue repair
"Polyphytochole"	Sandy everlasting flowers, dioecious nettle leaves, chamomile flowers, licorice root, rosehip fruits (dry extract)	Powder for preparation of solution for internal use	Choleretic, moderate anti-inflammatory, and spasmolytic effect; stimulation of liver and pancreatic function and biliary motility; moderate diuretic effect
"Choloshas"	Rosehip fruits (liquid extract), sucrose	Vials	Choleretic preparation of plant origin

Biologically active food supplements			
"Hepatocrit" with chamomile	Coriander fruits, bur marigold herb, rosehip fruits, calamus rhizomes and roots, peppermint leaves, yarrow herb, chamomile flowers	Filter bags	Choleretic preparation

Experimental and clinical studies have demonstrated the antioxidant, anti-inflammatory, antidiabetic, and antitumor activities of rosehip fruits.

The hepatoprotective activity of the fruits was demonstrated in experimental models on rats by the capacity to effectively protect liver cells from carbon tetrachloride (CCl_4) intoxication and to reduce the level of hepatotoxicity [110]. The results of *in vitro* studies showed that a galactolipid present in rosehip is capable of inhibiting the chemotaxis of neutrophils in human peripheral blood, thereby reducing the accumulation of cells at the site of inflammation. Furthermore, it has been demonstrated that the fruits of *Rosa canina* L. inhibit cyclooxygenase-1 (COX-1) and cyclooxygenase-2 (COX-2), the principal mediator enzymes of inflammatory processes [111, 112].

Clinical studies have demonstrated that a three-month course of aqueous rosehip extract administration in patients with type 2 diabetes mellitus significantly reduced blood glucose levels and serum total cholesterol/HDL-C ratios without adverse effects [113]. Fruit powder has been shown to enhance cell viability and improve skin elasticity through stimulation of collagen synthesis, while exhibiting *in vitro* antiproliferative activity against breast cancer (MCF-7), cervical cancer (HeLa), and colorectal cancer (HT-29) cell lines [49, 114]. Furthermore, the methanolic extract inhibits the conjugation of bacterial plasmids, which represents an important combinatorial factor in the treatment of antibiotic-resistant strains [54].

Owing to the high content of organic acids (8.06%) in dog rosehip roots, root extracts demonstrate superior bacteriostatic activity compared to other species of the genus *Rosa* (*R. pisiformis*, *R. villosa*) [115].

The antioxidant activity of aqueous-methanolic root extracts, confirmed by the CUPRAC method (191.86 ± 17.95 mg TEAC/g), is approximately three times higher than that recorded for fruit extracts (64.93 ± 12.41 mg TEAC/g). The bioaccessibility of root phenolic compounds was assessed using an *in vitro* gastrointestinal digestion model, confirming the substantial release of gallic and ellagic acids under simulated digestion conditions [103]. HPLC-UV-MS analysis identified the presence of 19α-hydroxyursane-type triterpenoids – specifically euscaphic acid and tormentic acid – in the roots, and demonstrated their *in vitro* and *in silico* inhibitory activity against collagenase and tyrosinase, thereby suggesting a protective role against photoaging and hyperpigmentation of the skin [104].

Data on the chemical composition and pharmacological activity of *Rosa canina* L. roots remain fragmentary in character and are based primarily on empirical practice. The scarcity of systematic pharmacognostic studies limits the use of this valuable raw material in official medicine. In this regard, the investigation of the phytochemical profile of dog rosehip roots growing in Katon-Karagay National Park represents a relevant and promising direction for domestic pharmacy.

CHAPTER 2. MATERIALS AND METHODS

2.1 Characterization of the Research Objects

The principal object of study was the roots of dog rosehip (*Rosa canina* L.) growing in the territory of Katon-Karagay National Park, East Kazakhstan Region: approximate geographic coordinates: 49°10′N, 85°37′E; altitude ~1,200–1,400 m a.s.l. (Figure 1). The preparation of medicinal plant raw material (MPRM) was carried out in strict compliance with the requirements of the State Pharmacopoeia of the Republic of Kazakhstan (SP RK) and the international standards of Good Agricultural and Collection Practice (GACP).

Figure 1. Dog rosehip (*Rosa canina* L.) in its natural habitat (Katon-Karagay National Park, 2025)

Collection of raw material. Collection was carried out under permit No. 27-2-20/5190-FWC issued by the administration of Katon-Karagay National Park (09.06.2023). The roots of *Rosa canina* L. were harvested in the autumn of 2025 (at the end of the growing season), taking into account the biological rhythm of the plant and the period of maximum accumulation of biologically active substances (BAS). To preserve the ecological balance of the population, in accordance with GACP guidelines, the root system of any individual shrub was not excavated in its entirety; only lateral, well-developed root branches were excised. Harvesting was conducted under dry, clear weather conditions.

Primary processing. Excavated roots were subjected to immediate primary processing. The raw material was shaken free of soil, stones, and sand, and rapidly rinsed with running cold water to prevent mineral contamination (ash insoluble in 10% HCl). Dead, decayed, mold-affected, or pest-damaged portions, as well as fine fibrous rootlets (diameter < 0.5 cm), were removed. Thick roots were sectioned into longitudinal and transverse pieces of 10–15 cm in length to facilitate the drying process.

Drying. The raw material was spread in a thin layer (3–5 cm) on clean paper sheets under a covered shelter in the shade, protected from direct sunlight, with adequate ventilation. To ensure uniform drying and prevent microbiological contamination, the raw material was turned 1–2 times daily. Completion of drying was assessed by the organoleptic (pharmacognostic) method: roots were considered sufficiently dried when they snapped without bending (fibrous fracture). The residual moisture content at this stage was reduced to no more than 10%.

Packaging and storage. The dried root raw material was packaged and stored in accordance with the requirements of the general article "Storage of Medicinal Plant Raw Material" of Volume 1 of the SP RK and GOST 6077-80. The raw material was placed in multi-layer kraft paper bags and natural fabric sacks to ensure air permeability and prevent the accumulation of hygroscopic moisture. A label indicating the collection site (Katon-Karagay National Park), date, and name of the raw material was affixed to each package. The packaged raw material was stored in a dedicated room – dry, light-protected, and well-ventilated – with relative air humidity not exceeding 60% and ambient temperature not exceeding +25°C.

Laboratory instruments and equipment:

- UV-Vis spectrophotometer Cary-60 (Agilent Technologies, USA): for measurement of optical density during the quantitative determination of polyphenols and flavonoids;
- Light microscope OPTIKA (Italy): equipped with a high-resolution digital camera, for recording anatomical features;
- Analytical balance: weighing accuracy of 0.0001 g;
- Electric muffle furnace: heating range 500–600°C, for ash content determination;
- Electric drying oven: temperature 105 ± 2°C, for determination of moisture content and extractable substances;
- Electric water bath: for maintaining constant temperature during BAS extraction;
- Auxiliary equipment: desiccator, 25/50 ml microburettes, reflux condensers, porcelain crucibles and evaporating dishes, weighing bottles.

2.2 Methods of Investigation

Morphological-anatomical analysis

Macroscopic and microscopic analyses were carried out in accordance with the requirements of the general pharmacopoeial article "Roots, Rhizomes, Bulbs, Tubers, and Corms" of Volume 1 of the SP RK, for the purpose of determining the authenticity and diagnostic features of the plant raw material.

Macroscopic analysis. The external characteristics of the roots (shape, surface texture, color, fracture, odor, and taste) were described visually and with the aid of optical magnification instruments.

Microscopic analysis. For the preparation of transverse sections, samples were softened in a water–glycerin–alcohol (1:1:1) mixture. A 5% aqueous sodium hydroxide (NaOH) solution was used for tissue clearing, and Lugol's solution for the identification of starch grains. Preparations were mounted in glycerin and examined under a light microscope (bright-field method) equipped with an OPTIKA digital imaging system.

Evaluation of qualitative indicators of raw material

Moisture content was determined according to SP RK method 2.2.32. A precisely weighed 2.0 g sample of raw material was placed in a pre-tared weighing bottle and dried in an electric oven at 105°C to constant weight (initial drying: 2 hours; subsequent intervals: 1 hour each). Cooling was performed in a desiccator for 30 minutes. Weighings were performed on an analytical balance with an accuracy of 0.0001 g. Moisture content (W, %) was calculated by the following formula:

$$W = \frac{m_2 - m_3}{m_2 - m_1}$$

Where:

- m_1 – mass of the empty weighing bottle, g;
- m_2 – mass of the weighing bottle with raw material before drying, g;
- m_3 – mass of the weighing bottle with raw material after drying to constant weight, g.

Total ash was determined according to SP RK method 2.4.16. A 1.0–2.0 g sample was incinerated without flame in a porcelain crucible and heated in a muffle furnace at 500–600°C until a uniform pale gray ash was obtained. Total ash content (X, %), expressed on an absolutely dry basis, was calculated as:

$$X = = \frac{(m_2 - m_1) \times 100 \times 100}{m \times (100 - W)}$$

Where:

- m_1 – mass of the empty crucible, g;
- m_2 – mass of the crucible with ash, g;
- m – initial mass of raw material taken for analysis, g;
- W – moisture content, %.

Ash insoluble in 10% hydrochloric acid was determined in accordance with SP RK article 2.8.1. To the total ash, 15 ml of a 10% HCl solution was added and the mixture was boiled in a water bath for 10 minutes. The mixture was filtered through an ash-free filter and the residue was washed with hot water until a negative reaction to silver nitrate was obtained. The filter was re-incinerated in the crucible at 500–600°C and weighed. The acid-insoluble ash content (X_1, %) on an absolutely dry basis was calculated as:

$$X_1 = = \frac{(m_2 - m_1) \times 100 \times 100}{m \times (100 - W)}$$

Where:

- m_1 – mass of the empty porcelain crucible, g;
- m_2 – constant mass of the crucible with the acid-insoluble residue after re-incineration, g;
- m – initial precisely weighed mass of raw material, g;
- W – loss on drying (moisture content), %.

Extractable substances were determined gravimetrically in accordance with GOST R ISO 9768-2011. A 2.0 g sample was extracted with 50 ml (V_1) of purified water under a reflux condenser in a water bath for 1 hour. From the filtered extract, a 25 ml aliquot (V_2) was evaporated in a pre-weighed porcelain dish and dried at 105°C to constant weight. The extractable substance content (X_2, %) on an absolutely dry basis was calculated as:

$$X_2 = = \frac{(m_1 - m_0) \times V1 \times 100 \times 100}{m \times V2 \times (100 - W)}$$

Where:

- m_0 – mass of the empty porcelain dish, g;
- m_1 – constant mass of the dish with dry residue, g;
- m – initial mass of raw material, g;
- V_1 – total volume of extractant (50 ml);
- V_2 – volume of filtrate taken for evaporation (25 ml);
- W – loss on drying (moisture content), %.

Phytochemical analysis

Preparation of extracts:

- Aqueous extract: 5.0 g of raw material was extracted with 50 ml of distilled water (1:10) in a water bath for 30 minutes;
- Alcoholic extract: 5.0 g of raw material was refluxed with 50 ml of 70% ethyl alcohol (1:10) for 45–60 minutes and filtered.

Qualitative analysis (color reactions):

- Phenolic compounds: 2 ml extract + 3 drops of 1% $FeCl_3$;
- Tannins: 2 ml aqueous extract + 1% gelatin solution (in the presence of 10% NaCl) or bromine water;
- Catechins: 1 ml alcoholic extract + 1% vanillin in alcohol + concentrated HCl;
- Flavonoids: 2 ml extract + 1 ml 2% $AlCl_3$ (fluorescence) and cyanidine test (Mg powder + concentrated HCl);
- Triterpenoids (Liebermann–Burchard): dry residue + acetic anhydride + concentrated H_2SO_4;
- Saponins: vigorous shaking of 5 ml aqueous extract for 15 seconds (foam test);

- Coumarins: 2 ml extract + 10% NaOH, heating, observation under UV light;
- Anthraquinones (Bornträger): hydrolyzed extract + NaOH solution;
- Alkaloids: acidified filtrate + Dragendorff's reagent;
- Amino acids: 2 ml extract + 1% ninhydrin solution, heating;
- Carbohydrates: Molisch reaction (α-naphthol + H_2SO_4);
- Organic acids: aqueous extract + $NaHCO_3$ powder.

Quantitative analysis was performed by spectrophotometry and titrimetry using a Cary-60 UV-Vis spectrophotometer (all measurements in triplicate, n=3). Results were expressed as a percentage (%) on an absolutely dry raw material basis.

Polyphenols: determined using the Folin–Ciocalteu reagent (GOST R ISO 14502-1-2010) at λ = 765 nm, expressed relative to a gallic acid reference standard.

Flavonoids: determined via $AlCl_3$ complexation at λ = 415 nm, expressed relative to a rutin reference standard.

The quantitative content (X_3) of both analytes was calculated using the following spectrophotometric formula:

$$X_3 = \frac{A \times m_0 \times V_1 \times 100}{A_0 \times m \times V_2 (100 - W)}$$

Where:

- A – optical density of the test extract;
- A_0 – optical density of the reference standard solution (gallic acid or rutin);
- m_0 – mass of the reference standard, g;
- m – mass of the test raw material, g;
- V_1 – total volume of the extract, ml;
- V_2 – volume of the aliquot taken for analysis, ml;
- W – moisture content (loss on drying), %.

