



ISSN: 2277- 7695

TPI 2015; 3(11): 72-74

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www.thepharmajournal.com

Received: 11-11-2014

Accepted: 19-12-2014

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Chromato-mass-spectrometry identification of lipophilic compounds in plant extract

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Abstract

Lipophilic compounds have been investigated by the method of chromato-mass-spectrometry using plant samples of wild carrot seeds, extracted with ethyl acetate in the ratio of 1: 6. To identify them, an Agilent Technologies chromatograph was used, equipped with a series 6890 chromatographic column (with the inner diameter of 0, 25 mm and the length of 30 m) with a series 5973 mass spectrometer. The results of the research conducted demonstrated that the method of chromato-mass-spectrometry could be used to control the quality of wild carrot seeds by the content of lipophilic substances, as its major advantages include quite a wide range of application and the guarantee of the quality of the assay conducted.

Keywords: identification, lipophilic compounds, wild carrot, chromato-mass-spectrometry.

1. Introduction

Nowadays, an urgent issue in the field of pharmaceutical assays is the study of the ultimate chemical composition of plants, which contributes to the extension of the raw material base of useful plants and identifying new opportunities to use them.

We obtained dense and hydroalcoholic extracts of the herbal raw material of pumpkin and hop, which can be used both as finished products, and as substances for obtaining medical preparations and cosmetic aids (ointments, syrups, suppositories, creams, gels etc.).

It is common knowledge that the pharmacological activity of the medicinal herb raw material and phytooids is predetermined by biologically active substances, including lipophilic compounds [1]. Wild carrot (*Daucus carota L.*) is a biennial herbaceous medicinal plant of the family *Apiaceae*. In the first year of the wild carrot's life, a fusoid, hard, woody, dirty-whitish root with a basal rosette of pinnatisect leaves is formed, and in the second year, an up to 100 cm high caulis develops. The leaves in the contour are triangle, egg-shaped or elongated egg-shaped, up to 14-20 cm long, lower leaves are on petioles, the stem leaves are sessile, becoming smaller to the top. The flowers are small, forming a compound umbel. The fruits are hairy, two-seeded, fragrant [2-6].

Wild carrot is widespread, mainly in the Mediterranean, it can be found throughout Europe, Africa, and Australia. It also occurs in Ukraine, Moldova, the Caucasus and Central Asia. It grows near roads, in vegetable gardens, on forest skirts [2].

The purpose of our research is to identify the lipophilic compounds of wild carrot seeds using the method of chromato-mass-spectrometry, widely applied to determine the qualitative composition of a sample. Mass spectrometry along with gas chromatography is a valuable tool for identification and quantitative estimation of substances in plant samples [7].

2. Research Material and Techniques

Wild carrot seeds were tested. The dried and fine-cut medical herbal raw material was extracted with ethyl acetate in the ratio of 1: 6. The internal standard tridecane was added in terms of 50 µg of the substance per a certain quantity of a plant sample.

An Agilent Technologies chromatograph was used for tests, equipped with a series 6890 chromatographic column (with the inner diameter of 0, 25 mm and the length of 30 m) with a series 5973 mass spectrometer.

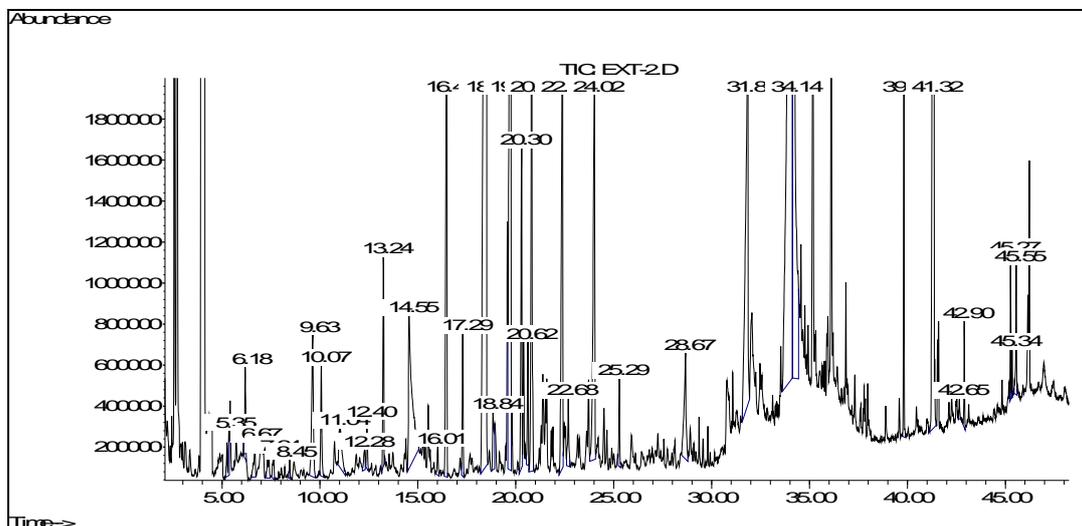
The thermostat temperature was set to 50 °C (1 min) and then to 320 °C with the velocity of 4 °C/min, the last temperature value was retained for 9 min. Helium was used as a carrier gas, the carrier gas velocity was 1, 2 ml/min.

