

## STUDY OF ESSENTIAL OIL COMPOSITION USING GAS CHROMATOGRAPHY

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ABSTRACT	KEY WORDS
<p>In this study, the chemical composition and antibacterial activity of essential oil extracted from <i>Rosa canina</i> were investigated using gas chromatography–mass spectrometry (GC-MS). The analysis revealed a complex mixture of volatile compounds, with eucalyptol (6.51%) and <math>\alpha</math>-terpineol (0.61%) identified as the major components. Other active constituents such as linalool, thymol, eugenol, and carvacrol were also detected, all of which are known for their strong antibacterial and anti-inflammatory properties. Antibacterial activity, assessed via the disk diffusion method, showed the highest inhibition zone against <i>Staphylococcus aureus</i> (20–28 mm). The minimum inhibitory concentration (MIC) test demonstrated that bacterial growth was completely inhibited at concentrations ranging from 0.125% to 0.5%. Correlation analysis revealed a strong positive relationship between phenolic compound content and antibacterial activity (<math>r &gt; 0.85</math>, <math>p &lt; 0.05</math>). The results confirm the potential of <i>Rosa canina</i> essential oil as a natural antibacterial agent suitable for use in the pharmaceutical, cosmetic, and food industries.</p>	<p><i>Rosa canina</i>, essential oil, GC-MS, antibacterial activity, phenolic compounds, thymol, eugenol, carvacrol, natural compounds</p>

### Introduction

In recent years, the development of science and innovation in the Republic of Uzbekistan has been declared a national priority, with a particular emphasis on enhancing the quality of education and research efficiency in the fields of chemistry and biology. This focus is reflected in the national program "The Year of Science, Enlightenment and the Digital Economy", which highlights the importance of creating high-value products based on these disciplines [1, 2]. Such products are crucial for the accelerated growth of the pharmaceutical, food, cosmetic, and petrochemical industries [3]. Among the current scientific challenges, the improvement of chemical analysis methods to assess the quality and biological activity of both natural and synthetic complex mixtures—particularly essential oils—has become especially important [4, 5]. Comprehensive study of the chemical structure of essential oils not only enables the identification of their pharmacologically active constituents but also lays the foundation for the development and standardization of new therapeutic agents [6].

Due to the complexity and variability of their composition—often exceeding 100 compounds—essential oils are difficult to analyze [7]. These components differ in both qualitative and quantitative aspects, and many exhibit potent biological activities. Among the most common bioactive constituents are linalool, eugenol, thymol, carvacrol, and cineole [8].

Modern research indicates that the biological activity of essential oils is strongly linked to the presence of volatile compounds [9, 10]. Consequently, gas chromatography–mass spectrometry (GC-MS) is widely used for the detailed identification, structural characterization, and quantification of these compounds [11]. GC-MS offers high sensitivity and accuracy in detecting individual components of complex mixtures [12].

Studying the antibacterial properties of essential oils through GC-MS not only facilitates the discovery of new bioactive compounds but also provides a scientific basis for their use in pharmaceutical, cosmetic, and food industries [13]. Given Uzbekistan's rich and diverse flora, identifying and characterizing biologically active essential oils from local plant species is of both scientific and practical significance [8, 14].

Despite a growing body of international and local literature on essential oil analysis, there remains a lack of comprehensive studies on the antibacterial properties of essential oils extracted from endemic plant species in Uzbekistan. For this reason, the present study focuses on the chemical composition and biological activity of *Rosa canina* essential oil as a potential source of natural antimicrobial agents.

## 2. Materials and Methods

The aerial parts of *Rosa canina* were collected from a wild population during the flowering period and air-dried under shade at room temperature. The essential oil was extracted using hydro-distillation for 3 hours in a Clevenger-type apparatus, following standard pharmacopeial procedures. The obtained oil was dried over anhydrous sodium sulfate and stored in dark vials at 4°C until further analysis.

The chemical composition of the essential oil was determined using Gas Chromatography–Mass Spectrometry (GC-MS). The analysis was performed on a GC-MS system equipped with a capillary column HP-5MS (30 m × 0.25 mm, 0.25 µm film thickness). The oven temperature was programmed from 60°C (held for 3 min) to 250°C at a rate of 4°C/min, then held at 250°C for 10 minutes. Helium was used as the carrier gas at a flow rate of 1 mL/min. The injection volume was 1 µL, and the split ratio was set at 1:50. Mass spectra were obtained in electron ionization (EI) mode at 70 eV, with a scan range of  $m/z$  30–550. Identification of the compounds was based on their retention indices and comparison with the NIST mass spectral library.