Tannins were determined by permanganatometric titrimetry in accordance with GOST 24027.2-80, using indigo sulfonic acid as indicator and titrating with a 0.02 mol/L potassium permanganate ($KMnO_4$) solution to a stable golden-yellow endpoint. Total tannin content (X_4) was calculated as:

$$X_4 = \frac{(V - V_0) \times K \times 0.004157 \times V_1 \times 100}{m \times V_a \times (100 - W)}$$

Where:

- V – volume of $KMnO_4$ consumed for the test solution, ml;
- V_0 – volume of $KMnO_4$ consumed for the blank, ml;
- 0.004157 – mass of tannins (g) equivalent to 1 ml of 0.02 mol/L $KMnO_4$;

- K – correction factor for $KMnO_4$;
- V_1 – total extract volume (250 ml);
- V_a – aliquot volume taken for titration (25 ml);
- m – mass of raw material, g;
- W – moisture content, %.

Ascorbic acid (Vitamin C) was determined by iodometric titration with a standard iodine solution in the presence of a starch indicator, titrating to a stable pale blue endpoint persisting for ≥ 1 minute. Vitamin C content (X_5) was calculated as:

$$X_5 = \frac{V \times T \times V_1 \times 100}{m \times V_2 \times (100 - W)}$$

Where:

- V – volume of iodine solution consumed, ml;
- T – titer of iodine solution in terms of ascorbic acid, mg/ml;
- V1 – total volume of the acid extract, ml;
- V2 – aliquot volume taken for titration, ml;
- m – mass of raw material, g;
- W – moisture content, %.

Comparative analysis of the chemical composition of roots and fruits

A comprehensive phytochemical analysis was conducted to comparatively evaluate the accumulation patterns of biologically active substances in the roots and fruits of the dog rose (Rosa canina L.) growing in the Katon-Karagay National Park. For the study, fruits (Figure 2) were collected in addition to the roots of Rosa canina L. from the same geographical point – Katon-Karagay National Park – during the autumn of 2025. Both types of raw materials were prepared in accordance with GACP (Good Agricultural and Collection Practices) standards and processed under identical laboratory conditions.

Figure 2. External morphology and transverse sections of dog rosehip fruits (Katon-Karagay National Park)

The comparative analysis encompassed five parameters: total polyphenols, total flavonoids, tannins, ascorbic acid. All determinations were performed on the fruit raw material under conditions identical to those applied for the roots – using

the same instrumental platform, identical calibration curves, and three parallel measurements (n=3) for each parameter. Results were expressed as a percentage (%) on an absolutely dry raw material basis.

CHAPTER 3. RESULTS

3.1 Morphological-Anatomical Analysis of Dog Rosehip Root Raw Material

For the purpose of confirming the authenticity and evaluating the quality of the medicinal plant raw material (MPRM), a comprehensive macro- and microscopic analysis was performed on samples collected from Katon-Karagay National Park.

Macroscopic evaluation was carried out in accordance with the general methodological guidelines of pharmacognostic analysis. The study was conducted under diffuse daylight conditions. The length and diameter of the roots were determined using millimeter-scale measuring instruments, and surface features were examined with the aid of a 10× magnifying loupe. Organoleptic indicators (odor and taste) were assessed in accordance with standard requirements.

Macroscopic diagnostic features identified in the course of the study (Figures 3 and 4):

- Shape and dimensions: the raw material consists of cylindrical, lignified, and branched root fragments with a length of 20–30 cm and a diameter of 0.5–1.5 cm.
- Surface texture and color: the outer surface of the root is rough and longitudinally furrowed. The outer protective tissue (periderm) exfoliates in some areas, resulting in the exposure of lighter-colored underlying tissues.
- Color: the external color ranges from yellowish-brown to dark brown.
- Bark layer: the thickness of the root bark ranges from 0.02 to 0.10 cm; it is very thin and closely adherent to the central wood.
- Fracture: fibrous and woody, indicating well-developed secondary xylem in mature root raw material.
- Organoleptic characteristics: the odor is faint and characteristic. The taste is slightly astringent, attributable to the presence of phenolic compounds and tannins.

Figure 3. External morphology of dog rosehip roots collected from Katon-Karagay National Park

Figure 4. Macroscopic view of the surface and fracture of a dog rosehip root

For microscopic analysis, transverse sections of the roots were prepared. Samples were pre-softened in a water–glycerin–alcohol (1:1:1) mixture and treated with an aqueous sodium hydroxide (NaOH) solution for tissue clearing. Cleared preparations were mounted in glycerin and examined under a light microscope (bright-field method) equipped with an OPTIKA digital imaging system. Lugol's solution (iodine test) was used for the identification of starch grains.

Microscopic examination of transverse root sections revealed a stable anatomical diagnostic complex characteristic of *Rosa canina* L. (Figures 5 and 6):

1. Integumentary and cortical tissues: a multi-layered periderm (cork tissue) is situated at the periphery of the root. Beneath it, cortical parenchyma composed of thin-walled polygonal cells is observed.
2. Conducting system (xylem and phloem): clearly visible medullary rays were identified in the phloem zone. The central portion of the root is occupied by secondary xylem (wood), in which lignified conducting vessels were identified. The vessel walls bear scalariform and bordered pits arranged in longitudinal rows. Thick-walled wood fibers and dense xylem fragments were also recorded.
3. Storage substances: starch grains of varying sizes and rounded or oval shape are accumulated in parenchyma cells. Upon application of Lugol's solution, a characteristic blue-violet coloration was observed.
4. Crystal inclusions: druses of calcium oxalate (star-shaped crystal aggregates) were identified in the parenchyma.

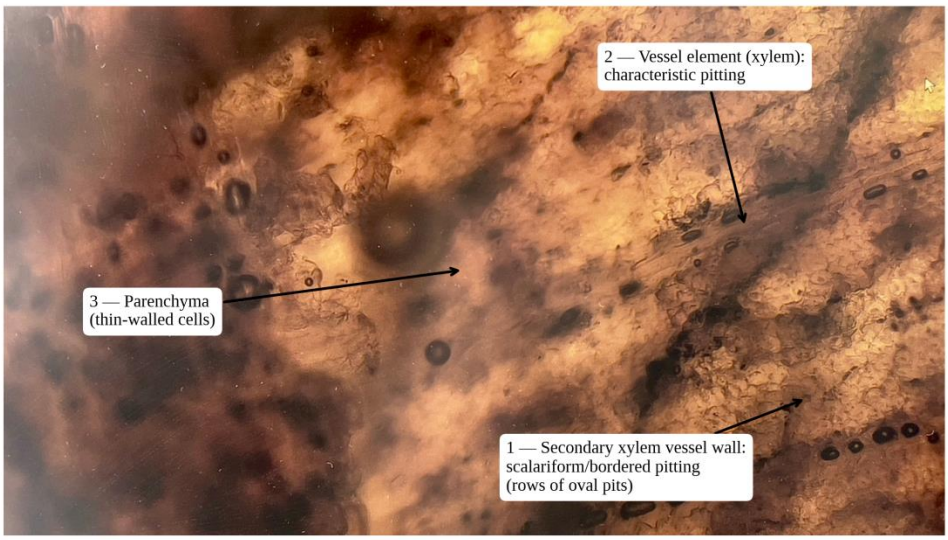

Figure 5. Root of dog rosehip; bright-field microscopy. *Labels: 1, 2 – conducting vessel elements of secondary xylem with characteristic pits; 3 – parenchymatous tissue*

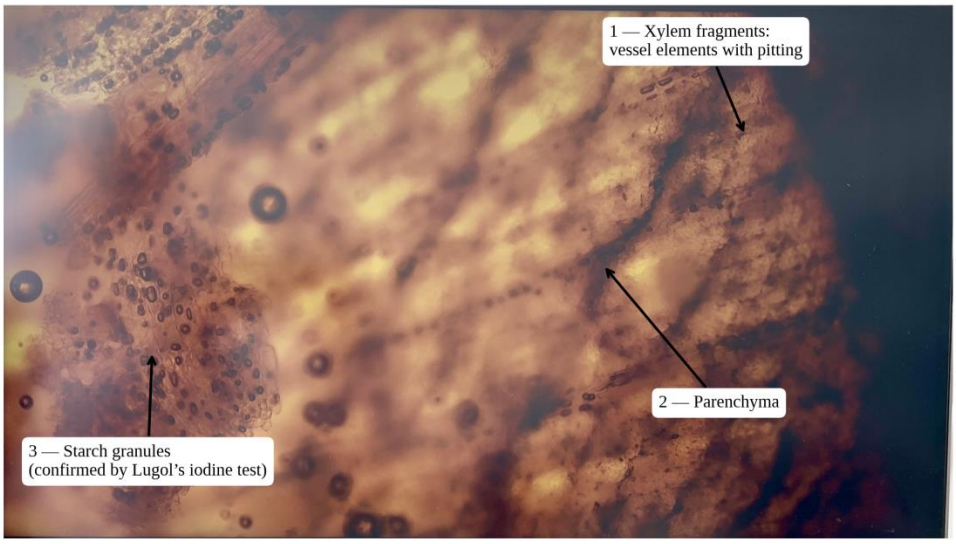

Figure 6. Root of dog rosehip; bright-field microscopy. *Labels: 1 – xylem fragments; 2 – parenchyma; 3 – starch grains (confirmed by Lugol's reaction)*

3.2 Evaluation of Qualitative Indicators of Dog Rosehip Root Raw Material

For the purpose of standardization and quality control of the MPRM, the principal qualitative indicators – moisture content, total ash, ash insoluble in 10% hydrochloric acid, and extractable substances – were determined in accordance with the requirements of the SP RK.

Moisture content is a critical parameter governing the stability of the raw material during storage and its resistance to microbiological deterioration.

Instruments and equipment: analytical balance (accuracy 0.0001 g), electric drying oven, glass weighing bottles, desiccator with desiccant.

Procedure: clean, dry glass weighing bottles were dried in an electric oven at 105°C to constant weight, cooled in a desiccator for 30 minutes, and weighed (m_1). Approximately 2.0 g of root raw material comminuted to a particle size of 1–3 mm was precisely weighed and placed into the prepared weighing bottle. The weighing bottle with the raw material was weighed (m_2). The weighing bottle containing the raw material (with the lid open) was placed in the drying oven and dried at 105°C for 2 hours. After the initial drying period, the bottle was cooled in a desiccator for 30 minutes and reweighed. Subsequent drying was carried out at 1-hour intervals. Constant weight (m_3) was considered achieved when the difference between two consecutive weighings did not exceed 0.0005 g. The results are systematized in Table 6.

Table 6. Determination of moisture content of dog rosehip root raw material

No.	**Initial mass of raw material (*m*), g**	**Mass of weighing bottle with raw material before drying (m_2), g**	**Mass after 2 h, g**	**Mass after 30 min, g**	**Constant mass after 30 min (m_3), g**	**Moisture content (*W*), %**
1	2.0041	48.2164	48.0712	48.0688	48.0675	7.43
2	2.0023	50.6477	50.5010	50.4998	50.4991	7.42
3	2.0038	44.2626	44.1165	44.1152	44.1144	7.39

Moisture content was calculated using the formula (W) presented in Section 2.2. Based on the results of three samples, the mean moisture content of the raw material was 7.41 ± 0.02%, which fully complies with the SP RK requirement (moisture content must not exceed 10%).

Total ash is the inorganic (mineral) residue remaining after complete ignition of the medicinal plant raw material in a muffle furnace at 500–600°C.

Instruments and equipment: muffle furnace, porcelain crucibles, desiccator with desiccant.

Procedure: porcelain crucibles were heated in a muffle furnace at 500–600°C to constant weight, cooled in a desiccator, and weighed (m_1). Approximately 1.0–2.0 g of comminuted raw material (precisely weighed) was placed in the crucible and carefully incinerated without flame on an electric hot plate. The incinerated sample was then heated in a muffle furnace at 500–600°C until all carbonaceous particles were completely eliminated and a uniform pale gray ash was formed. The crucible was cooled in a desiccator and brought to constant weight (m_2). Results are systematized in Table 7.

Table 7. Determination of total ash of dog rosehip root raw material

No.	Constant mass of crucible (m_1), g	Initial mass of raw material (m), g	Mass after 2 h, g	Mass after 30 min, g	Constant mass (m_2), g	Net ash mass, g	Total ash, %
1	31.3526	2.0041	31.4215	31.4198	31.4195	0.0669	3.61
2	35.2258	2.0023	35.2948	35.2935	35.2932	0.0674	3.64
3	28.5894	2.0038	28.6582	28.6571	28.6568	0.0674	3.64

Total ash was calculated using the formula (X) presented in Section 2.2, with the moisture content of the raw material (W = 7.4%) taken into account; results were expressed on an absolutely dry basis. The mean total ash content was 3.63 ± 0.02%, which fully complies with the SP RK requirement (total ash must not exceed 7.0%).

Ash insoluble in 10% hydrochloric acid is a specific parameter indicating the content of siliceous matter (primarily silicon dioxide, SiO_2) that remains undissolved after treatment of the total ash with 10% HCl.

Instruments and equipment: water bath, ash-free filter paper, porcelain crucible, muffle furnace.

Procedure: to the total ash obtained in the same crucible, 15 ml of a 10% HCl solution was added. The crucible was covered with a watch glass and heated in a boiling water bath for 10 minutes. The mixture was filtered through ash-free filter paper and the residue was washed with hot distilled water until a negative reaction to silver nitrate (absence of chloride ions) was obtained. The filter paper with the residue was returned to the crucible, dried, and re-incinerated in the muffle furnace at 500–600°C to constant weight. Results are systematized in Table 8.

Table 8. Determination of ash insoluble in 10% HCl of dog rosehip root raw material

No.	Constant mass of crucible (m_1), g	Initial mass of raw material (m), g	Mass after 2 h, g	Mass after 30 min, g	Constant mass (m_2), g	Acid-insoluble ash, %
1	31.3526	2.0041	31.3765	31.3758	31.3755	1.23

2	35.2258	2.0023	35.2495	35.2490	35.2489	1.25
3	28.5894	2.0038	28.6132	28.6126	28.6124	1.24

The content of ash insoluble in 10% HCl was calculated using the formula (X_1) in Section 2.2, with W = 7.4% applied; results were expressed on an absolutely dry basis. The mean value was 1.24 ± 0.01%, which fully complies with the SP RK requirement (must not exceed 3.0%).