The compounds were identified using Nist 05 and Wiley, 138 mass spectra libraries [5].

Wild carrot seeds contain coumarins (umbelliferone, esculetin, scopoletin, osthole), furanochromones (xanthotoxin, peucedanine), alkaloids, tannins, flavonoids, organic acids, sugars and over 20 microelements. The ether oil contains daucol, carotol and cineol [5,6].

The therapeutic application of wild carrot seeds includes

using them as anti-inflammatory, anthelmintic, antispasmodic, abortive, diuretic agents, for induction and normalization of menses [4]. The ether oil of wild carrot seeds is found in some cosmetic and medical phytooids, as it has a wound healing effect and improves the skin condition. These types of pharmacological activity are preconditioned, among other factors, by lipophilic compounds, analyzed in this paper.



Graph 1: Chromatographic profile of ethyl acetate extract of wild carrot seeds.

The value of the main components' content (> 0.1 % of the overall peak area), available from the experiment, are shown in Table 1.

Table 1: Chromatography-mass spectrometer identification of compounds in wild carrot seed samples

Sr. No.	Retention index, min	Compound	Component content, mg/kg
1	5.34	α -pinene	69.4
2	6.17	sabinene	85.9
3	6.67	myrcene	53.7
4	7.39	para-cymene	45.0
5	7.63	citriene	34.4
6	8.45	γ -terpinene	17.4
7	9.63	linalool	251.5
8	10.06	undecane	117.1
9	11.03	verbenol	124.0
10	12.27	α -terpineol	28.7
11	12.4	verbenone	52.8
12	13.24	dodecane	190.1
13	14.54	geraniol	538.7
14	16.01	2, 4-decadienal	31.6
16	17.29	α -terpinyl acetate	142.7
17	18.5	geranyl acetate	3273.1
18	18.84	coumarin	89.5
19	19.74	β -caryophyllene	1498.0
20	20.29	trans- α -bergamotene	340.2
21	20.62	humulene	110.9
22	20.81	β -farnesene	611.0
23	22.37	β -bisabolene	619.7
24	22.68	β -sesquiphellandrene	82.9
25	24.02	caryophyllene oxide	634.0
26	25.28	hexadecane	79.8
27	28.67	myristic acid	216.1
28	31.85	palmitic acid	936.5
29	34.12	linoleic acid	3730.1
30	34.13	oleic acid	2442.6
31	41.31	diterpene derivative of geraniol	1684.7
32	42.64	stigmasta-3, 5-diene	14.5
33	45.26	β -amyirin acetate	98.5
34	45.55	α - amyirin acetate	107.5

3. Research Findings and Discussion

Consequently, as is seen from Table 1, the chemical composition of wild carrot seeds is presented by 34 identified compounds: mono-, di-, triterpenes, fatty acids, hydrocarbons, alcohols etc.

Terpene compounds are of the greatest interest due to their antimicrobial, spasmolytic and anti-inflammatory properties. Terpenoids, geranyl acetate and β -caryophyllene, having the highest concentration in wild carrot seeds, are used, particularly, for formulation of perfume compositions, when obtaining flavours for soap, beauty products and in synthesis of some aromatic substances.

The hydrocarbon content in wild carrot seeds is quite low, with undecane and dodecane prevailing. Among alcohols, the concentration of geraniol in wild carrot seeds is the highest. Geraniol has a diuretic and hypotensive effect, and is used in cosmetic and fragrance industries, for perfuming soap and cleansing agents.

4. Conclusions

1. The findings of the research conducted showed that the method of chromat-mass-spectrometry can be applied to control the quality of wild carrot seeds by the content of lipophilic substances, since its major advantages are quite a wide range of application and the guaranteed quality of the assay conducted.
2. Based on the findings of the research conducted, it may be concluded that wild carrot seeds are promising raw material for extraction of terpene compounds and fatty acids in order to develop original medical, as well as cosmetic and preventive treatment preparations on its base.

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