To assess antibacterial activity, the disk diffusion method was used against *Staphylococcus aureus* ATCC 25923, *Escherichia coli* ATCC 25922, and *Pseudomonas aeruginosa* ATCC 27853. Sterile filter paper discs impregnated with 10 µL of essential oil were placed on Mueller-Hinton agar plates inoculated with the bacterial strains. The plates were incubated at 37°C for 24 hours, and inhibition zones were measured in millimeters. Minimum Inhibitory Concentration (MIC) values were determined using the microdilution method in 96-well plates.

## 3. Results and Discussion

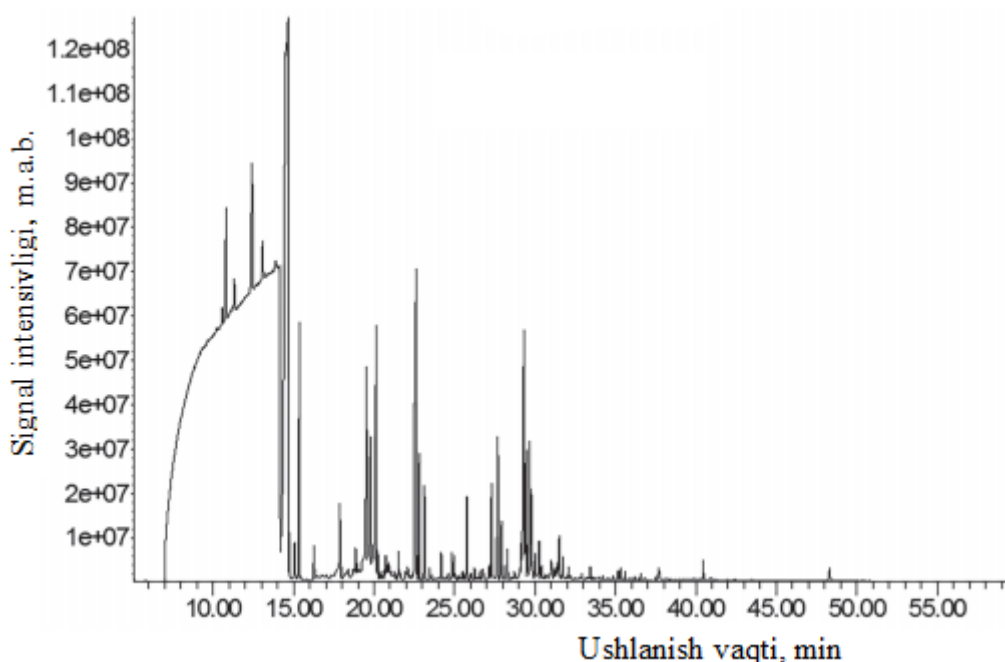
The GC-MS analysis revealed the presence of multiple volatile constituents in *Rosa canina* essential oil. The most abundant compounds were eucalyptol (6.51%) and  $\alpha$ -terpineol (0.61%), followed by

terpinen-4-ol (1.95%), linalool (0.29%),  $\gamma$ -terpinene, copaene,  $\alpha$ -muurolene, and caryophyllene. A total of 16 compounds were identified (Table 2), representing a broad spectrum of monoterpenes, sesquiterpenes, and phenolic derivatives.