Extractable substances represent the complex of organic and inorganic compounds extractable from medicinal plant raw material by a specified solvent (purified water, ethyl alcohol of various concentrations, etc.) and remaining as a residue after complete evaporation of the extractant.

Instruments and equipment: analytical balance, 200/250 ml conical flask with reflux condenser, water bath, glass pipettes, porcelain evaporating dishes, electric drying oven, desiccator.

Procedure: a clean porcelain dish was dried in the oven at 105°C to constant weight, cooled in a desiccator, and precisely weighed (m_0). Approximately 2.0 g (precisely weighed, m) of root raw material comminuted to 1–2 mm was placed in a conical flask and 50 ml (V_1) of purified water was added. The flask was connected to a reflux condenser and heated in a boiling water bath for 1 hour. After cooling to room temperature, the contents were made up to the original volume (50 ml) with the same solvent and filtered through paper filter into a dry flask. A 25 ml aliquot (V_2) of the clear filtrate was pipetted into the pre-weighed porcelain dish and evaporated to dryness in a water bath. The dish with the dry residue was then dried at 105°C to constant weight, cooled in a desiccator, and reweighed (m_2). Results are systematized in Table 9.

Table 9. Determination of extractable substance yield in dog rosehip roots

No.	**Mass of empty dish (m_0), g**	**Precisely weighed mass of raw material (m), g**	**Extractant volume (V_1), ml**	**Aliquot volume (V_2), ml**	**Mass of dish with dry residue (m_1), g**	**Extractable substances, %**
1	42.1534	2.0032	50	25	42.2597	11.461
2	45.3021	2.0015	50	25	45.4083	11.460
3	38.8912	2.0054	50	25	38.9976	11.459

The extractable substance yield was calculated using the formula (X_2) in Section 2.2, with W = 7.4% applied; results were expressed on an absolutely dry basis. The mean yield of extractable substances was 11.46 ± 0.002%.

3.3 Extraction of Biologically Active Substances from Dog Rosehip Roots

To ensure the analytical accuracy and reproducibility of phytochemical analyses, the extraction of biologically active substances (BAS) from the roots of *Rosa canina* L. was carried out according to a standardized stepwise scheme. Extraction conditions (nature and concentration of the extractant, time, and temperature) were selected in accordance with the physicochemical properties of the target compounds.

Pre-treatment of raw material. Dried *Rosa canina* L. roots collected from Katon-Karagay National Park were comminuted in a laboratory mill. The comminuted material was fractionated through analytical sieves to obtain a homogeneous fraction with a particle size of 1–3 mm. The physicochemical rationale for this particle size is that reduction in particle diameter increases the contact surface area between the extractant and plant cells, thereby enhancing the diffusion rate of BAS. The prepared raw material was stored in a sealed container in a dark, dry place until analysis.

Given the chemical diversity of the BAS to be determined, two extractants were used in parallel:

- Distilled water – as a highly polar extractant;
- 70% ethyl alcohol – as a medium-polarity extractant. The selection of 70% as the alcohol concentration is scientifically justified: this concentration maximizes the yield of phenolic compounds while minimizing protein precipitation compared to pure water, and improves the solubility of polar BAS relative to absolute ethanol.

Preparation of the aqueous extract. The aqueous extract was prepared primarily for qualitative reactions targeting polar compounds (tannins, saponins, polysaccharides, organic acids, amino acids) and for the quantitative determination of ascorbic acid.

Procedure: a precisely weighed 5.0 g sample of comminuted root raw material was placed in a 250 ml conical flask. 50 ml of distilled water at room temperature was added (raw material:extractant ratio 1:10). The flask was covered with a watch glass, placed in a boiling water bath, and extracted continuously for 30 minutes at 95–98°C. This temperature range was selected to maximize BAS yield while limiting the thermal degradation of thermolabile compounds (vitamin C).

After extraction, the hot extract (above 60°C) was filtered through a cotton-gauze filter into a 100 ml volumetric flask – hot filtration prevents tannin precipitation upon cooling. The filtrate was cooled to room temperature, the volume was adjusted to the original level (50 ml) with distilled water, and the extract was used immediately or stored at 4°C for no longer than 24 hours.

Preparation of the alcoholic extract. The alcoholic extract was prepared for qualitative reactions targeting flavonoids, catechins, phenolic compounds, and triterpenoids, as well as for the quantitative determination of polyphenols and flavonoids on the Cary-60 UV-Vis spectrophotometer.

Procedure: a precisely weighed 5.0 g sample of comminuted root raw material was placed in a 250 ml conical flask. 50 ml of 70% ethyl alcohol was added (raw material:extractant ratio 1:10). The flask was equipped with a reflux condenser, which prevents solvent evaporation and maintains a stable concentration throughout extraction. The mixture was heated in a boiling water bath under gentle reflux at 78–80°C for 45–60 minutes. This temperature was selected because it approximates the boiling point of 70% ethanol (78.1°C), ensuring maximum recovery of flavonoids and phenolic compounds while preventing their thermal degradation.

After cooling to room temperature, the extract was filtered through paper filter until clear. The filtrate was divided into two portions:

- First portion – used directly for qualitative phytochemical reactions;
- Second portion – diluted with 70% ethyl alcohol to the required working concentration for optical density measurement on the Cary-60 UV-Vis spectrophotometer.

Quality control of extracts. The quality and suitability of the prepared extracts were evaluated according to the following criteria:

- Clarity: all extracts were re-filtered until optically clear. Turbid extracts were considered analytically unsuitable, as suspended colloidal particles may produce false optical density readings in spectrophotometric measurements;
- Time of use: alcoholic extracts were used on the day of preparation, as oxidative degradation of phenolic compounds occurs during storage;
- Storage conditions: prior to quantitative determinations, extracts were stored at 4°C, protected from light.

3.4 Qualitative Analysis of the Principal Groups of Biologically Active Substances in Dog Rosehip Roots

For the purpose of characterizing the chemical composition of *Rosa canina* L. root raw material, qualitative analysis of the principal BAS groups was performed. The chemical reactions conducted and their results are presented below.

1. Total phenolic compounds ($FeCl_3$ test)

3 drops of a 1% iron(III) chloride solution were added to 2 ml of alcoholic or aqueous extract.

Result: positive (+++). An intensely dark green coloration was observed (Figure 7, No. 1), confirming complexation between phenolic hydroxyl groups and iron ions.

2. Total tannins (gelatin precipitation)

A 1% gelatin solution (in the presence of 10% NaCl) was added dropwise to 2 ml of aqueous extract.

Result: positive (++). A distinct pale flocculent precipitate was formed (Figure 7, No. 2), demonstrating the protein-precipitating property of tannins.

3. Identification of tannin type (bromine water reaction)

3–4 drops of bromine water were added to 2 ml of aqueous extract.

Result: positive (++). A distinct precipitate formed in the solution, demonstrating that the tannins present in the raw material belong to the condensed (pyrocatechol) group.

4. Catechins (vanillin–HCl reagent)

2 drops of a 1% vanillin solution in alcohol and 1 drop of concentrated HCl were added to 1 ml of 70% alcoholic extract in a porcelain dish. Result: positive (++). A distinct bright raspberry (crimson) coloration was observed (Figure 7, No. 3).

5. Total flavonoids (cyanidine test)

0.1 g of metallic magnesium (Mg) powder and 1 ml of concentrated HCl were added to 2 ml of alcoholic extract.

Result: positive (++). The solution acquired a pink-raspberry coloration of moderate intensity (Figure 7, No. 4), confirming reduction of the flavonoid nucleus.

6. Confirmation of flavonoids ($AlCl_3$ test)

1 ml of a 2% aluminum chloride solution was added to 2 ml of 70% alcoholic extract.

Result: positive (+++). An intensely yellow coloration developed (Figure 7, No. 5), accompanied by yellow-green fluorescence under UV light, confirming the presence of flavonoids bearing free hydroxyl groups.

7. Triterpenoids and steroids (Liebermann–Burchard reaction)

The dry residue obtained by evaporation of the alcoholic extract was dissolved in 1 ml of acetic anhydride, after which concentrated H_2SO_4 was carefully introduced along the wall of the flask.

Result: positive (++). A distinct blue-green or emerald-colored ring was formed at the interface of the two layers.

8. Saponins (foam test)

5 ml of aqueous extract in a test tube was vigorously shaken for 15 seconds.

Result: positive (+++). A stable, dense foam of 1.5 cm height persisting for more than 15 minutes was formed (Figure 7, No. 6).

9. Coumarins (lactone test)

10% NaOH was added to 2 ml of alcoholic extract, the mixture was heated, and then observed under UV light.

Result: positive (+). Clarification of the solution and weak yellow-green luminescence (fluorescence) were recorded.

10. Anthraquinones (Borntrager reaction)

NaOH solution was added to the hydrolyzed extract.

Result: negative (−). The alkaline layer did not acquire a cherry-red coloration, indicating the absence of anthracene derivatives in the raw material.

11. Alkaloids (Dragendorff's reagent)

2 drops of Dragendorff's reagent were added to 2 ml of acidified filtrate.

Result: negative (−). The characteristic pink-brown precipitate was not observed.

12. Amino acids (ninhydrin test)

A 1% ninhydrin solution was added to 2 ml of aqueous extract and heated in a water bath.

Result: positive (++). A distinct blue-violet (ink-like) coloration was recorded (Figure 7, No. 7).

13. Carbohydrates (Molisch reaction)

α-Naphthol solution was added to 2 ml of aqueous extract, after which concentrated H_2SO_4 was layered beneath.

Result: positive (++). A distinct violet ring was formed at the interface of the two liquids (Figure 7, No. 8).

14. Organic acids (reaction with sodium bicarbonate)

$NaHCO_3$ powder was added to the aqueous extract.

Result: positive (+++). Active effervescence – vigorous evolution of carbon dioxide (CO_2) – was observed.

15. Ascorbic acid (reaction with 2,6-dichlorophenolindophenol)

To 2 ml of aqueous extract, a solution of 2,6-dichlorophenolindophenol was added.

Result: positive (++). A rapid decolorization of the reagent's bluish-pink color was observed, which indicated the reducing properties of ascorbic acid.

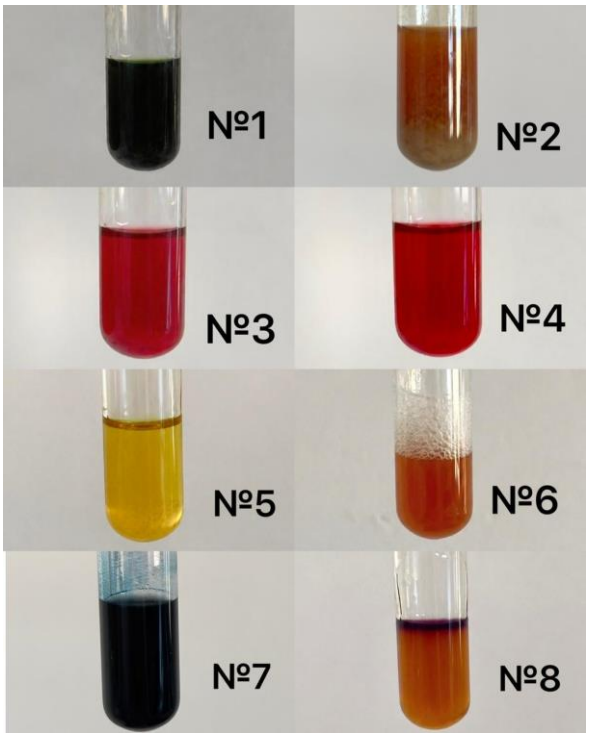

Figure 7. Results of qualitative analysis of BAS groups in dog rosehip roots. *Labels: No. 1 – total phenolic compounds ($FeCl_3$ test); No. 2 – total tannins (gelatin precipitation); No. 3 – catechins (vanillin–HCl reagent); No. 4 – flavonoids (cyanidine test); No. 5 – flavonoids ($AlCl_3$ test); No. 6 – saponins (foam test); No. 7 – amino acids (ninhydrin test); No. 8 – carbohydrates (Molisch reaction)*

The results of the analysis are systematized in Table 10.

Table 10. Results of qualitative analysis of BAS groups in dog rosehip roots

No.	BAS group	Reaction	Result*	Basis
1	Total phenolic compounds	$FeCl_3$	+++	Intensely dark green coloration; indicates high concentration of phenolic hydroxyl groups
2	Tannins (total)	Gelatin precipitation	++	Pale flocculent precipitate formed
3	Tannins (type identification)	Bromine water	++	Precipitate formed; confirms presence of condensed group
4	Catechins	Vanillin–HCl	++	Bright raspberry coloration – moderate intensity
5	Flavonoids (total)	Cyanidine test (Mg + HCl)	++	Pink-raspberry coloration – moderate intensity
6	Flavonoids (confirmation)	$AlCl_3$ test	+++	Intensely yellow coloration + fluorescence under UV light
7	Triterpenoids / steroids	Liebermann–Burchard	++	Emerald-colored ring – moderate intensity
8	Saponins	Foam test	+++	Stable dense foam, height 1.5 cm, persisting > 15 min
9	Coumarins	Lactone test (NaOH + UV)	+	Solution clarification + weak yellow-green fluorescence
10	Anthraquinones	Bornträger reaction	−	No cherry-red coloration observed
11	Alkaloids	Dragendorff's reagent	−	No pink-brown precipitate observed
12	Amino acids	Ninhydrin test	++	Blue-violet coloration – moderate intensity
13	Carbohydrates	Molisch reaction	++	Violet ring – moderate intensity

14	Organic acids	$NaHCO_3$ reaction	+++	Active effervescence – vigorous CO_2 evolution
15	Ascorbic acid	2,6-dichlorophenolindophenol	++	A rapid decolorization of the bluish-pink color was observed

**Intensity scale: +++ high intensity; ++ moderate intensity; + weak intensity; − negative*

3.5 Quantitative Analysis of the Principal Groups of Biologically Active Substances in Dog Rosehip Roots

For the purpose of evaluating the chemical composition of *Rosa canina* L. roots collected from Katon-Karagay National Park, quantitative analysis of the principal BAS groups was performed. All determinations were carried out on the basis of three independent parallel measurements (n=3). Results were expressed as a percentage (%) on an absolutely dry raw material basis; the moisture content of the raw material (W = 7.41%) was taken into account in all calculations. Statistical processing included calculation of the arithmetic mean (M) and standard deviation (SD).

