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CHEMICAL COMPOSITION OF ESSENTIAL OILS OF ROOTS AND LEAVES OF *FERULA TADSHIKORUM* PIMENOV

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This study describes the results of a comparative analysis of the chemical composition of essential oils of roots and leaves of 3 samples of virginile plants *Ferula tadshikorum* of various years of life from natural places (12–14 and 24–27-year-old individuals, Babatag ridge of the Surkhandarya region) of growth and in culture (4 years of life, Tashkent Botanical Garden).

The chemical composition of essential oils obtained by hydrodistillation was relatively investigated by means of GC-MSD analysis.

The results obtained showed that the isolated chemicals differ and are related to the age of plants. For example, introduced plants had an almost similar qualitative composition of essential oils of leaves and roots, and in natural individuals, the leaves, irrespective of the age of the plant, contained the same main components, while the roots had a different compound composition. The main component of the essential oils of the introducents were myristicin and methanethioamide, N,N-dimethyl, and in individuals of natural habitats the sulfur-containing components were thiopropionamide; disulfide, methyl 1-(methylthio) propyl and 1-(1-Propen-1-yl)-2-(2-thiopent-3-yl) disulfide, and also Germacyclobutane, 1,1-dimethyl-.

According to the available literature, they have anti-cancer, antioxidant and antimicrobial activity, and in the future research should be aimed at studying their therapeutic abilities.

Keywords: essential oil, *Ferula tadshikorum*, introduction, leaves, natural conditions, root, sulfur-containing components.

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Introduction

In modern society, each ethnic group has centuries of experience in the use of, tinctures, decoctions, and alcohol solutions from local medicinal plants, used to prevent and treat many ailments and diseases, in particular, colds, neuropsychiatric, gastrointestinal, liver, kidney, genital disorders, cardiovascular system problems, etc. [1, 2].

One of the promising groups of plants for the search for new sources of biologically active substances of particular interest in the production of drugs, in particular, is the genus *Ferula* L. from the family of Apiaceae Lindl. [3].

H. Khalifaev et al. [4] first investigated the chemical composition of root essential oils in *Ferula kuhistanica* Korovin growing in three different soil conditions and collected from two regions of Tajikistan. The authors compared the results obtained on the composition of essential oils with the data available in the literature for other species of *Ferula.* As a result, they established that *Ferula kuhistanica* root essential oils were dominated by α-pinen, β-pinen, β-fellandren, and myrcene. According to literary sources, these components have been found in the essential oils of many *Ferula* species as the main components that are probably characteristic of this genus.

Z. Bohrgati, M. Iranshahi [5] collected data on the properties of secondary metabolites and their physiological activity *Ferula assa-foetida* L. and *F. gummosa* Boiss. These plants are widely used for the treatment of infectious diseases, including diarrhea, skin infections, intestinal parasites, influenza, etc. In many species of the genus *Ferula*

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L., potent metabolites were isolated, most of them showed various biological activity, especially antimicrobial activity. The authors emphasized the importance of drymane-type sesquiterpene coumarins of the genus *Ferula* with potential antiviral properties.

The toxicity of new disesquiterpene and five-desquiterpene coumarins from the roots of *Ferula pseudalliacea* Rech.f. and the effect of these compounds on seed germination of some weeds and crops was evaluated [6]. In addition, sanandajin, farnesiferol B and camolonol acetate showed the greatest activity against HeLa human cancer cells. The study results showed that disesquiterpene and sesquiterpene coumarins isolated from *F. pseudalliacea* root extract can be considered potent herbicides and anticancer chemoprevention agents.

V. Karimian et al. [7] investigated the chemical composition and antibacterial activity of *Ferula assa-foetida* L camede resin essential oils. The authors identified 3 types of essential oils, containing α -pinene, and (Z)-propenyl sec-butyl disulfide. The highest content of these substances was found in an essential oil of the tear type, that is, there is α-pinene 38.2%, and (Z)-propenyl sec-butyl disulfide 12.4% with high antibacterial, anti-biofilm and cytotoxic activities, followed by paste and mass types, respectively.

