CHEMICAL COMPOSITION OF THE VOLATILE OIL FROM DIFFERENT PLANT PARTS OF ANETHUM GRAVEOLENS L. (UMBELLIFERAE) CULTIVATED IN ROMANIA

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Abstract

The essential oil from dried leaves, flowers and fruits of *Anethum graveolens* L. (dill) cultivated in Romania was isolated by hydrodistillation and analysed by gaschromatography coupled with mass spectrometry (GC-MS). The main components in leaves were α - phellandrene (62.71%), limonene (13.28%) and anethofuran (16.42%). The main components in flowers were α - phellandrene (30.26%), limonene (33.22%) and anethofuran (22%). *Cis*-carvone and limonene are the major constituents of seeds volatile oil with 75.2% and, respectively 21.56%.

Rezumat

Uleiul volatil obținut din frunze, flori și fructe uscate de *Anethum graveolens* L. (mărar) cultivat în România, a fost izolat prin hidrodistilare și analizat prin cromatografie de gaze cuplată cu spectrometrie de masă (GC-MS). Principalii componenți din frunze au fost α -felandrenul (62,71%), limonenul (13,28%) și anetofuranul (16,42%). Principalii componenți din flori au fost α -felandrenul (30,26%), limonenul (33,22%) și anetofuranul (22%). Cis-carvona și limonenul sunt principalii compuși din uleiul volatil obținut din semințe, reprezentând 75,2% și, respectiv, 21,56%.

Keywords: Anethum graveolens, dill, essential oil, carvone, limonene.

Introduction

Anethum graveolens L. or dill, belonging to Apiaceae (Umbelliferae) family, is an annual aromatic herb known for culinary and medicinal use since ancient times. It is cultivated in the most parts of Europe and the United States of America. A variant called east Indian dill or sowa (Anethum sowa Roxb.) is cultivated in India, Egipt and Japan. The chemical composition of the essential oil of the two chimiotypes of dill and sowa differs mainly by the dillapiole content. The typical flavour of herb dill oil is due to α -phellandrene, limonene and dill ether (anethofuran). For flavouring purposes the herb oil with low content of carvone is preferred [1]. The dill seed oil contains a small quantity of dillapiole up to 3% when grown in tropical climate [1, 2]. In the east Indian dill (sowa) the content of

dillapiole ranges from 5 to 27% [3].

In recent years the scientific literature reports pharmacological effects of dill such as antibacterial [4, 5], antimycobacterial [6], antioxidant [7-10], cancer chemopreventive [11]. The well-known properties of dill from the traditional medicine, such as carminative, stomachic, diuretic have been reported [12, 13]. The dill essential oil has hypolipidemic activity and could be a cardioprotective agent [14]. Many studies showed that dill essential oil quantity and chemical composition varies depending on the plant parts and the developing stage of the plant at harvest time [13, 15-17]. The scientific literature data concerning chemical composition of volatile oil from different plant parts of *Anethum graveolens* L. are poor and differ from one author to the other [3, 13-15, 18].

The aim of this paper is to elucidate the chemical composition of the essential oil from leaves, flowers and fruits of dill cultivated in Romania. In order to study the complex chemical composition of volatile compound from plants, advanced analytical GC-MS techniques must be used, these allowing the identification of compounds even in minute quantities [23, 24].

Materials and methods

Reagents and solvents

All solvents and reagents were purchased from Merck, Darmstadt, Germany: dichloromethane, supraSolv for gas chromatography, anhydrous Na₂SO₄ granulated for organic trace analysis, the C₈-C₂₀ and C₂₁-C₄₀ *n*-alkanes used for the determination of Kovats retention indices.

Plant material

The raw material consisted of the leaves (harvested during the plant's flowering), flowers (on the blossom stage), and fruits (at their full maturity) of *Anethum graveolens* L., dill (*Apiaceae*) harvested in 2008 from Arges county (the southern part of Romania; 500 m altitude). The products were naturally dried in shadow and stored in controlled laboratory conditions.

Isolation of the essential oil

100 grams of fragmented dried vegetal products were hydrodistilled with 500 mL water in a Clevenger-type apparatus without organic solvent for 3h [25]. The essential oil was dried over anhydrous Na₂SO₄, stored in a dark glass bottle and kept at 4°C until analysis.

Gas chromatography-mass spectrometry

GC-MS analyses was performed on a Fisons Instrument GC 8000 equipped with an electron impact quadrupole, MD 800 mass spectrometer detector. The electron ionisation energy was 70 eV, ion-source temperature 200°C and the interface temperature 280°C.

A fused silica capillary column 5% phenyl-poly-dimethyl-siloxane (DB-5MS 30 m x 0.32 mm i.d. and 0.25 μ m film thickness, J&W Scientific) was used. The column temperature was programmed as follows: from 40°C (3 min hold) raised at 4°C/min to 250 °C and finally held at 250 °C for 10 min. A split-splitless injection (split ratio 1:30) at 280 °C was employed. The carrier gas (helium) flow rate was 2 mL/min. Two μ L of sample were injected. Data acquisition was performed with MassLab software for the mass range 30 - 600 u with a scan speed of 1 scan/s. The identification of compounds was performed by comparing their mass spectra with data from Adams [19], US National Institute of Standards and Technology (NIST, USA), WILEY 1996 Ed. mass spectra library and a personal library of 600 spectra. The identification of compounds was also based on the Kovats retention indices.