Determination of total polyphenol content

The determination of polyphenolic compounds was performed by the spectrophotometric method using the Folin–Ciocalteu reagent. In accordance with the Bouguer–Lambert–Beer law, in an alkaline medium the phenolic hydroxyl groups of polyphenolic compounds reduce the phosphotungstic-molybdic heteropolyacids of the Folin–Ciocalteu reagent, forming an intensely blue-colored complex with maximum absorbance at 765 nm. Quantitative calculation was performed relative to the state reference standard (SRS) of gallic acid.

Construction of the calibration curve. A stock solution of gallic acid SRS (C_0 = 1.0 mg/ml) was prepared in 70% ethyl alcohol. A series of working solutions was prepared at six concentrations by sequential dilution. The optical density (A) of each concentration was measured in triplicate (n=3) on a Cary-60 UV-Vis spectrophotometer at 765 nm. Calibration data are presented in Table 11.

Table 11. Optical densities of gallic acid reference solutions (λ = 765 nm, n = 3)

No.	**Concentration, mg/ml**	**A_1**	**A_2**	**A_3**
1	0.00	0.000	0.000	0.000
2	0.02	0.168	0.170	0.169
3	0.04	0.337	0.341	0.339
4	0.06	0.507	0.510	0.508

5	0.08	0.676	0.679	0.677
6	0.10	0.845	0.848	0.847

Based on the experimental data in Table 11, a calibration curve of optical density versus gallic acid concentration was constructed (Figure 8). The linear regression equation is:

$$y = 8.4686x - 0.0001,\ R^2 = 0.999$$

Where *y* is optical density and *x* is gallic acid concentration (mg/ml). The high coefficient of determination confirms compliance with the Bouguer–Lambert–Beer law throughout the studied concentration range (0.00–0.10 mg/ml).

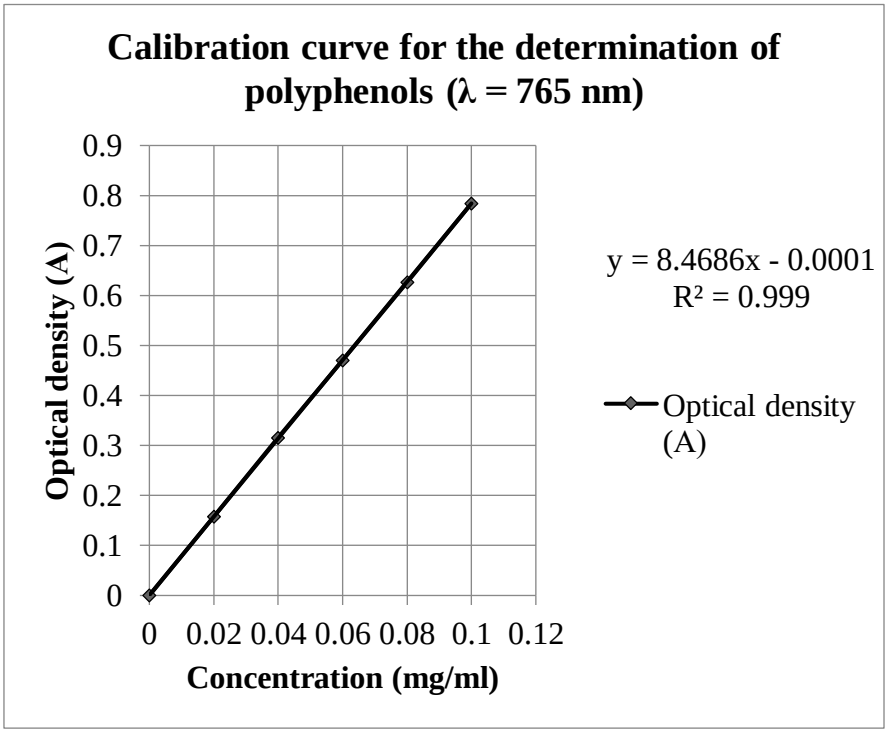

Figure 8. Calibration curve of gallic acid reference solution for the determination of total polyphenols. Folin–Ciocalteu spectrophotometric method (λ = 765 nm). X-axis: gallic acid concentration (mg/ml); Y-axis: optical density (A). Regression equation: $y = 8.4686x - 0.0001$; $R^2 = 0.999$.

Preparation of the test extract. A precisely weighed 1.0 g sample of comminuted root raw material (m) was placed in a 100 ml volumetric flask. 70% ethyl alcohol was added and the mixture was extracted under a reflux condenser in a water bath at 80°C for 45 minutes. After cooling, the extract was filtered through paper filter and the volume was adjusted to the mark with 70% alcohol (V_1 = 100 ml).

Analytical procedure. From the test extract and from the gallic acid reference solution (C = 0.10 mg/ml), 1.0 ml aliquots (V_2) were transferred into two separate 25 ml volumetric flasks. To each, 5.0 ml of Folin–Ciocalteu reagent (diluted 1:10 with water) was added and mixed thoroughly. After 3 minutes, 4.0 ml

of a 7.5% sodium carbonate (Na_2CO_3) solution was added to both flasks and the volumes were adjusted to the mark with distilled water. The mixtures were kept in the dark for 30 minutes. A mixture of 1 ml of 70% alcohol and reagents (without raw material extract) served as the control solution. The optical densities of the test extract (A) and the reference solution (A_0) were measured at 765 nm.

Calculation. The total polyphenol content (X_3, %) was calculated using the formula from Section 2.2:

$$X_3 = \frac{0.285 \times 0.10 \times 100 \times 100}{0.847 \times 1.0043 \times 1 \times (100 - 7.41)} = 3.62\%$$

The results of the spectrophotometric determination of total polyphenols in the root extract are systematized in Table 12.

Table 12. Determination of total polyphenols in roots of *Rosa canina* L.

No.	**m, g**	**A**	**A_0**	**X, %**
1	1.0042	0.285	0.847	3.62
2	1.0038	0.287	0.847	3.65
3	1.0051	0.284	0.847	3.60
Mean				3.62 ± 0.03

Result: the total polyphenol content in roots of *Rosa canina* L. is 3.62 ± 0.03% (expressed as gallic acid).

Determination of total flavonoid content

The determination of flavonoids was based on the complexation reaction with aluminum chloride ($AlCl_3$). $AlCl_3$ forms stable chelate complexes with the 4-keto group and the 3- or 5-hydroxyl groups of flavonoids, yielding a yellow-green coloration with maximum absorbance at 415 nm. Quantitative calculation was performed relative to the SRS of rutin.

Construction of the calibration curve. A stock solution of rutin SRS (C_0 = 1.0 mg/ml) was prepared by dissolution with heating in 95% ethyl alcohol. A series of working solutions was prepared at five concentrations. Calibration data are presented in Table 13.

Table 13. Optical densities of rutin reference solutions (λ = 415 nm, n = 3)

No.	**Concentration, mg/ml**	**A_1**	**A_2**	**A_3**
1	0.00	0.000	0.000	0.000
2	0.02	0.156	0.159	0.158
3	0.04	0.313	0.316	0.315
4	0.06	0.469	0.472	0.471
5	0.08	0.625	0.628	0.627
6	0.10	0.782	0.785	0.784

Based on the experimental data in Table 13, a calibration curve was constructed (Figure 9). The linear regression equation is:

$$y = 7.8329x + 0.0009,\ R^2 = 0.999$$

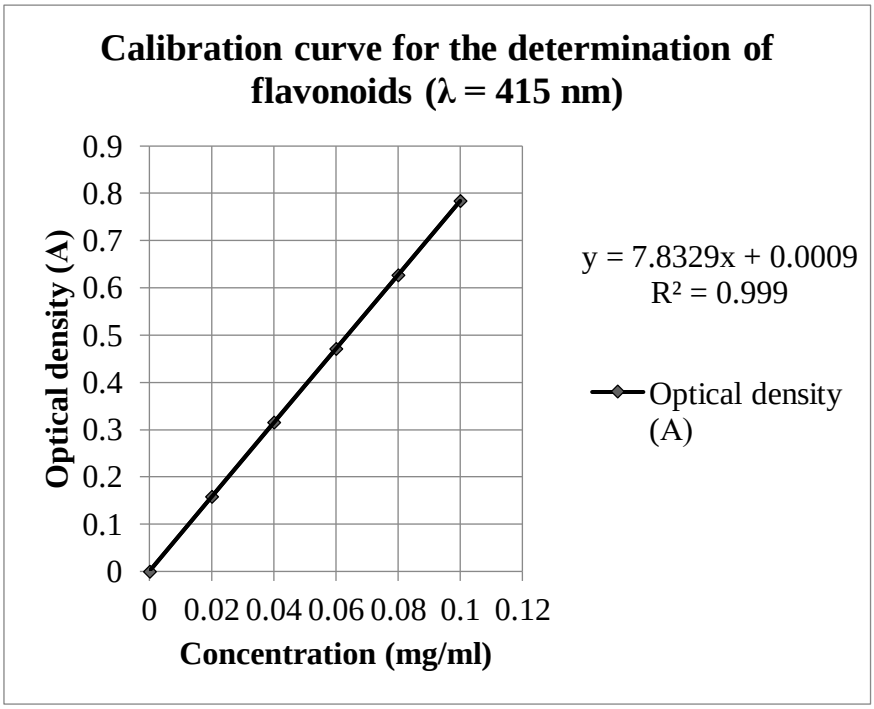

Figure 9. Calibration curve of rutin reference solution for the determination of total flavonoids. $AlCl_3$ spectrophotometric method (λ = 415 nm). X-axis: rutin concentration (mg/ml); Y-axis: optical density (A). Regression equation: y = 7.8329x + 0.0009; $R^2 = 0.999$.

Analytical procedure. From the alcoholic extract prepared for polyphenol determination, and from the rutin reference solution (C = 0.06 mg/ml), 2.0 ml aliquots (V_a) were transferred into two separate 25 ml volumetric flasks. To each, 2.0 ml of a 2% $AlCl_3$ solution in alcohol and 1 drop of 30% acetic acid were added. The solutions were adjusted to the mark with 95% ethyl alcohol and left at room temperature for 30 minutes. Mixtures consisting of the respective extract or reference standard and alcohol without $AlCl_3$ were used as control solutions. Optical density was measured at 415 nm.

Calculation. The flavonoid content (X_4, %) was calculated using the formula from Section 2.2:

$$X_4 = \frac{0.118 \times 0.0060 \times 100 \times 100}{0.471 \times 1.0025 \times 2 \times (100 - 7.41)} = 0.83\%$$

The results are systematized in Table 14.

Table 14. Determination of total flavonoids in roots of *Rosa canina* L.

No.	**m, g**	**A**	**A_0**	**X, %**

1	1.0025	0.118	0.471	0.83
2	1.0031	0.121	0.471	0.85
3	1.0028	0.115	0.471	0.81
Mean				0.83 ± 0.03

Result: the total flavonoid content in roots of *Rosa canina* L. is 0.83 ± 0.03% (expressed as rutin).

Quantitative determination of tannins

The determination of tannins was performed by permanganatometric titrimetry. Indigo sulfonic acid was used as indicator; tannins and other oxidizable compounds in the extract are oxidized by $KMnO_4$. Subtraction of the background oxidation value determined in the blank experiment yields the actual tannin content.

Procedure. A precisely weighed 2.0 g sample (m) was placed in a 500 ml flask and 250 ml (V_3) of boiling distilled water was added. The mixture was extracted under gentle reflux for 30 minutes on an electric hot plate, then filtered through a cotton filter into a 250 ml volumetric flask. A 25 ml aliquot (V_a) was transferred into a 1000 ml conical flask; 500 ml of distilled water and 25 ml of indigo sulfonic acid (indicator) were added. The resulting blue solution was titrated with a 0.02 mol/L $KMnO_4$ solution to a stable golden-yellow endpoint (V). A blank experiment was performed without raw material (V_0).

Calculation. The tannin content (X_5, %) was calculated using the formula from Section 2.2:

$$X_5 = \frac{2.0 \times 1.000 \times 0.004157 \times 250 \times 100}{2.0145 \times 25 \times (100 - 7.41)} = 0.42\%$$

The results are systematized in Table 15.

Table 15. Determination of tannins in roots of *Rosa canina* L.

No.	**m, g**	**V, ml**	**V_0, ml**	**ΔV, ml**	**X, %**
1	2.0145	19.8	17.8	2.0	0.42
2	2.0152	19.9	17.8	2.1	0.43
3	2.0148	19.7	17.8	1.9	0.41
Mean					0.42 ± 0.01

Result: the total tannin content in roots of *Rosa canina* L. is 0.42 ± 0.01%.

Quantitative determination of ascorbic acid (Vitamin C)

The determination of ascorbic acid was performed by iodometric titration.

At the equivalence point, an excess drop of iodine reacts with the starch indicator, imparting a distinct blue color to the solution. The extract is prepared in a 2% HCl medium to inhibit the natural oxidation of ascorbic acid and to provide the optimal acidic environment for the iodometric reaction.

