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INVESTIGATION OF FATTY ACID COMPOSITION OF HERB AND ROOTS OF FETID MEADOW RUE (*THALICTRUM FOETIDUM* L.)

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Abstract. The manuscript presents the results of the determination of fatty acids in the herb and roots of fetid meadow rue. Investigation has been performed by the gas chromatography-mass spectrometry (GC/MS) that is based on the getting of fatty acid methyl esters and their further analysis. Identification of fatty acid methyl esters was carried out by the comparison of the retention times of standard mixture of fatty acid methyl esters of bacteria (Supelco, USA) and by the usage of mass spectral library NIST 02.

From the results of the chromatographic analysis 34 lipophilic compounds were identified in the herb of fetid meadow rue and 44 lipophilic compounds were identified in the roots of this plant. It was found that Hexadecanoic acid, methyl ester (palmitic acid, methyl ester) and 9,12,15-Octadecatrienoic acid, methyl ester (linolenic acid, methyl ester) are abundant in the herb of fetid meadow rue in comparison with other identified fatty acids. Concentrations of these acids are 3.582 mg/g and 3.830 mg/g, respectively. During the analysis of fatty acid composition of the meadow rue it was established that its composition is more varied although identified compounds are contained in much less quantities. Among the identified fatty acids 9,12-Octadecadienoic acid (Z,Z)-, methyl ester (linolenic acid, methyl ester) (1.066 mg/g), 9-Octadecenoic acid (Z)- methyl ester (oleic acid, methyl ester) (0.842 mg/g), Hexadecanoic acid, methyl ester (palmitic acid, methyl ester) (0.801 mg/g) are found in higher quantity

Keywords: fetid meadow rue, fatty acid composition, herb, roots, gas chromatography-mass spectrometry.

Introduction. Fetid Meadow Rue (*Thalictrum foetidum* L.) is a member of Ranunculaceae family. This is a perennial herbaceous plant with a height of 65 cm. Caules are leafy, thick, adenotrichous with foul smell. Leaves are compound, tri- or tetrapinnate, rounded-oval. Flowers in loose panicles are small. The fruitlets are ovate or ovate-oblong. The plant grows in plains, mountain meadows of Central Asia. This and other species of plants are very popular in modern traditional medicine of the countries of Central Asia.

This plant is understudied from a chemical composition and pharmacological properties of view. In Tajik traditional medicine tea from the fetid meadow rue herb is used for treatment of falling sickness, malaria, jaundice, edema, pulmonary tuberculosis, it can act as haemostatic at the epitaxises, female diseases and such as general tonic. Tea from the roots of fetid meadow rue is used for treatment of diarrhea, stomach ulcer, liver and kidney diseases. Soaking therapy based on the applications of fetid meadow rue herb is used in the cases of ulcer, wounds and at the rheumatic disease. Tea from the seeds of fetid meadow rue also relieves tormina, hypertensive disease, faintness, bronchitis [1, 2, 5].

Detail pharmacognostic investigation of herbal raw material with the extension of information data about the chemical composition of plant is important today taking into account widespread use of this plant in traditional medicine and its perspective as herbal raw material for the development of medicinal preparations.

Therefore, *the aim of our work* was investigation of fatty acid composition of herb and roots of fetid meadow rue.

Materials and Methods. Determination of fatty acid composition of herb and roots of fetid meadow rue has been performed by the gas chromatography-mass spectrometry (GC/MS) that is based on the getting of fatty acid methyl esters and their further analysis.

The chromatographic separation was performed on gas chromatograph/mass spectrometer system Agilent 6890N/5973inert (Agilent technologies, USA). All chromatographic separations were performed on capillary column HP-5ms (30m×0,25mm×0,25mkm, Agilent technologies, USA). Evaporator temperature was 250 °C, interface temperature was 280 °C.

Separation was performed in programmed temperature mode – program started at 60 °C for 4 min and changed to 250 °C at the rate of 4 °C/min and held for 6 min. The temperature was raised to 300 °C at the rate of 20 °C/min and held for 5 min. Sample injection was 1 µl. Injector was operated in a split mode with a split ratio of 1:20. MS scanning was performed from m/z 38-400. Flow rate of gas-carrier was 1.0 mL/min.