In traditional medicine of Iran, the local population often uses *Ferula cupularis* (Boiss.) Spalik et S.R. Downie (Apiaceae), native to the foothills of Mount Dena, but there have been no scientific studies proving its medicinal properties [8]. Consequently, for the first time, the chemical composition and antibacterial effect of essential oils of flowers, leaves and stems were studied. The essential oil of *F. cupularis* flowers contained 15 monoterpenic, 13 oxygenated monoterpenic and 2 sesquiterpenic hydrocarbons. The essential oil of the leaves contained 12 monoterpenes, 13 oxygenated monoterpenes, 2 sesquiterpenes, 6 oxygenated sesquiterpene hydrocarbons and 3 non-penoid components. The stem essential oil contained one monoterpene, 23 oxygenated monoterpene, 2 sesquiterpenes and 6 oxygenated sesquiterpene hydrocarbons. The results of the study revealed a strong antibacterial effect of *F. cupularis* essential oils, which may be associated with a high content of oxygenated monoterpenic hydrocarbons in them.

Studies on the chemical composition and biological features of essential oils of underground or aboveground organs of *Ferula tadshikorum* can be said to be almost absent. There is only one work in which underground organs have been studied on the chemical composition and biological activity of essential oils [9]. According to the authors, the essential oil was dominated by the sulfur-containing compounds (Z)-1-propenyl *sec*-butyl disulfide (37.3%), (E)-1-propenyl *sec*-butyl disulfide (29.9%), (E)-1-propenyl 1-(methylthio) propyl disulfide (16.8%), and propyl sec-butyl disulfide (4.8%). The antioxidant, antimicrobial and cytotoxic activity of the essential oil was also evaluated.

In early work, we reported the content of protein and different groups of polysaccharides in roots and leaves in 3 virginile individuals of various years of life *Ferula tadshikorum* [10].

In the present study, a chemical analysis of essential oils from leaves and roots was carried out in virginiles of different years of life of *Ferula tadshikorum*, both introduced in culture and from natural habitats.

Materials and Methods

Ferula tadshikorum is a perennial monocarpic plant; a flower shoot appears in the last year of life. The plant has been in a virginal age state for a long time. At this stage of development, the plant has a rosette of leaves. For our analysis, we took plants of virginal age, but of different years of life. For thus study virginile individuals *Ferula tadshikorum* plants were collected in April 2022: – individuals of the 4th year of life, grown under conditions of introduction in the Tashkent Botanical Garden; – medium-aged (12–14 years) and mature 24–27-year-old plants collected from the Babatag ridge of Surkhandarya region. The experiments were carried out three times. Fresh samples from the above-ground (leaves) and underground (roots) parts of various *Ferula tadshikorum* individuals were cut into small pieces and essential oils were isolated by hydrodistillation. Analysis of the obtained extracts was carried out on an Agilent 8890 GC gas chromatograph with fission and non-fission evaporators, which was used in conjunction with the Agilent GC-MSD series 5977V in SCAN mode and ionization by Electron impact (EI).

Parameters of gas chromatograph analysis. Analytical column HP-5ms Ultra Inert 30 m x 250 μm x 0.25 μm. Injection volume 1 μm. Flow division injection mode (20 : 1). Evaporator temperature 280 °C. Liner UI, without dividing the flow, with one narrowing, glass fiber Gasket with spraying Gilded, Ultra Inert with washer. Carrier gas Hydrogen, constant stream, 1.2 mL/min. The thermostat program was 60 °C for 3 minutes, then 5 °C/min to 170 °C for 5 minutes, then 10 °C/min to 310 °C, then held for 2 minutes. The temperature in the transport line was 280 °C.

MS conditions. Delay to eliminate solvent effects was 3.5 minutes. SCAN data collection mode. The gain as 1.00. The source temperature as 250 °C. The quadrupole temperature 150 °C. Retention time (RT) was determined.

Components were identified based on comparison of mass spectrum characteristic indices with NIST electronic library data [11].