Procedure. A precisely weighed 5.0 g sample (m) was triturated in a porcelain mortar with a small amount of 2% HCl to a homogeneous mass, quantitatively transferred into a 100 ml volumetric flask, and the volume was adjusted to the mark with 2% HCl (V_1). After standing for 15 minutes, the mixture was filtered through a dry paper filter. A 10 ml aliquot (V_2) was transferred into a conical flask, and 1 ml of a 1% starch indicator solution was added. The standard iodine solution (0.001 N I_2, titer in terms of ascorbic acid T = 0.22 mg/ml) was added dropwise from a burette; titration was continued until a stable pale blue color persisted for ≥ 1 minute (V).

Calculation. The ascorbic acid content (X_6, %) was calculated using the formula from Section 2.2:

$$X_6 = \frac{1.25 \times 0.22 \times 100 \times 100}{5.0042 \times 10 \times (100-7.41)} = 0.065\%$$

The results are systematized in Table 16.

Table 16. Determination of ascorbic acid in roots of *Rosa canina* L.

No.	**m, g**	**V, ml**	**T, mg/ml**	**X, %**
1	5.0042	1.25	0.22	0.065
2	5.0038	1.28	0.22	0.067
3	5.0055	1.22	0.22	0.063
Mean				0.065 ± 0.002

Result: the ascorbic acid content in roots of *Rosa canina* L. is 0.065 ± 0.002%.

All results obtained are summarized in Table 17.

Table 17. Results of quantitative analysis of BAS groups in roots of *Rosa canina* L.

Indicator	**Value, % of dry raw material mass (M±SD)**	**Analytical method**
Total polyphenols	3.62 ± 0.03	Spectrophotometry (Folin–Ciocalteu), λ = 765 nm
Total flavonoids	0.83 ± 0.03	Spectrophotometry ($AlCl_3$ complexation), λ = 415 nm
Tannins	0.42 ± 0.01	Titrimetry (permanganatometry)
Ascorbic acid (Vitamin C)	0.065 ± 0.002	Titrimetry (Iodometry)

3.6 Comparative Analysis of the Chemical Composition of Dog Rosehip Roots and Fruits from the Katon-Karagay Region

A comparative analysis of BAS accumulation in the underground (roots) and aboveground (fruits) organs of *Rosa canina* L. growing in Katon-Karagay National Park was conducted. The primary objective was to correlate the proposed new root raw material with the standard indicators of the official fruit raw material included in the SP RK. To ensure the reliability of the experimental data, all quantitative determinations were performed using uniform analytical methods under identical laboratory conditions (Table 18).

Table 18. Comparative BAS content in roots and fruits of *Rosa canina* L.

Plant organ	**Polyphenols, %**	**Flavonoids, %**	**Tannins, %**	**Vitamin C, %**
Roots	3.62 ± 0.03	0.83 ± 0.03	0.42 ± 0.01	0.065 ± 0.002
Fruits	4.87 ± 0.03	1.58 ± 0.03	2.01 ± 0.01	0.336 ± 0.002

The following quantitative fold differences between the morphological organs were recorded:

- Polyphenolic compounds: the value in fruits (4.87 ± 0.03%) was 1.3 times higher than in roots (3.62 ± 0.03%);
- Flavonoids: the flavonoid content in fruits (1.58 ± 0.03%) exceeded that in root raw material (0.83 ± 0.03%) by a factor of 1.9;
- Tannins: the concentration of this BAS group in fruits (2.01 ± 0.01%) was 4.8 times higher than in the root system (0.42 ± 0.01%);
- Ascorbic acid (Vitamin C): the ascorbic acid content in fruits (0.336 ± 0.002%) was 5.2 times higher than the root value (0.065 ± 0.002%), and the fruit raw material was found to fully comply with the SP RK normative requirement (≥ 0.3%).

Across all parameters studied, BAS concentrations were found to be maximally concentrated in the fruits relative to the roots.

For the purpose of characterizing the phytochemical uniqueness of the root raw material and verifying the quantitative results, a series of specific qualitative reactions targeting individual phenolic compounds was performed on the root extract. The results demonstrated that the root system possesses a distinct phytochemical profile compared to the fruits:

1. Reaction for gallic acid ($FeCl_3$ test). Upon addition of a 1% iron(III) chloride ($FeCl_3$) solution to the root extract, an intense blue-black precipitate was formed. This constitutes an important analytical indicator confirming the presence of free gallic acid and hydrolyzable tannins. In fruit samples, the color intensity of this reaction was lower than in the roots (Figure 10).

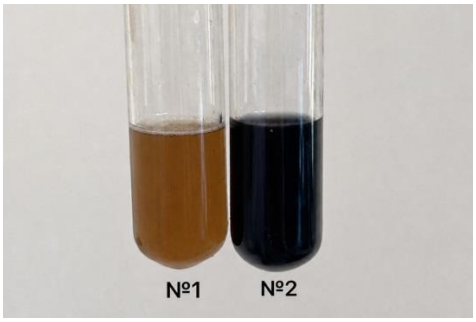

Figure 10. Results of the reaction for gallic acid. *Labels: No. 1 – fruit; No. 2 – root*

2. Reaction for ellagic acid (Griesmeyer–Reichel test). Upon addition of nitrous acid (HNO_2) to the root extract, followed by alkalinization with sodium hydroxide (NaOH) after the exposure period, a shift in solution color from bright red to violet was observed. In fruit samples, this reaction yielded a negative result (Figure 11).

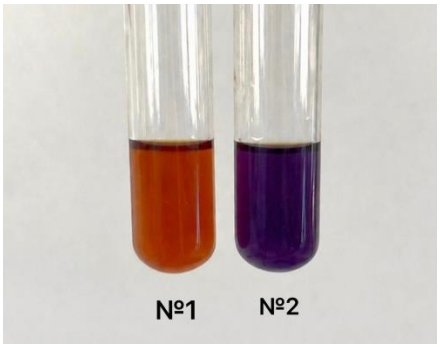

Figure 11. Results of the reaction for ellagic acid. *Labels: No. 1 – fruit; No. 2 – root*

3. Reaction for catechins (epicatechin) (vanillin–HCl test). Upon interaction with a solution of vanillin in concentrated hydrochloric acid, the root extract produced a distinct raspberry-colored staining, indicating a high concentration of epicatechin and other catechin-type compounds in the root. In fruit samples, this reaction yielded a weakly positive result (Figure 12).

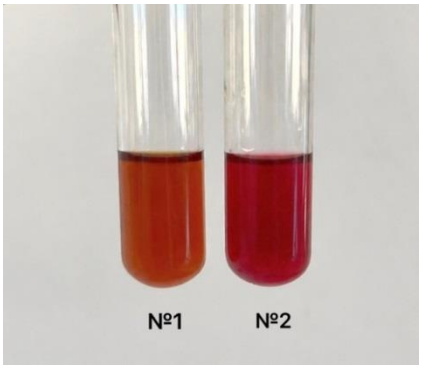

Figure 12. Results of the reaction for epicatechin. *Labels: No. 1 – fruit; No. 2 – root*

Although the root raw material contains lower quantities of BAS than the fruits, it is distinguished by the individual uniqueness of its phenolic complex. The presence of specific markers – ellagic acid, gallic acid, and epicatechin – confirmed by qualitative reactions, defines the characteristic pharmacognostic profile of the root system. The results obtained provide a scientific basis for establishing the qualitative and quantitative normative parameters required for the standardization of dog rosehip roots as an independent medicinal raw material.

3.7 Comparative Analysis of the Chemical Composition of Dog Rosehip Roots from the Katon-Karagay Region with Other Geographical Populations

For the purpose of identifying the regional characteristics of the population under investigation, a comprehensive comparative analysis of the chemical composition of the roots was conducted against published data from other geographical locations (Table 19).

The following fundamental studies served as reference sources for comparison:

1. Pharmacognostic studies of *Rosa canina* L. roots from the North Caucasus flora of Russia, by N.N. Vdovenko-Martynova et al. (2011) [97, 98];
2. Comprehensive analyses of *Rosa canina* L. roots collected from seven regions of Ukraine, by T.V. Oproshanska et al. (2017–2021) [76, 101, 102];
3. Data on the quantitative content of phenolic compounds and their bioavailability in *Rosa canina* L. roots from the Erzurum (Aktoprak) region of Turkey, by M. Masit et al. (2024) [103].

Table 19. Geographical variability of the chemical composition of *Rosa canina* L. roots

Region	**Geographical location**	**Polyphenols, %**	**Flavonoids, %**	**Tannins, %**	**Vitamin C, %**	**Source**
Kazakhstan	Katon-Karagay district, East Altai	3.62 ± 0.03	0.83 ± 0.03	0.42 ± 0.01	0.065 ± 0.002	Own data
Russia	North Caucasus	N/D	N/D	10.77	0.26	[Vdovenko-Martynova et al., 2011]
Ukraine	Kharkiv, Chernivtsi, Kyiv, Vinnytsia, Khmelnytskyi, Ternopil, Ivano-	0,083–0,092	0.80–2.50	4.50–11.0	0.11–0.13	[Oproshanska et al., 2021]

	Frankivsk regions					
Turkey	Erzurum (Aktoprak)	2.72–4.80	1.11–2.4	N/D	N/D	[Masit et al., 2024]

Notes: 1. N/D – not determined (data not available in the cited literature). 2. To ensure comparability of data, values for the Turkish population were recalculated from mg/g to %.

The following quantitative observations were recorded in the course of the comparative analysis:

- Polyphenolic compounds: the value in Katon-Karagay samples was 3.62 ± 0.03%. This indicator is significantly higher compared to Ukrainian populations (0.083–0.092%), and falls well within the characteristic range of the Turkish population (2.72–4.80%).
- Flavonoids: the value in the raw material under study (0.83 ± 0.03%) corresponds to the minimum threshold of Ukrainian samples (0.80%), but is lower than the range reported for the Turkish population (1.11–2.4%);
- Tannins: compared to the Russian (10.77%) and Ukrainian (4.5–11.0%) populations, the tannin accumulation level in Katon-Karagay samples (0.42 ± 0.01%) was considerably lower;
- Vitamin C: the Kazakhstani samples showed the lowest ascorbic acid concentration (0.065 ± 0.002%) among all populations compared; this value is 2–4 times lower than those reported for Ukrainian (0.11–0.13%) and Russian (0.26%) populations.

The data obtained demonstrate that the roots of *Rosa canina* L. from the Katon-Karagay region are distinguished from other geographical populations by their quantitative phytochemical composition profile.

DISCUSSION OF RESULTS

Discussion of Morphological-Anatomical Analysis Results

The comprehensive morphological-anatomical analysis performed on *Rosa canina* L. roots collected from Katon-Karagay National Park confirmed the authenticity of the raw material and enabled a complete characterization of its diagnostic features.

The macroscopic features identified – cylindrical shape of root fragments (length 20–30 cm, diameter 0.5–1.5 cm), deeply longitudinally furrowed rough outer surface, color range from yellowish-brown to dark brown, and thin bark closely adherent to the central wood – are fully consistent with published pharmacognostic monographs for *Rosa canina* L. The fibrous-woody fracture character anatomically demonstrates well-developed secondary xylem in mature root raw material, which in turn serves as an important indicator of biological maturity and appropriate timing of collection. The organoleptic characteristics of the raw material – a slightly astringent taste and characteristic odor – are physicochemically attributable to the presence of phenolic compounds and tannins, subsequently confirmed quantitatively in later stages of the study, which underscores the analytical value of preliminary organoleptic screening prior to targeted analytical investigations.

Microscopic analysis revealed a stable anatomical diagnostic complex characteristic of *Rosa canina* L. The multi-layered periderm (cork tissue) at the periphery of the root – functioning as an ecologically important adaptive mechanism – protects biologically active substances, particularly phenolic compounds susceptible to degradation by moisture and temperature fluctuations. The scalariform and bordered pits of the lignified conducting vessels in the xylem zone (manifested macroscopically as a dense and heavy root fracture) demonstrate the transition of the root wood from active water-conducting function to metabolite storage function. Druses of calcium oxalate (star-shaped crystal aggregates) in the parenchyma cells constitute a taxonomic-diagnostic marker common to all studied species of the genus *Rosa* and play an important role in distinguishing this taxon from other examined representatives of the family Rosaceae. Accumulation of starch grains, confirmed by the Lugol reaction, anatomically demonstrates that the root functions as an active storage organ.

The results of the morphological-anatomical analysis reliably confirm the identity of the raw material under study as *Rosa canina* L. and demonstrate its compliance with the requirements of the general pharmacopoeial article "Roots, Rhizomes, Bulbs, Tubers, and Corms" of the SP RK.

Discussion of Qualitative Indicators of Raw Material

The qualitative indicators determined for standardization purposes – moisture content, total ash, ash insoluble in 10% HCl, and extractable substances – were evaluated by comparison with the normative limits of the SP RK.

Moisture content. The mean value of 7.41 ± 0.02% was considerably below the SP RK normative limit (≤ 10%). This moisture level ensures microbiological stability of the raw material during storage. The moisture content being below the

normative limit experimentally confirms compliance of the autumn-harvested raw material drying technology (3–5 cm thin layer, turning 1–2 times daily, under a covered shelter) with GACP standards.

Total ash. The mean value across three samples was 3.63 ± 0.02%, more than twice below the upper limit established by the SP RK (≤ 7.0%). Total ash characterizes the aggregate mineral-inorganic component (calcium, magnesium, iron, etc.) of the raw material. The low total ash value in the raw material under study attests to minimal contamination of the roots with soil minerals, confirming proper harvesting and primary processing procedures. This result may also be regarded as ecologically significant data characterizing the intensity of mineral element accumulation in plant roots from the high-mountain soil of Katon-Karagay National Park.