Herbal raw material was crushed to powder in a glass mortar for analysis. A weighed portion of herbal material was mixed in a glass vial with 2 mL of reaction mixture consisting of methanol:toluene:sulfuric acid (44:20:2 v/v) and solution of internal standard in 0.3 mL of heptane (it corresponds to 200 mg of sample). Test sample was kept at 80 °C for 2 hours on ultrasonic bath then it was cooled to room temperature and centrifugated at 5000 rpm for 10 min. 0.2 mL of upper hexane phase containing fatty acid methyl esters was separated.

Identification of fatty acid methyl esters was carried out by the comparison of the retention times of standard mixture of fatty acid methyl esters of bacteria (Supelco, USA) and by the usage of mass spectral library NIST 02. Assay was done by the internal standard addition to the test sample. Undecanoic acid (C11:0) was used as internal standard [3, 4].

Results and Discussion. As follows from the analysis 34 lipophilic compounds were identified in the herb of fetid meadow rue (table 1), 44 lipophilic compounds were identified in the roots of this plant (table 2). The chromatograms of fatty acid methyl esters of the herb and roots of fetid meadow rue are given on the fig. 1 and fig. 2, respectively.

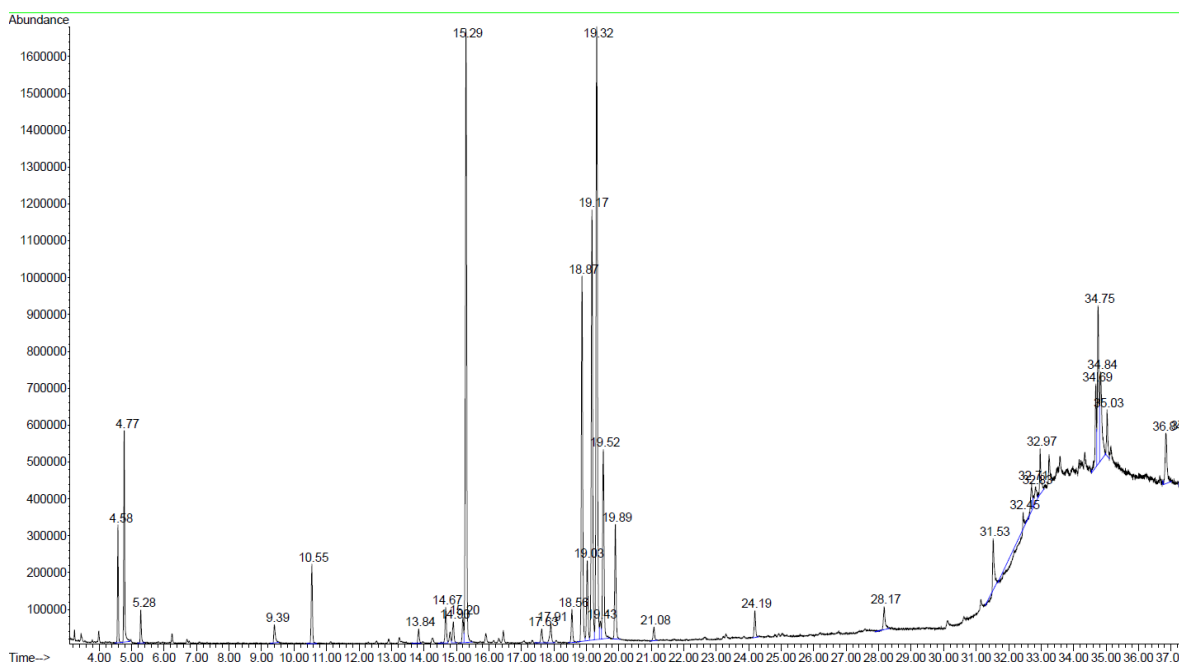


Fig. 1. Chromatogram of fatty acid methyl esters of the fetid meadow rue herb

It was found that Hexadecanoic acid, methyl ester (palmitic acid, methyl ester) and 9,12,15-Octadecatrienoic acid, methyl ester (linolenic acid, methyl ester) are abundant in the herb of fetid meadow rue in comparison with other identified fatty acids. Concentrations of these acids are 3.582 mg/g and 3.830 mg/g, respectively. Among the other fatty acids Citric acid, trimethyl ester (0.147 mg/g), 7-Hexadecenoic acid, methyl ester (palmitolic acid, methyl ester) (0.136 mg/g), Heptadecanoic acid, methyl ester (margaric acid, methyl ester) (0.088 mg/g), 6-Octadecenoic acid, methyl ester, (Z)- (Petroselinic acid) (0.111 mg/g), 5-Octadecenoic acid, methyl ester (1.203 mg/g), Octadecanoic acid, methyl ester (0.664 mg/g), Eicosanoic acid, methyl ester (arachic acid, methyl ester) (0.161 mg/g), Docosanoic acid, methyl ester (behenic acid) (0.180 mg/g), Tetracosanoic acid, methyl ester (lignoceric acid, methyl ester) (0.278 mg/g) were found. Valeric acid, 2,6-dimethylnon-1-en-3-yn-5-yl ester was also found (0.468 mg/g).