The retention index of a sample component is a number, obtained by interpolation (usually logarithmic), relating the adjusted retention volume (time) or the retention factor of the sample component to the adjusted retention volumes (times) of two standards eluted before and after the peak of the sample component. In the Kováts index or Kováts retention index used in gas chromatography, *n*-alkanes serve as the standards and logarithmic interpolation is utilized:

$$I = 100\left[\frac{\log_{10} X_i - \log_{10} X_z}{\log_{10} X_{z+1} - \log_{10} X_z} + z\right]$$

where X refers to the adjusted retention volumes or times, z is the number of carbon atoms of the *n*-alkane eluting before and (z+1) is the number of carbon atoms of the *n*-alkane eluting after the peak of interest:

$$V'Rz < V'Ri < VR(z+1)$$

The Kováts (retention) index expresses the number of carbon atoms (multiplied by 100) of a hypothetical normal alkane which would have an adjusted retention volume (time) identical to that of the peak of interest when analysed under identical conditions. The Kováts retention index is always measured under isothermal conditions. In the case of temperature-programmed gas chromatography a similar value can be calculated utilizing direct numbers instead of their logarithm. Since both the numerator and denominator contain the difference of two values, here we can use the total retention volumes (times). Sometimes this value is called the linear retention index:

$$I^T = 100\left[\frac{t^T_{Ri} - t^T_{Rz}}{t^T_{R(z+1)} - t^T_{Rz}} + z\right]$$

where t^T_R refers to the total retention times (chart distances) measured under the conditions of temperature programming. The value of I^T will usually differ from the value of I measured for the same compound under isothermal conditions, using the same two phases [12].

Results and Discussion

Extracts of leaves and roots of virginile specimens of different years of life of *Ferula tadshikorum* were identified using GC-MSD analysis, which are presented in Tables 1–2.

The main substance in the roots and leaves of 4-year-old introducers is myristicin, respectively 67.98% and 47.04%. According to the literature, myristicin has antitumor properties, and is also found in the essential oils of *Petroselinum crispum* (Mill.) Fuss. It helped inhibit tumor formation in the lungs of animals [13].

In addition, in the roots of introducents, the highest content was also methanethioamide, N,N-dimethyl (20.95%). S. Mugendhiran, B.D. Sheeja [14] studied the chemical composition of *Erigeron canadensis* L. by gas chromatography-mass spectrometry and identified various substances. The main component was methanethioamide, N,N-dimethyl (46.57%). The authors recommend using this plant as an antioxidant, anti-inflammatory, antitumor and antibacterial agent.

In natural representatives of 12–14 years of life in the roots, the main substance consists of sulfur–containing components – thiopropionamide (83.86%) and (Z)-sec-butyl propenyl disulfide (7.24%), and in individuals 24–27 years of life in the same organs 1-(1-Propen-1-yl)-2-(2-thiopent-3-yl) disulfide (62.86%). According to X. Li et. al. [15], essential oils of *Ferula ovina* Boiss. seeds they contain thiopropionamide up to 1.85%.

Sulfur-containing compounds are known to occur in plants and often have bioactive properties.

1-(1-Propenyl)-2-(2-thiopentyl) disulfide, or commonly known as allicin, is a sulfur-containing compound found in various plants, particularly in the genus *Allium* L., which includes garlic (*Allium sativum* L.), onions (*Allium cepa* L.), and shallots (*Allium cepa* var. *aggregatum* G. Don). Allicin is responsible for the characteristic aroma and flavor associated with these plants. For example, certain organosulfur compounds found in plants of the genus *Allium* (such as garlic and onions) have potential anticancer, antioxidant, and antimicrobial activities [16].