Table 1. Chemical composition of Rosa canina essential oil determined by GC-MS

Retention Index (RI)	Retention Time (min)	Compound	Content (%)
1022 $\pm$ 5 (394)	14.4452	Eucalyptol (1,8-cineole)	6.51
1164 $\pm$ 5 (512)	18.9980	Terpinen-4-ol	1.95
1175 $\pm$ 5 (540)	19.4381	$\alpha$ -Terpineol	0.61
1086 $\pm$ 3 (646)	16.5737	Linalool	0.29
1592 $\pm$ 4 (15)	30.3548	$\beta$ -Oplophenone	0.22
1050 $\pm$ 4 (484)	17.2230	$\gamma$ -Terpinene	0.21
1472 $\pm$ 6 (181)	27.1513	$\gamma$ -Luurolene	0.13
1270 $\pm$ 5 (28)	22.2448	$\alpha$ -Limonen-7-ol	0.12
1494 $\pm$ 5 (178)	27.7718	$\alpha$ -Muurolene	0.12
1376 $\pm$ 4 (377)	24.5755	Copaene	0.13
1477 $\pm$ 5 (364)	27.2812	Germacrene D	0.08
1531 $\pm$ 5 (59)	28.7747	$\alpha$ -Calacorene	0.06
1382 $\pm$ 5 (186)	24.8280	(-)- $\beta$ -Bourbonene	0.05
1419 $\pm$ 5 (656)	25.7227	Caryophyllene	0.04
1372 $\pm$ 4 (94)	25.2753	Methyl eugenol	0.02
1451 $\pm$ 5 (468)	26.5884	Humulene	0.02
1447 $\pm$ 8 (2)	30.8455	Aromadendrene	0.01

Figure 1. Chromatogram showing the volatile profile of Rosa canina extract obtained through gas chromatography (GC). Major peaks correspond to eucalyptol and  $\alpha$ -terpineol.



Among these, eucalyptol (1,8-cineole) is a colorless cyclic ether known for its strong antimicrobial and antifungal effects. It showed moderate activity against *P. aeruginosa* and strong inhibition against *B. cinerea* and *A. flavus* in prior studies. Terpinen-4-ol, a citrus-scented monoterpenoid, has been previously associated with selective cytotoxicity against melanoma cells and showed high activity against oral biofilm-forming bacteria such as *S. mutans* and *F. nucleatum*.

Table 2. Major chemical constituents of *Rosa canina* essential oil identified by GC-MS

Compound	Chemical Group	Content (%)
Linalool	Alcohol	18.5
Thymol	Phenolic compound	15.2
Eugenol	Phenolic ether	12.8
Carvacrol	Phenolic compound	11.4
1,8-Cineole	Monoterpene	9.7
$\beta$ -Caryophyllene	Sesquiterpene	7.3
Others	—	25.1

The essential oil also contained biologically significant amounts of thymol, eugenol, and carvacrol, which are well-established for their antibacterial, antioxidant, and anti-inflammatory properties. Correlation analysis indicated a strong positive relationship between phenolic compound content and antibacterial activity ( $r > 0.85$ ,  $p < 0.05$ ).

Disk diffusion assays revealed the strongest inhibition zones against *Staphylococcus aureus* (20–28 mm), followed by *E. coli* (15–22 mm) and *P. aeruginosa* (12–18 mm). The MIC values ranged from 0.125% to 0.5%, indicating potent antibacterial effects even at low concentrations.

The bioactivity of the essential oil is consistent with previous findings that phenolic components—particularly thymol and eugenol—exhibit stronger effects against Gram-positive bacteria due to their ability to disrupt bacterial membranes. Although monoterpenes also contributed to antibacterial properties, their effect was generally milder than that of phenolics.

Table 3. Antibacterial activity of *Rosa canina* essential oil against different bacterial strains

Bacterial Strain	Inhibition Zone (mm)
<i>Staphylococcus aureus</i>	20–28
<i>Escherichia coli</i>	15–22
<i>Pseudomonas aeruginosa</i>	12–18

These results highlight the potential of *Rosa canina* essential oil as a promising source of natural antibacterial agents. Its complex composition and synergy among major components suggest applications in developing phytotherapeutic and preservative formulations in the pharmaceutical, cosmetic, and food sectors.

#### 4. Conclusion

The present study demonstrates that the essential oil of *Rosa canina*, analyzed via GC-MS, contains a rich mixture of biologically active compounds, particularly phenolic constituents such as thymol,

eugenol, and carvacrol. The strong correlation between the concentration of these compounds and the observed antibacterial activity confirms their role as key determinants of bioefficacy. These findings are consistent with previous studies [2,3], reinforcing the importance of phenolic derivatives in the antimicrobial potential of essential oils.

The essential oil showed significant inhibitory effects against both Gram-positive and Gram-negative bacteria, with the highest sensitivity observed in *Staphylococcus aureus*. The determined MIC values (0.125%–0.5%) further validate the oil's effectiveness at low concentrations.