Ash insoluble in 10% HCl. This parameter primarily reflects the content of silicon dioxide (SiO_2) – the most stringent indicator of mineral contamination in raw material. The mean value obtained (1.24 ± 0.01%) was considerably below the normative limit (≤ 3.0%). This result demonstrates that thorough rinsing with running water during primary processing effectively eliminates mineral contamination reflected by the SiO_2 content, and confirms that harvesting technology conducted in accordance with GACP standards ensures purity levels within normative requirements for raw material intended for public health applications.

Extractable substances. The mean yield of water-soluble extractives determined using purified water was 11.46 ± 0.002%. Since no standard normative value has been established in the SP RK for extractable substances in dog rosehip root raw material, this result is of primary importance as baseline data: it provides the scientific basis for the target parameter as a normative requirement for the raw material under study. The selection of purified water as an extractant is biochemically justified, as this highly polar solvent ensures the efficient extraction of polar biologically active substances, including water-soluble polyphenols, glycosides, and organic acids. The value obtained demonstrates the suitability of the raw material for pharmaceutical processing and its capacity to provide a sufficient source for producing dosage forms based on extracts, infusions, or decoctions.

Full compliance of all determined qualitative indicators with normative requirements constitutes a prerequisite for the standardization of dog rosehip roots as an independent medicinal plant raw material.

Discussion of Qualitative Phytochemical Analysis Results

Qualitative analysis of 15 BAS groups in the raw material enabled characterization of the complexity and diversity of the phytochemical profile. Scientific interpretation of the results obtained is central to the evaluation of the pharmacological potential of the raw material under study.

Reactions yielding positive results. Total phenolic compounds, tannins, catechins, flavonoids, triterpenoids and steroids, saponins, coumarins, amino acids, carbohydrates, organic and ascorbic acids were identified. The simultaneous presence of these ten BAS groups in a single raw material provides an anatomico-

chemical basis for the broad-spectrum pharmacological activity of *Rosa canina* L. roots. Specifically:

- Phenolic compounds and tannins ($FeCl_3$ reaction – dark green coloration; gelatin – pale flocculent precipitate): the bromine water typing reaction confirmed that the tannins in the root belong to the condensed (pyrocatechol) group. Condensed tannins have been demonstrated to exhibit higher antioxidant activity than hydrolyzable tannins, permitting a preliminary assessment of the antioxidant potential of the raw material under study.
- Catechins (vanillin–HCl reaction – bright raspberry coloration): the intensely positive result confirms the presence of epicatechin and other flavan-3-ols in the root. The high concentration of epicatechin in the raw material under study (approximately 3% of total catechin content) is consistent with published literature data and highlights its significance as a specific biomarker distinguishing *Rosa canina* L. roots from the fruits.
- Flavonoids ($AlCl_3$ – yellow fluorescence; cyanidine test – pink-raspberry coloration): positive results in both reactions reliably confirm the presence of flavonoids. The $AlCl_3$ reaction identifies hydroxyflavonoids (flavonol and flavone subgroups), while the cyanidine test detects flavonols and flavones via reduction of the flavan skeleton. The synergistic positive outcome of these two reactions demonstrates that the flavonoid complex of the raw material under study comprises compounds belonging to at least two chemical subgroups.
- Triterpenoids and steroids (Liebermann–Burchard reaction – emerald ring): confirmation of this group is consistent with published reports of 19α-hydroxyursane-type compounds (euscaphic acid, tormentic acid, 2-oxopomolic acid) isolated from *Rosa canina* L. roots, and corroborates the specific phytochemical profile of the raw material under study.
- Saponins (stable dense foam, ≥15 min): the presence of saponins characterizes the root raw material as a technologically promising substrate for the development of accurately dosed dosage forms (syrups, infusions, extracts), as saponins facilitate absorption and enhance the bioavailability of biologically active substances.
- Coumarins (NaOH + UV light – yellow-green fluorescence): the presence of coumarins in the root raw material suggests anticoagulant, spasmolytic, and dermatological potential, indicating important target areas for future pharmacological investigation.
- Ascorbic acid (reaction with 2,6-dichlorophenolindophenol – rapid decolorization of the reagent): A positive reaction confirms the presence of ascorbic acid in the studied raw material and demonstrates its distinct reducing properties.

Reactions yielding negative results. Anthraquinones and alkaloids were not detected. The absence of anthraquinones is a taxonomically expected result for plants of the family Rosaceae. The absence of alkaloids is regarded as favorable from the perspective of the general toxicological profile of the raw material under study.

Discussion of Quantitative Analysis Results

Quantitative phytochemical analysis of *Rosa canina* L. roots collected from Katon-Karagay National Park enabled the first characterization of the quantitative BAS profile of this raw material. The quantitative results were evaluated by comparison with the SP RK normative base, with the fruits of the plant, and with published data from other geographical populations.

Total polyphenols. The total polyphenol content determined by the Folin–Ciocalteu spectrophotometric method was 3.62 ± 0.03%. The polyphenols of *Rosa canina* L. roots are represented primarily by hydrolyzable (gallic acid- and ellagic acid-based) and condensed (proanthocyanidin) tannins, as well as phenolcarboxylic acids.

Total flavonoids. The total flavonoid content determined by the $AlCl_3$ spectrophotometric method, expressed as rutin, was 0.83 ± 0.03%. The high coefficients of determination of the linear calibration models for spectrophotometric determinations (R^2 = 0.998) confirm that all measurements were performed within the linear working range and that the Bouguer–Lambert–Beer law was fully upheld.

Tannins. The total tannin content determined by permanganatometric titrimetry was 0.42 ± 0.01%. Cross-referencing with the qualitative analysis results (Section 3.4) provides scientific grounds for postulating that the predominant fraction of the 0.42% value is attributable to condensed tannins.

Ascorbic acid. The ascorbic acid content determined by iodometric titration was 0.065 ± 0.002%. This level is a theoretically expected result: ascorbic acid biosynthesis occurs primarily in the photosynthetically active green organs of the plant, while the root system, functioning as a storage organ, synthesizes ascorbic acid in limited quantities.

The internal consistency of the quantitative analysis is further corroborated by the qualitative reaction results (Section 3.4): all positive qualitative reactions – for phenolic compounds, tannins, and flavonoids – are in logical concordance with the corresponding measured values from the quantitative determinations.

The quantitative data obtained – total polyphenols (3.62 ± 0.03%), flavonoids (0.83 ± 0.03%), tannins (0.42 ± 0.01%), and ascorbic acid (0.065 ± 0.002%) – were determined with high analytical reliability and constitute the first scientific characterization of the chemical composition of *Rosa canina* L. root raw material from Katon-Karagay National Park.

Discussion of Comparative Analysis of Roots and Fruits

Across all quantitative parameters studied, BAS concentrations were found to be higher in the fruits than in the roots: 1.3-fold for total polyphenols, 1.9-fold for flavonoids, 4.8-fold for tannins, and 5.2-fold for ascorbic acid. These results are consistent with the morphological and physiological functions of the respective plant organs.

Notwithstanding these quantitative differences, the pharmacological value of the root raw material warrants independent consideration on the following scientific grounds:

First argument – phytochemical uniqueness. The high concentrations of ellagic acid (Griesmeyer–Reichel test – positive), free gallic acid ($FeCl_3$ reaction –

intense blue-black precipitate), and epicatechin (vanillin–HCl – distinct raspberry coloration), confirmed by qualitative reactions, demonstrate that the root raw material possesses a set of specific biomarkers absent or present only in negligible amounts in the fruits. According to published data, these compounds – epicatechin, ellagic acid, and gallic acid – constitute the principal chemical substrate accounting for the antioxidant potential of the root extract (CUPRAC: 191.86 ± 17.95 mg TEAC/g) being three times higher than that of the fruit extract (64.93 ± 12.41 mg TEAC/g) [103].

Second argument – chemical stability. The BAS of the roots are considerably more resistant to oxidative degradation than the components characteristic of the fruits. The anaerobic conditions within the root system, protected by the periderm, prevent the oxidation of phenolic compounds, providing grounds for postulating that dosage forms prepared from root raw material will have a longer shelf life than fruit-based preparations.

Dog rosehip roots and fruits, while quantitatively distinct, represent independent medicinal raw materials with complementary phytochemical profiles.

Discussion of Comparative Analysis with Geographical Populations

Comparative analysis of the chemical composition of the Katon-Karagay population against data from Russia (North Caucasus), Ukraine (7 regions), and Turkey (Erzurum) enabled identification of the scientific patterns of regional variability of BAS in *Rosa canina* L.

The Katon-Karagay population aligns with the established international ranges for polyphenolic compounds (3.62%) and flavonoids (0.83%). Specifically, the polyphenolic content is well-positioned within the range reported for Turkish populations (2.72–4.80%), while the flavonoid levels are comparable to the indices observed in Ukrainian populations (0.80%). The tannin value (0.42%) were considerably lower than Russian (10.77%; 25.62%) and Ukrainian (4.5–11.0%) data. Similarly, the concentration of ascorbic acid was recorded at 0.065%, representing the minimum value among the compared populations.

The following factors may be identified as contributing to the observed variability in BAS content:

1. Altitude factor. At the high-mountain elevations of Katon-Karagay National Park (1,500–2,500 m a.s.l.), the brevity of the growing season restricts the time available for biosynthesis of accumulative metabolites, producing a clear contrast with lowland or lower-mountain regions such as the North Caucasus or Ukrainian plains.

2. Climatic factor. The continental climate of the East Altai (marked temperature contrasts, prolonged frost periods) may suppress the synthesis of osmotic stress-related metabolites) while enhancing the biosynthesis of xeromorphic adaptation metabolites in populations developing under near-polar altitudinal conditions.

3. Analytical methodological differences. The comparability of data across studies is conditional: the analytical standards employed (GOST, pharmacopoeial, ISO), extractant concentration, extraction time, and temperature

all exert a measurable influence on reported values, which must be taken into account when interpreting inter-study differences.

4. Genetic population specificity. *Rosa canina* is a polyploid, highly polymorphic species; the genotype of the East Altai population, which has developed under conditions of geographical isolation, may differ fundamentally in its metabolic profile from European or Caucasian populations.

The aggregate results of the study demonstrate that *Rosa canina* L. roots from the Katon-Karagay region warrant pharmacognostic evaluation as an independent medicinal raw material with distinctive regional characteristics.

CONCLUSION

This master's thesis was devoted to a comprehensive pharmacognostic analysis of the chemical composition of the roots of dog rosehip (*Rosa canina* L.) growing in the high-mountain ecosystem of Katon-Karagay National Park, East Kazakhstan Region. Based on the results of the research objectives accomplished, the following scientific conclusions were drawn.

1. Results of morphological-anatomical analysis

Macro- and microscopic examination of *Rosa canina* L. root raw material collected from Katon-Karagay National Park revealed a stable diagnostic complex fully consistent with published pharmacognostic monographs. At the macroscopic level, the following features were recorded: cylindrical shape of root fragments (length 20–30 cm, diameter 0.5–1.5 cm), deeply longitudinally furrowed surface, color range from yellowish-brown to dark brown, and fibrous-woody fracture. At the microscopic level, the diagnostic complex characteristic of *Rosa canina* L. was identified – multi-layered periderm, lignified secondary xylem elements, druses of calcium oxalate in parenchyma cells, and starch grains confirmed by the Lugol reaction. The morphological-anatomical data obtained reliably confirm the authenticity of the raw material under study and demonstrate its full compliance with the requirements of the general pharmacopoeial article "Roots, Rhizomes, Bulbs, Tubers, and Corms" of the SP RK.

2. Results of qualitative normative indicators

The principal qualitative indicators of *Rosa canina* L. root raw material were determined within the normative limits of the SP RK:

- Moisture content: 7.41 ± 0.02% (requirement: ≤ 10%);
- Total ash: 3.63 ± 0.02% (requirement: ≤ 7.0%);
- Ash insoluble in 10% HCl: 1.24 ± 0.01% (requirement: ≤ 3.0%);
- Water-soluble extractives: 11.46 ± 0.002%.

Full compliance of all qualitative normative indicators with the established limits demonstrates the microbiological stability of the raw material, the absence of mineral contamination, and its technological suitability for pharmaceutical processing.

3. Results of qualitative phytochemical analysis

Qualitative phytochemical screening encompassing 15 reactions performed on aqueous and alcoholic extracts of *Rosa canina* L. roots identified the presence of a complex of biologically active substances. 11 reactions yielded positive results: total phenolic compounds ($FeCl_3$ reaction), total tannins and condensed (pyrocatechol-type) tannins (gelatin precipitation, bromine water reaction), catechins (vanillin–HCl), flavonoids ($AlCl_3$ and cyanidine tests), triterpenoids and steroids (Liebermann–Burchard reaction), saponins (foam test), coumarins (lactone test), amino acids (ninhydrin test), carbohydrates (Molisch reaction), organic acids ($NaHCO_3$ reaction) and ascorbic acid (2,6-dichlorophenolindophenol reaction). Anthraquinones and alkaloids were not detected, which represents a taxonomically expected and toxicologically favorable result for representatives of the family Rosaceae.

4. Results of quantitative analysis

The content of the principal BAS groups, determined by spectrophotometric (Folin–Ciocalteu, $AlCl_3$) and titrimetric (permanganatometry, Tillmans' reagent) methods, was as follows:

- Total polyphenols (expressed as gallic acid): 3.62 ± 0.03%;
- Total flavonoids (expressed as rutin): 0.83 ± 0.03%;
- Tannins: 0.42 ± 0.01%;
- Ascorbic acid (Vitamin C): 0.065 ± 0.002%.

All quantitative determinations were performed on the basis of three parallel measurements (n=3) and results were expressed on an absolutely dry raw material basis.

