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INVESTIGATION OF FATTY ACID COMPOSITION OF DEVIL'S CLAW ROOTS

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The manuscript presents the results of the determination of fatty acids composition in the roots of devil's claw.

Materials and methods. As the study object the devil's claw roots was used. Chromatographic separation was performed using gas chromatography-mass spectrometry system Agilent 6890N / 5973inert (Agilent Technologies, USA).

Results. The results investigation of fatty acid composition of devil's claw roots obtained 28 lipophilic compounds by the GC/MS. Among the identified fatty acids dominating is propionic, stearic and palmitic acid (2.08 ±0.01, 1.17 ±0.07 and 0.89 ±0.01, respectively).

Among the acids determinationed, the most important is octadecenoic acid, methyl ester (0.22±0.05), which is part of cell membranes, is involved in the metabolism and the synthesis of prostaglandins, which are necessary for cell growth and regeneration.

Conclusions. The results of the study of fatty acid composition of the devil's claw roots indicate the prospect of further research of devil's claw as an object for the development of new drugs.

Keywords: *Devil's claw, roots, fatty acid composition, gas chromatography-mass spectrometry*

Introduction

Devil's Claw (Pedaliaceae plant family, Harpagophytum procumbens,) – is a traditional African plant which has been used in folk-medicine during a long time for the treatment of various diseases especially arthragra and rheumatism [1].

Roots of this plant are used as medicines that have anti-inflammatory, antirheumatic, analgesic, sedative and diuretic activity. Harpagoside and harpagide are main biologically active substances found in H. procumbens, as well as chemical composition is represented with 8-p-coumaroylharpagide, 8-feruloylharpagide, 8-cinnamoylmyoporoside, pagoside, acteoside, isoacteoside, 6'-O-acetylacteoside, 2,6-diacetylacteoside, cinnamic acid, caffeic acid, procumbide, and procumboside [2, 3].

Pharmacognostic experiment of herbal raw material with the chemical composition data information of plant is important task and has widespread use of this plant in traditional medicine and its perspective as herbal raw material for the medicinals development.

Therefore, the aim of our work was investigation of fatty acid composition of devil's claw roots.

Methods

Determination of fatty acid composition of devil's claw roots 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.

Chromatographic separation was performed using gas chromatography-mass spectrometry system Agilent 6890N / 5973inert (Agilent technologies, USA). Column capillary HP-5ms (30m × 0,25 mm × 0,25 mkm, Agilent technologies, USA). Evaporator temperature is 250 °C, interface temperature 280 °C. Separation was carried out in programming mode temperature - the initial temperature of 60 °C was heated for 4 min., gradient from 4 °C / min to 250 °C. The final temperature was kept for 5 min. Sample volume of 1 µl was injected in the split mode flow 1:20. Detection was performed in SCAN mode in the range (38-400 m / z). The flow rate of carrier gas through the column at 1.0 ml / min [4].

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 mkg of sample). Test sample was kept at 80 0C 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. The quantitative analysis was done by the internal standard addition to the test sample. Undecanoic acid (C11:0) was used as internal standard [5, 6].

Results

As follows from the analysis 28 lipophilic compounds were identified in the roots of Devil's Claw (table 1). The chromatograms of fatty acid methyl esters of standard mixture and fatty acid methyl esters of the devil's claw roots of are given on the fig. 1 and fig. 2, respectively.

Discussion

As can be seen from the above, 28 lipophilic compounds were identified in the devil's claw roots by the GC/MS.

Among the identified fatty acids dominating is propionic, stearic and palmitic acid (2.08 ±0.01, 1.17 ±0.07 and 0.89 ±0.01, respectively).

Among the acids determinationed, the most important is octadecenoic acid, methyl ester (0.22±0.05), which is part of cell membranes, is involved in the metabolism and the synthesis of prostaglandins, which are necessary for cell growth and regeneration.

The results of the study of fatty acid composition of the devil's claw roots indicate the prospect of further research of devil's claw as an object for the development of new drugs.

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Figure 1. Chromatogram of fatty acid methyl esters of standard mixture