Table 1. Quantitative result of fatty acid composition of the fetid meadow rue herb

Peak number	Retention time	Compound	Content, mg/g
1	4.5797	Undecanoic acid, methyl ester	Internal Standard
2	4.7717	1-Propene-1,2,3-tricarboxylic acid, trimethyl ester, (E)-	0.871
3	5.2805	Citric acid, trimethyl ester	0.147
4	9.3956	2,6,6-Trimethyl-9-undecen-1-ol	0.116
5	10.5472	Methyl tetradecanoate	0.408
6	13.8366	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	0.074
7	14.6713	9,12,15-Octadecatrien-1-ol, (Z,Z,Z)-	0.210
8	14.9034	2-Cyclopenten-1-one, 2-pentyl-	0.131
9	15.1979	7-Hexadecenoic acid, methyl ester, (Z)-	0.136
10	15.2961	Hexadecanoic acid, methyl ester	3.582
11	17.626	Heptadecanoic acid, methyl ester	0.088
12	17.9072	1,1'-Bicyclopentyl-1,1'-diol	0.137
13	18.5543	Tetrahydropyran 12-tetradecyn-1-ol ether	0.192
14	18.8668	Cis-4-methyl-exo-tricyclo[5.2.1.0(2,6)]decane	2.106
15	19.0275	3.beta.-Methyl-trans-hexahydrophthalide	0.500
16	19.1703	9,12-Octadecadienoic acid, methyl ester	2.610
17	19.322	9,12,15-Octadecatrienoic acid, methyl ester, (Z,Z,Z)-	3.830
18	19.4292	6-Octadecenoic acid, methyl ester, (Z)-	0.111
19	19.5229	5-Octadecenoic acid, methyl ester	1.203
20	19.8933	Octadecanoic acid, methyl ester	0.664
21	21.0806	d-Ribose, 2-deoxy-bis(thioheptyl)-dithioacetal	0.089
22	24.187	Eicosanoic acid, methyl ester	0.161
23	28.1683	Docosanoic acid, methyl ester	0.180
24	31.5247	Tetracosanoic acid, methyl ester	0.278
25	32.4486	Cyclotrisiloxane, hexamethyl-	0.452
26	32.7075	Heptasiloxane, 1,1,3,3,5,5,7,7,9,9,11,11,13,13-tetradecamethyl-	0.200
27	32.8369	Methyltris(trimethylsiloxy)silane	0.175
28	32.9708	Heptasiloxane, 1,1,3,3,5,5,7,7,9,9,11,11,13,13-tetradecamethyl-	0.267
29	34.6847	Valeric acid, 2,6-dimethylnon-1-en-3-yn-5-yl ester	0.468
30	34.7561	Cyclohexane, 1,1'-(2-propyl-1,3-propanediyl)bis-	0.989
31	34.8365	Silane, trimethyl[5-methyl-2-(1-methylethyl)phenoxy]-	0.778
32	35.0329	Silicic acid, diethyl bis(trimethylsilyl) ester	0.236
33	36.8405	1,3-Bis(trimethylsilyl)benzene	0.421
34	37.4073	Hexestrol di-TMS	0.382