The Kovats retention indices were calculated using *n*-alkanes C_{8} - C_{20} and C_{21} - C_{40} and the experimental values were compared with those reported in literature [20, 21].

Results and discussion

The average content in essential oil of *Anethum graveolens* samples (3 determinations) was: 12 mL/kg for leaves, 32 mL/kg for flowers and 34 mL/kg for fruits (the results were calculated with reference to the dried material). These values are comparable with the results mentioned in the scientific literature about *Anethum graveolens* [3, 22].

In figure 1 are shown the chromatograms of essential oil from leaves (a), flowers (b) and fruits (c).



Table I shows the relative content of volatile compounds from essential oil, expressed as percentage from total area.

Table I.

Nr	Compounds	t _R /min*	Kovats retention indices	Area/ %		
				Leaves	Flowers	Fruits
1	α-Tujene	6.186	925	0.14	0.07	-
2	α-Pinene	6.328	930	1.66	0.90	0.02
3	Sabinene	7.721	974	0.12	0.05	-
4	β-Myrcene	8.472	994	0.12	0.06	-
5	α-Phellandrene	8.839	1004	62.71	30.26	0.12
6	<i>p</i> -Cymene	9.406	1024	1.11	0.65	-
7	Limonene	9.531	1028	13.28	33.22	21.56
8	p-Menth-3-en-2-one	14.394	1181	0.07	-	-
9	Dill Ether	14.536	1185	16.42	22.00	-
10	cis-Dihydrocarvone	15.088	1201	-	0.18	0.04
11	trans-Dihydrocarvone	15.154	1204	-	1.88	3.02
12	Carvone	16.481	1249	-	10.29	75.21
13	α-Copaene	22.752	1378	-	0.18	-
14	Cadinol	27.391	1667	-	0.09	-
15	γ-Muurolene	22.743	1477	0.29	-	-
16	p-Menth-1,8-dien-6-ol	24.003	1527	0.07	-	-
17	Neophytadiene	31.243	1838	0.42	-	-
18	<i>n</i> -Nonadecane	32.636	1900	0.07	-	-
19	<i>n</i> -Eicosane	34.696	2000	0.05	-	-
20	n-Heneicosane	36.632	2100	0.09	-	-
21	<i>n</i> -Docosane	38.484	2200	0.12	-	-
22	<i>n</i> -Tricosane	40.277	2300	0.19	-	-
23	<i>n</i> -Tetracosane	42.004	2400	0.19	-	-
24	<i>n</i> -Pentacosane	43.664	2500	0.19	-	-
25	<i>n</i> -Hexacosane	45.266	2600	0.16	-	-
26	<i>n</i> -Heptacosane	47.051	2700	0.08	-	-
Identified from total area				97.55	99.83	99.97

Chemical composition of essential oil from leaves, flowers and fruits of *Anethum graveolens* L.

In the essential oil from *Anethum graveolens* L. leaves 21 compounds were identified, adding up to 97.55% of the total area. Monoterpenic hydrocarbons were found predominant in the leaves oil representing 79.14% of the total content, where α -phellandrene (62.71%) constituted the major compound. This result is in agreement with that reported by Amin and Sleem [13]. The oxygenated compounds reached up to 16.56% with 16.42% dill ether (3,9-epoxy-1-p-menthen), which was the major compound. The *n*-alkanes from C₁₉-C₂₇were found in small quantities between 0.05 – 0.19% totalling 1.14%.

In the flower essential oil 13 compounds were identified adding up to 99.83% of the total area. The main components of the volatile oil from flowers are monoterpenic hydrocarbons: α -phellandrene (30.26%) and limonene (33.22%), total monoterpenic hydrocarbons representing 65.21%. In flowers essential oil the content of α -phellandrene is only 30.26% compared with 62.71% in the leaves oil. On the other hand, the content of limonene and dill ether in flowers oil is higher (33.22%) and respectively 22.00%) than the one determined in leaves oil.

In the leaves oil, the ketonic compound carvone was not present but in the flowers oil its content is 10.29%. In the essential oil of fruits, carvone is the main compound with 75.21%.

Another ketonic compound absent in the leaves essential oil was present in the flowers and fruits oils as *cis*- and *trans*-dihydrocarvone isomers. The fruit essential oil was rich in limonene 21.56% and *cis*- and *trans*-dihydrocarvone adding up to 3.06%. The amount of α -phellandrene in leaves oil and flowers oil was 62.71%, respectively 30.26%, and only 0.12% in fruits oil.

Conclusions

The chemical composition of dill volatile oil varies depending on the plant parts. In the leaves oil monoterpenic hydrocarbons are predominant, amounting to 79.14% (62.71% α -phellandrene and 13.28% limonene). In the flowers oil the content of α -phellandrene and limonene is 32.26% and 33.22%, respectively. Anethofuran (dill ether) is present in leaves and flowers with 16.42% and 22%, respectively, but is missing in the fruit oil. The main compound in fruits essential oil is carvone (75.21%), while the content of α -phellandrene is only 0.12% and limonene is 21.56%.

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