5. Results of comparative analysis of roots and fruits

Comparative phytochemical analysis of roots and fruits demonstrated that BAS concentrations are higher in the fruits across all quantitative parameters studied. However, quantitative inferiority does not negate the independent pharmacognostic significance of the root raw material: the accumulation in the root system of specific biomarkers (ellagic acid, free gallic acid, and epicatechin) absent or present only in negligible amounts in the fruits, scientifically demonstrates that the two organs constitute independent medicinal raw materials with complementary and distinct phytochemical profiles.

6. Results of comparative analysis with geographical populations

Comparative analysis of the chemical composition of the Katon-Karagay population against data from Russia (North Caucasus), Ukraine (7 regions), and Turkey (Erzurum) revealed pronounced geographical variability in the BAS of *Rosa canina* L. The polyphenolic complex of the Katon-Karagay samples (3.62%) falls well within the established range of the Turkish population (2.72–4.80%), while the flavonoid content (0.83%) is consistent with the indices reported for Ukrainian populations (0.80%). Conversely, the concentrations of tannin (0.42%) and ascorbic acid (0.065 %) were notably lower than those of the compared regions. Such results are characteristic of the low-vitamin profile of *Rosa canina* L. and are attributable to the synergetic influence of high altitude, a shortened growing season, specific continental climatic conditions, and population-specific genetic traits. The results obtained represent the first data for the Central Asian region in this field and contribute new information to the geographical chemotaxonomy of *Rosa canina* L.

The totality of results obtained forms the scientific basis for the standardization of *Rosa canina* L. roots as an independent medicinal plant raw material and for their introduction into pharmaceutical circulation as a new domestic medicinal plant raw material.

LIST OF REFERENCES:

1. Linnaeus C. Species Plantarum. Stockholm: Laurentius Salvius; 1753. p. 1200.
2. Maryrm IS, Muhammad IS, Mir AK. Bibliography of the genus *Rosa* L. Hamdard Medicus. 2008;46(3):5-11.
3. World Flora Online. *Rosa* L. [Internet]. WFO; 2024 [cited 2024]. Available from: http://www.worldfloraonline.org/taxon/wfo-4000033476
4. Wissemann V, Ritz CM. The genus *Rosa* (Rosoideae, Rosaceae) revisited: molecular analysis of nrITS-1 and atpB-rbcL intergenic spacer (IGS) versus conventional taxonomy. Bot J Linn Soc. 2005;147(3):275-290.
5. Sokolova TA, editor. Decorative plant growing. Arboriculture [Dekorativnoye rasteniyevodstvo. Drevovodstvo]: textbook. Moscow: Academia; 2004. 352 p. Russian.
6. Wulf EV, Maleeva OF. World resources of useful plants: A handbook [Mirovyye resursy poleznykh rasteniy: Spravochnik]. Bakhteev FK, editor. Leningrad: Nauka; 2009. p. 179-200. Russian.
7. Budantsev AL, Lesiovskaya EE, editors. Wild useful plants of Russia [Dikorastushchiye poleznyye rasteniya Rossii]. St. Petersburg: SPHFA; 2001. p. 493-495. Russian.
8. Buzunova IO, editor. *Rosa* L. – Rosehip, or rose. Flora of the middle zone of the European part of Russia [Rosa L. – Shipovnik, ili roza. Flora sredney polosy yevropeyskoy chasti Rossii]: textbook. Moscow: KMK; 2014. p. 158-179. Russian.
9. Jian HY, Yan MS, Zhang HJ, et al. Molecular phylogeny of the genus *Rosa* (Rosaceae) based on chloroplast and nuclear DNA sequences. Sci Rep. 2015;5:16838. doi:10.1038/srep16838.
10. Wissemann V. The evolutionary history of the genus *Rosa* L. (Rosaceae). Biol J Linn Soc. 2007;90(1):15-30.
11. Fougère-Danezan M, Joly S, Bruneau A, et al. Phylogeny and biogeography of wild roses with specific attention to polyploids. Ann Bot. 2015;115(2):275-291.
12. Ryabinina OA, Lyavdanskaya GT, Bastaeva GT, et al. IOP Conf Ser: Earth Environ Sci. 2020;624:012015. doi:10.1088/1755-1315/624/1/012015.
13. Baytenov MS. Genus 27. Rosehip – *Rosa* Linnaeus. In: Baytulin IO, editor. Flora of Kazakhstan. Generic complex of flora [Flora Kazakhstana. Rodovoy kompleks flory]. Almaty: Gylym; 2001. Vol 2. p. 116. Kazakh.
14. Pavlov NV, editor. Flora of Kazakhstan [Flora Kazakhstana]. Alma-Ata: Academy of Sciences of the Kazakh SSR; 1961. Vol 4. 571 p. Russian.
15. Andrianova NG, Shyntasova NA, Ivlev VI, et al. Preliminary resource assessment of some species of rose hips in Central Kazakhstan. KazNU Bull Biol Ser. 2013;3/2(59):458-461.
16. Fisyun VV. *Rosa* L. – Rosehip. In: Pavlov NV, editor. Flora of Kazakhstan [Flora Kazakhstana]. Alma-Ata: AS of the Kazakh SSR; 1961. Vol 4. p. 485–502. Russian.

17. Khapugin AA. The genus *Rosa* L. in the Moksha river basin [Rod Rosa L. v basseyne reki Moksha] [PhD thesis]. Moscow; 2015. p. 15-43. Russian.

18. Tsioutsiou EE, Giordani P, Hanlidou E, et al. Ethnobotanical study of medicinal plants used in Central Macedonia, Greece. Evid Based Complement Alternat Med. 2019;2019:4513792. doi:10.1155/2019/4513792.

19. Wu M, Li W, Zhang Y, et al. Structure characteristics, hypoglycaemic and immunomodulatory activities of pectic polysaccharides from *Rosa setate* x *Rosa rugosa* waste. Carbohydr Polym. 2021;268:117190.

20. Chang SW, Du YE, Qi Y, et al. New depsides and neuroactive phenolic glucosides from the flower buds of Rugosa Rose (*Rosa rugosa*). J Agric Food Chem. 2019;67(26):7289-7296.

21. Dani KGS, Fineschi S, Michelozzi M, et al. Diversification of petal monoterpene profiles during floral development and senescence in wild roses. Oecologia. 2021;197(4):957-969.

22. Kwon EK, Lee DY, Lee H, et al. Flavonoids from the buds of *Rosa damascena* inhibit the activity of 3-hydroxy-3-methylglutaryl-coenzyme a reductase and angiotensin I-converting enzyme. J Agric Food Chem. 2010;58(2):882-886.

23. Ren G, Xue P, Sun X, Zhao G. Determination of the volatile and polyphenol constituents and the antimicrobial, antioxidant, and tyrosinase inhibitory activities of bioactive compounds from *Rosa rugosa* Thunb. by-product.

24. Kerasioti E, Apostolou A, Kafantaris I, et al. Polyphenolic composition of *Rosa canina, Rosa sempervivens* and *Pyrocantha coccinea* extracts and assessment of their antioxidant activity in human endothelial cells. Antioxidants. 2019;8(4):92.

25. Naumenko KO, Samoshkina OY, Zharinova ZI, Dmitrieva DV. The effect of rosehip infusion regime on the ascorbic acid content in the decoction. Bull Med Inst REAVIZ. 2022;2:374. Russian.

26. Mármol I, Sánchez-De-Diego C, Jiménez-Moreno N, et al. Therapeutic applications of rose hips from different *Rosa* species. Int J Mol Sci. 2017;18(6):1137.

27. Wu H, Li M, Yang X, et al. Extraction optimisation, physicochemical properties and antioxidant and hypoglycaemic activities of polysaccharides from roxburgh rose (*Rosa roxburghii* Tratt.) leaves. Int J Biol Macromol. 2020;165(Pt A):517-529.

28. Kubczak M, Khassenova AB, Skalski B, et al. Bioactive compounds and antiradical activity of the *Rosa canina* L. leaf and twig extracts. Agronomy. 2020;10(12):1897.

29. Roman I, Stanil A, Stanil S. Bioactive compounds and antioxidant activity of *Rosa canina* L. biotypes from spontaneous flora of Transylvania. Chem Cent J. 2013;7(1):73.

30. Aleksashina SA, Makarova NV, Demenina LG. Antioxidant potential of rosehip fruits. Nutrition Issues [Voprosy Pitaniia]. 2019;88(3):84-89. doi:10.24411/0042-8833-2019-10033. Russian.

31. Muranets AP, Esimseitova AK, Dyusembekova DA, et al. Study of the biodiversity of wild plants of the genus *Rosa* L. in Kazakhstan and their molecular genetic identification. Bull LN Gumilyov ENU Biosci Ser. 2022;139(2):44.

32. Nowak R, Gawlik-Dziki U. Polyphenols of *Rosa* L. leaves extracts and their radical scavenging activity. Z Naturforsch C J Biosci. 2007;62(1-2):32-38.

33. Christensen L. Galactolipids as potential health promoting compounds in vegetable foods. Recent Pat Food Nutr Agric. 2012;1(1):50-58.

34. Tang Y, Zhu ZY, Liu Y, et al. The chemical structure and anti-ageing bioactivity of an acid polysaccharide obtained from rose buds. Food Funct. 2018;9(4):2300-2312.

35. Chang SW, Du YE, Qi Y, et al. New depsides and neuroactive phenolic glucosides from the flower buds of Rugosa Rose (*Rosa rugosa*). J Agric Food Chem. 2019;67(26):7289-7296.

36. Polumackanycz M, Kaszuba M, Konopacka A, et al. Phenolic composition and biological properties of wild and commercial Dog Rose fruits and leaves. Molecules. 2020;25(22):5272.

37. Phetcharat L, Wongsuphasawat K, Winther K. The effectiveness of a standardised rose hip powder, containing seeds and shells of *Rosa canina*, on cell longevity, skin wrinkles, moisture, and elasticity. Clin Interv Aging. 2015;10:1849-1856.

38. Moradkhani S, Rezaei-Dehghanzadeh T, Nili-Ahmadabadi A. *Rosa persica* hydroalcoholic extract improves cadmium hepatotoxicity by modulating oxidative damage and tumour necrosis factor-alpha status. Environ Sci Pollut Res Int. 2020;27(25):31259-31268.

39. Sadeghi H, Hosseinzadeh S, Touri MA, et al. Hepatoprotective effect of *Rosa canina* fruit extract against carbon tetrachloride induced hepatotoxicity in rat. Avicenna J Phytomed. 2016;6(2):181-188. PMID: 27222831.

40. Shaushekov K, Omarova KT, Pak RN, et al. Cholagogue and hepatoprotective properties of thick extract from *Rosa laxa* (Rosaceae) fruit. Plant Resources [Rastitel'nyye resursy]. 2005;41(3):117-124. Russian.

41. Komiazyk M, Palczewska M, Sitkiewicz I, et al. Neutralisation of cholera toxin by Rosaceae family plant extracts. BMC Complement Altern Med. 2019;19(1):140.

42. Ashtiyani SC, Najafi H, Jalalvandi S, Hosseinei F. Protective effects of *Rosa canina* L. fruit extracts on renal disturbances induced by reperfusion injury in rats. Iran J Kidney Dis. 2013;7(4):290-298.

43. Tayefi-Nasrabadi H, Sadati-Eshikli S, Aghapour Z. The effects of the hydroalcohol extract of *Rosa canina* L. fruit on experimentally nephrolithiasic Wistar rats. Phytother Res. 2012;26(1):78-85.

44. Cheng BCY, Fu XQ, Guo H, et al. The genus *Rosa* and arthritis: Overview on pharmacological perspectives. Pharmacol Res. 2016;114:219-234.

45. Taghizadeh M, Rashidi AA, Taherian AA, et al. The protective effect of hydroalcoholic extract of *Rosa canina* (Dog Rose) fruit on liver function and

structure in streptozotocin-induced diabetes in rats. J Diet Suppl. 2018;15(5):624-635.

46. Ninomiya K, Matsuda H, Kubo M, et al. Potent anti-obese principle from *Rosa canina*: Structural requirements and mode of action of trans-tiliroside. Bioorg Med Chem Lett. 2007;17(11):3059-3064.

47. Turan I, Demir S, Kilinc K, et al. Cytotoxic effect of *Rosa canina* extract on human colon cancer cells through repression of telomerase expression. J Pharm Anal. 2018;8(6):394-399.

48. Sabitov A, Gaweł-Beben K, Sakipova Z, et al. *Rosa platyacantha* Schrenk from Kazakhstan–Natural source of bioactive compounds with cosmetic significance. Molecules. 2021;26(9):2578.

49. Jiménez S, Gascón S, Luquin A, et al. *Rosa canina* extracts have antiproliferative and antioxidant effects on caco-2 human colon cancer. PLoS ONE. 2016;11(7):e0159136.

50. Guimarães R, Barros L, Calhelha RC, et al. Bioactivity of different enriched phenolic extracts of wild fruits from Northeastern Portugal: A comparative study. Plant Foods Hum Nutr. 2014;69(1):37-42.

51. Lee YH, Jung MG, Kang HB, et al. Effect of anti-histone acetyltransferase activity from *Rosa rugosa* Thunb. (Rosaceae) extracts on androgen receptor-mediated transcriptional regulation. J Ethnopharmacol. 2008;118(3):412-417.

52. Sargin SA. Potential anti-influenza effective plants used in Turkish folk medicine. J Ethnopharmacol. 2021;265:113319.

53. European Commission. Cosmetic Ingredient Database (CosIng) [Internet]. [cited 2024]. Available from: https://ec.europa.eu/growth/sectors/cosmetics/cosing_en

54. Kwak CS, Yang J, Shin CY, Chung JH. *Rosa multiflora* Thunb flower extract attenuates ultraviolet-induced photoaging in skin cells and hairless mice. J Med Food. 2020;23(9):988-997.