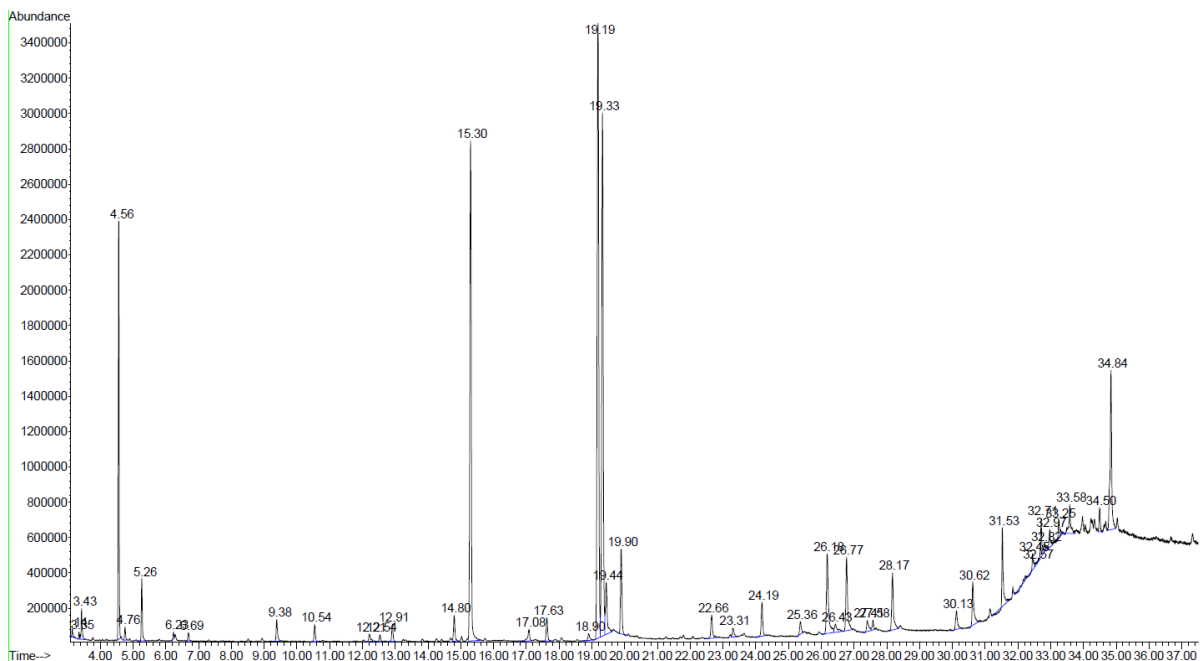


Fig. 2. Chromatogram of fatty acid methyl esters of the fetid meadow rue roots

Table 2. Quantitative result of fatty acid composition of the fetid meadow rue roots

Peak number	Retention time	Compound	Content, mg/g
1	3.1425	.alpha.-d-Ribopyranoside, methyl	0.007
2	3.3478	Decanoic acid, methyl ester	0.005
3	3.4281	Methyl 3,3-dimethoxypropionate	0.031
4	4.5618	Undecanoic acid, methyl ester	Internal Standard
5	4.7537	1-Propene-1,2,3-tricarboxylic acid, trimethyl ester	0.014
6	5.267	Citric acid, trimethyl ester	0.068
7	6.2355	Dodecanoic acid, methyl ester	0.005
8	6.6908	Nonanedioic acid, dimethyl ester	0.010
9	9.3866	Isoleucine, N-(trifluoroacetyl)-, sec-butyl ester, L-	0.031
10	10.5426	Methyl tetradecanoate	0.023
11	12.2119	Cyclohexan-1-ethanol, 1-hydroxymethyl-	0.014
12	12.5377	9-Dodecenoic acid, methyl ester, (E)-	0.010
13	12.9126	Pentadecanoic acid, methyl ester	0.024
14	14.8006	11-Hexadecenoic acid, methyl ester	0.040
15	15.296	Hexadecanoic acid, methyl ester	0.801
16	17.0814	9-Octadecenoic acid (Z)-, methyl ester	0.024
17	17.6259	Heptadecanoic acid, methyl ester	0.036
18	18.9024	13-Tetradecen-1-ol acetate	0.013
19	19.1881	9,12-Octadecadienoic acid (Z,Z)-, methyl ester	1.066
20	19.3264	9-Octadecenoic acid (Z)-, methyl ester	0.842
21	19.438	9-Octadecenoic acid (Z)-, methyl ester	0.098
22	19.8932	Octadecanoic acid, methyl ester	0.128
23	22.656	Tridecanedioic acid, dimethyl ester	0.043
24	23.3077	Cyclohexane, 1-(1,5-dimethylhexyl)-4-(4-methylpentyl)-	0.017
25	24.1914	Eicosanoic acid, methyl ester	0.055
26	25.3653	Cyclodecanol	0.021
27	26.1821	13-Octadecenal, (Z)-	0.172
28	26.432	Glycyl-dl-alanine	0.024
29	26.7712	Nonadecanedioic acid, dimethyl ester	0.135
30	27.405	Cyclopentadecane	0.025
31	27.5791	Octadecane	0.016
32	28.1727	Docosanoic acid, methyl ester	0.108
33	30.1276	Tricosanoic acid, methyl ester	0.036
34	30.623	Eicosanebioic acid, dimethyl ester	0.079
35	31.5246	Tetracosanoic acid, methyl ester	0.097
36	32.4485	Cyclotrisiloxane, hexamethyl-	0.023
37	32.569	Heptasiloxane, 1,1,3,3,5,5,7,7,9,9,11,11,13,13-tetradecamethyl-	0.004
38	32.7074	1H-Indole, 1-methyl-2-phenyl-	0.057
39	32.819	Cyclotrisiloxane, hexamethyl-	0.011
40	32.9707	Cyclotrisiloxane, hexamethyl-	0.028
41	33.2519	2-(Acetoxymethyl)-3-(methoxycarbonyl)biphenylene	0.030
42	33.5822	Silane, trimethyl[5-methyl-2-(1-methylethyl)phenoxy]-	0.056
43	34.4972	Tetrasiloxane, decamethyl-	0.031
44	34.8409	N-Benzyl-N-ethyl-p-isopropylbenzamide	0.274