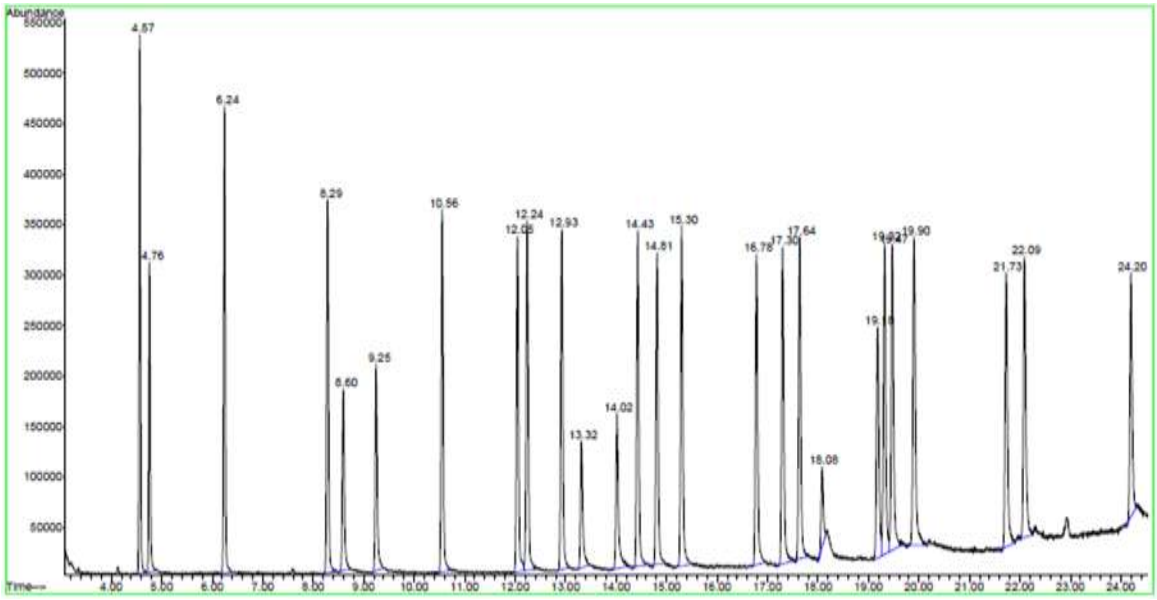

Figure 2. Chromatogram of fatty acid methyl esters of the Devil's claw roots

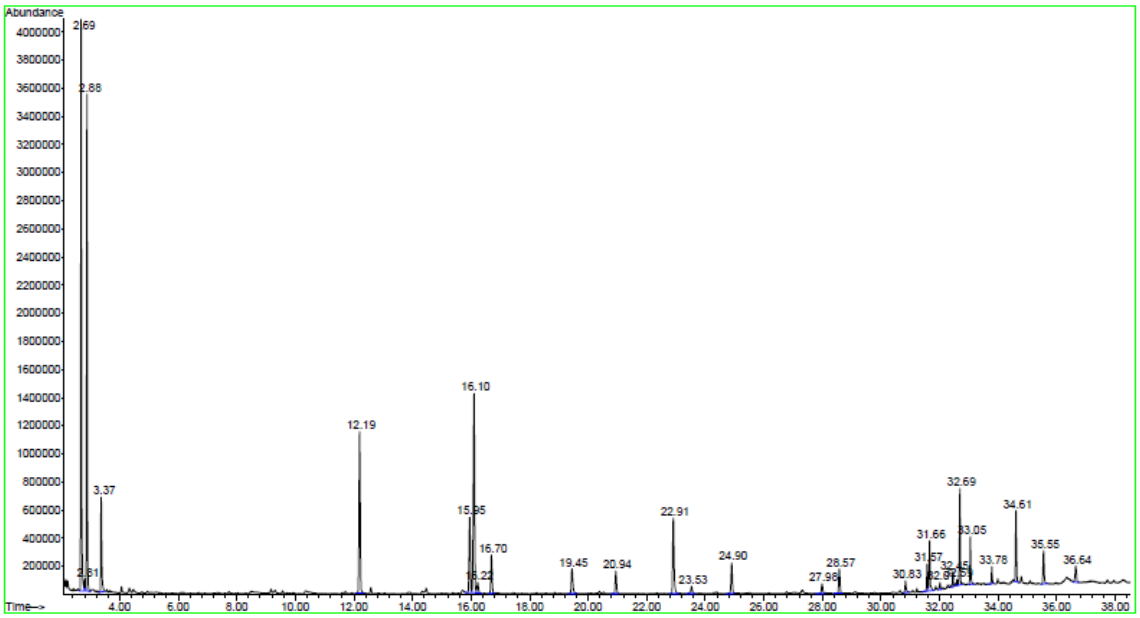

Table 1. Quantitative result of fatty acid composition of the Devil's claw roots (n=5)

Peak number	Retention time	Compound	Content, mg/g
1.	2.687	2-Propenoic acid, 3-phenyl-, methyl ester	2.08±0.01
2.	2.8164	Butanedioic acid, methoxy-, dimethyl ester	0.05±0.03
3.	2.8834	Undecanoic acid, methyl ester	Internal Standard
4.	3.3699	Citric acid, trimethyl ester	0.42±0.02
5.	12.1938	Hexadecanoic acid, methyl ester	0.89±0.01
6.	15.9519	9,12-Octadecadienoic acid, methyl ester	0.42±0.03
7.	16.0992	9-Octadecenoic acid (Z)-, methyl ester	1.17±0.07
8.	16.2242	9-Octadecenoic acid, methyl ester, (E)-	0.07±0.02
9.	16.6973	Octadecanoic acid, methyl ester	0.22±0.05
10.	19.4467	Tridecanedioic acid, dimethyl ester	0.17±0.03
11.	20.9419	Eicosanoic acid, methyl ester	0.13±0.06
12.	22.9058	Methyl 2-octylcyclopropene-1-octanoate	0.48±0.04
13.	23.5262	Octadecanedioic acid, dimethyl ester	0.05±0.02
14.	24.8964	Docosanoic acid, methyl ester	0.18±0.01
15.	27.985	Pentacosane	0.05±0.04
16.	28.5742	Tetracosanoic acid, methyl ester	0.14±0.01
17.	30.8326	1-Octadecene	0.06±0.03
18.	31.5691	hexacosanoic acid, methyl ester	0.11±0.03
19.	31.6538	Ledene oxide-(II)	0.24±0.02
20.	32.0109	1-Methylcholest-1,3,5(10)-trien-3-ol	0.03±0.01
21.	32.4528	Hexacosanoic acid	0.05±0.04
22.	32.5911	Octadecanedioic acid	0.02±0.03
23.	32.6893	Stigmastan-3,5-diene	0.35±0.05
24.	33.0464	montanic acid, methyl ester	0.18±0.04
25.	33.7784	nonacosylic acid, methyl ester	0.06±0.02
26.	34.6086	triacontanoic acid, methyl ester	0.30±0.01
27.	35.5459	hentriacontanoic acid, methyl ester	0.18±0.09
28.	36.6438	dotriacontanoic acid, methyl ester	0.10±0.01