55. Hayta S, Polat R, Selvi S. Traditional uses of medicinal plants in Elazig (Turkey). J Ethnopharmacol. 2014;154(3):613-623.

56. Shin EJ, Han AR, Lee MH, et al. Extraction conditions for *Rosa gallica* petal extracts with anti-skin ageing activities. Food Sci Biotechnol. 2019;28(5):1439-1446.

57. Fujii T, Ikeda K, Saito M. Inhibitory effect of rose hip (*Rosa canina* L.) on melanogenesis in mouse melanoma cells and on pigmentation in brown guinea pigs. Biosci Biotechnol Biochem. 2011;75(3):489-495.

58. Song YR, Lim WC, Han A, et al. Rose petal extract (*Rosa gallica*) exerts skin whitening and anti-skin wrinkle effects. J Med Food. 2020;23(8):870-878.

59. Plant resources of the USSR: Flowering plants, their chemical composition, and use [Rastitel'nyye resursy SSSR: tsvetkovyye rasteniya, ikh khimicheskiy sostav, ispol'zovaniye]. Hydrangeaceae-Haloragaceae family. Leningrad; 1987. p. 326. Russian.

60. State register of medicines and medical devices [Gosudarstvennyy reyestr lekarstvennykh sredstv i meditsinskikh izdeliy] [Internet]. [cited 2024]. Available from: http://register.ndda.kz/category/search_prep137. Russian/Kazakh.

61. Mashkovsky MD. Medicinal products [Lekarstvennyye sredstva]. Moscow: Novaya Volna; 2021. 1216 p. Russian.

62. Medicinal plants of Kazakhstan and their use [Lekarstvennyye rasteniya Kazakhstana i ikh ispol'zovaniye]. Almaty; 1996. 344 p. Russian.

63. Kukenov MK, editor. Medicinal plants of Kazakhstan and their use [Lekarstvennyye rasteniya Kazakhstana i ikh ispol'zovaniye]. Almaty; 1998. 345 p. Russian.

64. Grudzinskaya LM, Gemedzhieva NG, Nelina NV, Karzhaubekova ZhZh. Annotated list of medicinal plants of Kazakhstan: Reference publication [Annotirovannyy spisok lekarstvennykh rasteniy Kazakhstana: Spravochnoye izdaniye]. Almaty; 2014. p. 200. Russian.

65. Haas LF. *Rosa canina* (dog rose). J Neurol Neurosurg Psychiatry. 1995;59(5):470. doi:10.1136/jnnp.59.5.470.

66. Knowles E. Dog. In: The Oxford Dictionary of Phrase and Fable. 2nd ed. Oxford University Press; 2005.

67. Howard M. Traditional Folk Remedies. Century; 1987. p. 133.

68. Annenkov N. Botanical dictionary [Botanicheskiy slovar']. St. Petersburg: Imperial Academy of Sciences; 1878. p. 300-301. Russian.

69. Khrzhanovsky VG. Roses. Phylogeny and systematics [Rozy. Filogeniya i sistematika]. Moscow: Sovetskaya Nauka; 1958. p. 177-183. Russian.

70. Yakovlev GP, Chelombitko VA. Botany [Botanika]: textbook for universities. Kamelin RV, editor. St. Petersburg: SpetsLit; 2003. p. 434. Russian.

71. Atlas of medicinal plants of Russia [Atlas lekarstvennykh rasteniy Rossii]. Moscow: Nauka; 2021. 646 p. Russian.

72. Lavrenova GV. Family encyclopedia of folk medicine from Dr. Lavrenova [Semeynaya entsiklopediya narodnoy meditsiny ot doktora Lavrenovoy]. Moscow: OLMA Media Group; 2010. 550 p. Russian.

73. Sorokopudov VN, Netrebenko NN, Evtukhova MV, Pisarev DI. Morphological and anatomical aspects of some representatives of the genus *Rosa* L. [Morfologo-anatomicheskiye aspekty nekotorykh predstaviteley roda Rosa L]. Bull KrasGAU. 2009;11(38):50-54. Russian.

74. Sergunova EV. Research on the standardization of rose hips and dosage forms based on it [Issledovaniya po standartizatsii plodov shipovnika i lekarstvennykh form na ego osnove] [PhD thesis]. Moscow; 2002. 149 p. Russian.

75. Selivanova EM, Babaeva EY. Sepals of rose hips - a promising type of medicinal plant raw material [Chashelistiki plodov shipovnika - perspektivnyy vid lekarstvennogo rastitel'nogo syr'ya]. IX Int Sci Conf Young Scientists. Moscow: VILAR; 2021. p. 372-378. Russian.

76. Oproshanska T, Khvorost O. Comparative pharmacognostical study of roots of *Rosa majalis* and *Rosa canina* L. ScienceRise: Pharm Sci. 2021;33(5):77. doi:10.15587/2519-4852.2021.243385.

77. Ibn Sina AA. The Canon of Medicine [Kanon vrachebnoy nauki]. Tashkent: Fan; 1996. Russian translation.

78. Kovaleva NG. Crimean rose (*Rosa gallica* L.). In: Treatment with plants [Lecheniye rasteniyami]. Moscow: Meditsina; 1972. p. 209-211. Russian.

79. Lavrenova GV. Family encyclopaedia of folk medicine. Moscow: OLMA Media Group; 2010.

80. Gammerman AF, Kadaev GN, Yatsenko-Khmelevsky AA. Medicinal plants (healer plants) [Lekarstvennyye rasteniya (rasteniya-tseliteli)]. Moscow: Vysshaya Shkola. Russian.

81. Korsun VF, Korsun EV. Phytotherapy: traditions and modernity [Fitoterapiya: traditsii i sovremennost']. Moscow. Russian.

82. Nosal MA, Nosal IM. Medicinal plants and ways of their use in the people [Lekarstvennyye rasteniya i sposoby ikh primeneniya v narode]. Kyiv. Russian.

83. Sadykov NS. Medicinal plants of Kazakhstan [Kazakstannyn darilik osimdikteri]. Almaty. Kazakh.

84. Duke JA. Handbook of Medicinal Herbs. 2nd ed. CRC Press; 2002.

85. Mukanov BM. Rose hips of Kazakhstan (*Rosa* L. genus): biology, ecology and use [Kazakstannyn itmuryndary (Rosa L. tuysy): biologiyasy, ekologiyasy zhane paidalanyluy]. Almaty: Gylym; 2012. 180 p. Kazakh.

86. Otieva AV. Analysis of biologically active substances in the medicinal plant raw material of fruits and roots of rosehip. XIV All-Russ Sci-Pract Conf. Vladikavkaz: NOSU; 2023. Vol 2. p. 156-160. Russian.

87. Zhdanov DA, Kurkin VA, Braslavsky VB, et al. Actual aspects of quality control and standardization of rose hips. Dev Reg Med Prod [Razrabotka i registratsiya lekarstvennykh sredstv]. 2021;10(3):167-175. Russian.

88. Kokaeva FF, Dzhatieva DN. Study of the chemical composition of rose hips (*Rosa majalis*). Izv Gorsk State Agrar Univ. 2018;55(1):120-124. Russian.

89. Magomedova ZM, Gasanova MG. Study of the phytochemical composition of rosehip. Bull Dagestan State Univ Nat Sci. 2016;31(2):54-59. Russian.

90. Khanzhina EG, Pushkareva PV. Comparative analysis of the chemical composition of rose hips for the manufacture of oils and herbal teas. XXII All-Russ Sci-Pract Conf. Kirov: VyatSU; 2022. Vol 2. p. 159-164. Russian.

91. Shomirov BA, Choriev AZh, Akhrarov UB, Tukhtaev ShK. Study of the chemical composition of rosehips. Sci Educ. 2023;4(9):131-137. Russian.

92. Dubtsova GN, Kusova IU, Kunitsyna IK. Nutritional value of rosehip products. Nutr Issues [Voprosy Pitaniia]. 2018;87(S5):85-86. Russian.

93. Laman NA, Kopylova NI. Rosehip - a natural concentrate of vitamins and antioxidants. Sci Innov [Nauka i innovatsii]. 2017;176(10):45-49. Russian.

94. Dubtsova GN, Negmatulloeva RN, Bessonov VV, et al. Composition and content of biologically active substances in rose hips. Nutr Issues [Voprosy Pitaniia]. 2012;81(6):84-88. Russian.

95. Khomova SV. Phytochemical study and standardization of raw materials and preparations of rosehip [Fitokhimicheskoye issledovaniye i standartizatsiya syr'ya i preparatov shipovnika] [PhD thesis]. Perm; 2005. 162 p. Russian.

96. Koczorkiewicz G, et al. Cytotoxic and antimicrobial activity of triterpenoids isolated from *Rosa canina* L. roots. Molecules. 2019;24(11):21-25.

97. Vdovenko-Martynova NN, Kobylchenko NV, Blinova TI. Content of biologically active compounds in the roots of rosehip (Rosa canina L.) of the North Caucasus flora [Soderzhanie biologicheski aktivnykh soedineniy v kornyakh shipovnika (Rosa canina L.) flory Severnogo Kavkaza]. Modern Problems of Science and Education [Sovremennye problemy nauki i obrazovaniya], 2011;4:312. Russian.

98. Vdovenko-Martynova NN, Kobylchenko NV, Blinova TI. Pharmacognostic study of the roots of Dog Rose (*Rosa canina* L.) of the North Caucasus flora [Farmakognosticheskoe izuchenie korney shipovnika sobach'ego (Rosa canina L.) flory Severnogo Kavkaza] [PhD thesis abstract]. Pyatigorsk; 2011. 24 p. Russian.

99. Ousaaid D, Laaroussi H, Kamari FE, et al. Phytoactive compounds, chemical and biological functionalities of *Rosa canina* L.: a review. Discov Plants. 2025;2:258. doi:10.1007/s44372-025-00293-2.

100. Kerasioti E, Apostolou A, Kafantaris I, et al. Polyphenolic composition of *Rosa canina, Rosa sempervivens* and *Pyrocantha coccinea* extracts and assessment of their antioxidant activity in human endothelial cells. Antioxidants. 2019;8(4):92.

101. Oproshanskaya TV, Khvorost OP. Study of polyphenolic compounds of rosehip roots [Izucheniye polifenol'nykh soyedineniy korney shipovnika]. Pharm Kazakhstan. 2021;2:45-49. Russian.

102. Oproshanska TV, Khvorost OP, Ocheredko LV. Determination of the quantitative content of the amount of organic acids and ascorbic acid in different batches of *Rosa majalis* and *Rosa canina* raw material. Bull Pharm. 2017.

103. Macit M, Aras A, Capanoglu E, Bakir S. Investigating the content and bioaccessibility of phenolic compounds in roots of *Rosa canina* L. and *Rosa pimpinellifolia* L. Yüzüncü Yıl Univ J Agric Sci. 2023;33(2):163-173.

104. Subaş T, Özgen U, Kanbolat Ş, et al. Bioactive compounds from *Rosa canina* L. roots with anti-collagenase, anti-tyrosinase and antioxidant activities: In vitro and in silico approaches. Chem Biodivers. 2025. doi:10.1002/cbdv.202500357.

105. Mosina LM, Matveeva LV, Kilovataya OA, et al. Inflammatory bowel diseases as a medical and social problem. Mod Probl Sci Educ [Sovremennyye problemy nauki i obrazovaniya]. 2022;6-2:30. Russian.

106. Shaushekov K, Omarova KT, Pak RN, et al. Cholagogue and hepatoprotective properties of thick extract from *Rosa laxa* (Rosaceae) fruits. Plant Resour [Rastitel'nyye resursy]. 2005;41(3):117-124. Russian.

107. Mishchenko OYa, Yurchenko KYu, Kovalenko EN. Pharmacoeconomic aspects of the use of phytopreparations for the treatment of gastrointestinal diseases. II Int Sci-Pract Conf. Kharkiv; 2018. p. 305-308. Russian.

108. Sambukova TV, Ovchinnikov BV, Ganapolsky VP, et al. Prospects for the use of phytopreparations in modern pharmacology. Rev Clin Pharmacol Drug Ther [Obzory po klinicheskoy farmakologii i lekarstvennoy terapii]. 2017;15(2):56-63. Russian.

109. Ronzhin IV, Ponomareva EA. Statistics of gastrointestinal tract diseases: causes, symptoms, prevention. Young Sci [Molodoy uchenyy]. 2015;103(23):375-379. Russian.

110. Sadeghi H, Hosseinzadeh S, Touri MA, et al. Hepatoprotective effect of *Rosa canina* fruit extract against carbon tetrachloride induced hepatotoxicity in rat. Avicenna J Phytomed. 2016;6(2):181-188.

111. Larsen E, Kharazmi A, Christensen LP, Christensen SB. An antiinflammatory galactolipid from Rose Hip (*Rosa canina*) that inhibits chemotaxis of human peripheral blood neutrophils in vitro. J Nat Prod. 2003;66(7):994-995.

112. Jäger AK, Eldeen IM, van Staden J. COX-1 and -2 activity of rose hip. Phytother Res. 2007;21(12):1251-1252.

113. Hashem Dabaghian F, Abdollahifard M, et al. Effects of *Rosa canina* L. fruit on glycemia and lipid profile in type 2 diabetic patients: a randomized, double-blind, placebo-controlled clinical trial. J Med Plants. 2015;55(3):95-104.

114. Tumbas VT, Canadanovic-Brunet JM, et al. Effect of rosehip (*Rosa canina* L.) phytochemicals on stable free radicals and human cancer cells. J Sci Food Agric. 2012;92(6):1273-1281.

115. Yilmaz SO, Ercisli S. Antibacterial and antioxidant activity of fruits of some rose species from Turkey. Rom Biotech Lett. 2011;16(4):6407-6411.