Table 1. Chemical composition of essential oils of the roots of different virginile individuals *Ferula tadshikorum*

Retention time	Substances name	RI	percentage (%)
Introduced virginile individuals 4 years of life			
12.220	Disulfide, methyl 1-(methylthio) propyl	2286	1.19
12.364	**Methanethioamide, N,N-dimethyl-**	2329	**20.95**
13.687	**Myristicin**	2527	**67.98**
Total			**90.12**
Unidentified			**9.88**
Virginile individuals 12–14 years of life			
8.864	**(Z)-sec-Butyl propenyl disulfide**	1163	**7.24**
12.231	Disulfide, methyl 1-(methylthio)propyl	2286	2.20
12.425	**Thiopropionamide**	2349	**83.86**
Total			**93.3**
Unidentified			**6.7**
Virginile individuals 24–27 years of life			
12.223	Disulfide, methyl 1-(methylthio)propyl	2286	2.01
12.434	**1-(1-Propen-1-yl)-2-(2-thiopent-3-yl)disulfide**	2324	**62.86**
12.496	Methyl cis-3-chloropropenoate	2398	22.2
12.514	2-Methyl-2,3-epoxy-2,3-dihydro-1,4-naphthoquinone	2415	10.89
Total			**97.96**
Unidentified			**2.04**

Table 2. Chemical composition of essential oils of leaves of virginile individuals *Ferula tadshikorum*

Retention time	Substances name	RI	Percentage (%)
Introduced virginile individuals4 years of life			
12.320	8-Ethyl-4,5,6,7,9-pentathiadecane	1907	1.09
13.687	**Myristicin**	2527	**47.04**
18.111	Methyl cis-2-trimethylsilyl-cyclopropane-1-carboxylate	2671	1.42
Total			**49.55**
Unidentified			**50.45**
Virginile individuals 12-14 years of life			
12.221	**Disulfide, methyl 1-(methylthio)propyl**	2286	**3.69**
12.381	**Germacyclobutane, 1,1-dimethyl-**	2315	**83.01**
Total			**86.7**
Unidentified			**13.3**
Virginile individuals 24-27 years of life			
12.226	**Disulfide, methyl 1-(methylthio)propyl**	2286	**8.07**
12.375	**Germacyclobutane, 1,1-dimethyl-**	2315	**70.60**
13.609	1,3-Benzodioxole, 4-methoxy-6-(2-propenyl)-	2527	2.51
14.064	Benzene, 1,2,3-trimethoxy-5-(2-propenyl)-	2548	2.67
Total			**83.85**
Unidentified			**16.15**

In the leaves of these individuals, the highest content is in germacyclobutane, 1,1-dimethyl-, in middle-aged representatives up to 83.01%, and in mature – almost 70.60% (Table 2). This compound has unique properties, and its presence in plants is of interest among researchers. However, there is no information on the content of this component in plants in the literature, there are some data on its chemical synthesis [17–19]. Germanium is present in all living plant and animal matter in micro-trace quantities. Its therapeutic attributes include immuno-enhancement, oxygen enrichment, free radical scavenging, analgesia and heavy metal detoxification [20]. Germacyclobutane in plants is an interesting object of study, and its role in various aspects of plant physiology and biology requires further study. A deeper understanding of the functions and mechanisms of action of germacyclobutane may open up new possibilities for the application of this compound in various branches of medicine, agriculture and industry.

It can also be noted that in the leaves of mature virginile plants, compared to the rest of the components, a greater amount is a sulfur-containing substance – disulfide, methyl 1-(methylthio) propyl (8.07%).

Conclusions

According to the results of the experiments, it can be concluded that isolated chemicals from essential oils of roots and leaves of 3 samples of virginile plants of various years of life and places of growth of *Ferula tadshikorum* have a number of therapeutic features. The main component of the essential oils of the roots and leaves of the introducents was myristicin and methanethioamide, N, N-dimethyl, and in individuals of natural habitats in the roots, sulfur-containing components - thiopropionamide; disulfide, methyl 1-(methylthio)propyl и 1-(1-Propen-1-yl)-2-(2-thiopent-3-yl)disulfide. All of the above chemicals have anticancer, antioxidant and antimicrobial activity, but in the future, more in-depth research is needed on the biological properties of this plant. In addition, the main component found in plant leaves from the natural habitat is from the class of amines – Germacyclobutane, 1,1-dimethyl-, which may be of interest among researchers for future work on its pharmaceutical features.

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Conflict of Interest
The authors of this work declare that they have no conflicts of interest.

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