From the results of the analytical characterizations of the fatty acids from the *Thalictrum foetidum* L. roots it is found that its composition is more varied although identified compounds are contained in much less quantities.

Among the identified fatty acids 9,12-Octadecadienoic acid (Z,Z)-, methyl ester (linolenic acid, methyl ester) (1.066 mg/g), 9-Octadecenoic acid (Z)- methyl ester (oleic acid, methyl ester) (0.842 mg/g), Hexadecanoic acid, methyl ester (palmitic acid, methyl ester) (0.801 mg/g) are found in higher quantity. Besides, the content of the following acids was established: Citric acid, trimethyl ester (0.068 mg/g), Decanoic acid, methyl ester (0.005 mg/g), Dodecanoic acid, methyl ester (0.005 mg/g), Nonanedioic acid, dimethyl ester (0.010 mg/g), 9-Dodecenoic acid, methyl ester, (E)- (0.010 mg/g), Pentadecanoic acid, methyl ester (0.024 mg/g), 11-Hexadecenoic acid, methyl ester (0.040 mg/g), 9-Octadecenoic acid (Z)-, methyl ester (0.024 mg/g), Heptadecanoic acid, methyl ester (0.036 mg/g), 9-

Octadecenoic acid (Z)-, methyl ester (0.098 mg/g), Octadecanoic acid, methyl ester (0.128 mg/g), Tridecanedioic acid, dimethyl ester (0.043 mg/g), Nonadecanedioic acid, dimethyl ester (0.135 mg/g), Docosanoic acid, methyl ester (0.108 mg/g), Tricosanoic acid, methyl ester (0.036 mg/g), Eicosanebioic acid, dimethyl ester (0.079 mg/g), Tetracosanoic acid, methyl ester (0.097 mg/g).

Conclusions. As can be seen from the above, 34 lipophilic compounds were identified in the herb of fetid meadow rue and 44 lipophilic compounds were identified in the roots of this plant by the gas chromatography-mass spectrometry (GC/MS). Among the identified fatty acids Hexadecanoic acid, methyl ester (palmitic acid, methyl ester) and 9,12,15-Octadecatrienoic acid, methyl ester (linolenic acid, methyl ester) are found in the herb of the fetid meadow rue in higher quantities, their concentrations are 3.582 mg/g and 3.830 mg/g, respectively. 9,12-Octadecadienoic acid (Z,Z)-, methyl ester (linolenic acid, methyl ester) (1.066 mg/g), 9-Octadecenoic acid (Z)- methyl ester (oleic acid, methyl ester) (0.842 mg/g), Hexadecanoic acid, methyl ester (palmitic acid, methyl ester) (0.801 mg/g) are found in the higher quantities in the roots of fetid meadow rue.

Obtained experimental findings give additional information about the chemical composition of herbal raw material such as fetid meadow rue and it can be used in further complex pharmacological investigations.

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