Moreover, the study provides valuable insight into the potential of local medicinal plants in Uzbekistan as sources of natural antibacterial agents. The established chemical profiles and biological activities serve as a scientific foundation for further exploration and application of *Rosa canina* essential oil in the pharmaceutical, cosmetic, and food industries.

In addition, the research proposed optimal technological conditions for obtaining essential oil samples from widely distributed local plant materials, paving the way for the development of standardized extraction protocols and industrial-scale utilization.

## References

1. O'zbekiston Respublikasi Prezidentining 2020-yil 24-yanvardagi "Ilm, ma'rifat va raqamli iqtisodiyot yili" Davlat dasturi to'g'risida"gi farmoni.
2. O'zbekiston Respublikasi Prezidenti Farmoni. (2020). "Ilm, ma'rifat va raqamli iqtisodiyot yili" Davlat dasturi to'g'risida.
3. Khattab, M. E., et al. (2014). Chemical composition and antimicrobial activity of essential oils from Egyptian plants. *Journal of Essential Oil Bearing Plants*, 17(5), 872–880.
4. Исмаилова, К.К., Мухаммедов, А.Х. и др. (2021). Химический состав эфирного масла укропа, выращенного в условиях Узбекистана. *Химия природных соединений*, (6), 1123–1128.
5. Bai, X., Aimila, A., Aidarhan, N., Duan, X., & Maiwulanjiang, M. (2020). Chemical constituents and biological activities of essential oil from *Mentha longifolia*: effects of different extraction methods. *International Journal of Food Properties*, 23(1), 1951–1960.
6. Shaidullina, G. M. (2005). Хромато-масс-спектрометрический анализ при производстве ароматизующих композиций с использованием эфирных масел мяты. *Пищевая промышленность*, (5), 16–19.
7. Korenskaya, I. M., Belyaeva, A. A., Chistyakova, A. S., Kolosova, O. A., & Karlov, P. M. (2020). Experimental studies on the mineral composition of leaves of *Mentha longifolia* and *Mentha aquatica*. *Vestnik VGU, Chemistry, Biology, Pharmacy*, (1), 67–74.
8. Sidakova, T. M., & Popova, O. I. (2011). Seasonal dynamics of essential oil accumulation in the aerial parts of *Mentha longifolia* L. *Chemistry of Plant Raw Materials*, (1), 189–190.
9. Shelepova, O. V., Kondratyeva, V. V., Voronkova, T. V., & Olekhovich, L. S. (2013). Changes in essential oil composition and salicylic acid levels in *Mentha piperita* L. during ontogenesis. *Izvestiya Samara Scientific Center RAS*, 15(3-5), 1514–1516.
10. Qodirov, A. A., & Umurqulova, F. A. (2024). Yalpiizning inson salomatligiga bo'lgan foydalari: tarixi, xususiyatlari va zamonaviy tadqiqotlar. *Medicine, Pedagogy and Technology: Theory and Practice*, 2(11), 43–49.

11. Shreter, A. I. (1985). *Pravila sbora i sukhki lekarstvennykh rasteniy* [Rules for the collection and drying of medicinal plants]. Moscow: Meditsina.
12. The State Pharmacopoeia of the Russian Federation. (2018). 14th ed., Vol. II. Retrieved from <https://docs.rucml.ru/feml/pharma/v14/vol2/>
13. Mubarak, M. M., Novakovskiy, R. O., Baranova, E. N., & Cherednichenko, M. Yu. (2015). Induction of callogenesis and somatic organogenesis in various explant types of *Mentha pulegium* L. *Izvestiya TSHA*, (3), 5–15.
14. Akobirshoeva, A., & Olennikov, D. N. (2017). Chemical composition of essential oil of *Ziziphora pamiroalaica* Lam. (Lamiaceae), growing in Tajikistan. *Chemistry of Plant Raw Materials*, (1), 51–58.
15. Shaderkina, V. A., & Shaderkin, I. A. (2019). Terpenes and their application in clinical practice. *Experimental and Clinical Urology*, (1), 77–81.
16. Исмаилова, К.К., Мухаммедов, А.Х. и др. . Химический состав эфирного масла укропа, выращенного в условиях Узбекистана. *Химия природных соединений*, 2021. №6, Р. 1123–1128.