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Chemical composition of essential oil of two *Achillea* cultivars in Khorasan, Iran

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Abstract

Medicinal plants used in folk medicine are being increasingly studied and used on pharmaceutical, foodand nutraceutical fields.Medicinal plants mainly grow in plains, roadsides and mountainous regions. They are mostly used as wound healer due to tannins properties, besides having bitter and aromatic materials effects on the nervous system and heart. This paper presents a study on the chemical composition of essential oils of the flower of *A. millefolium*L. subsp and *A. eriophora*fromkhorasan, obtained by the hydrodistillation method and analyzed by using gas chromatography mass spectroscopy.Recent researches show that these plants have anti-cancer property. Forsampling, these plantsweretaken fromKhorasan, Iran,during the full flowering stage in July 2012. The plants' constituents were identified by their mass spectra and Kovats' indices Camphor, in which Fragranyl acetate, Thymol, Linalool and 1,8-Cineole were found to be the major constituents.

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Introduction

Achillea (Asteraceae) species are important medicinal plants commonly used in folk medicine against wounds, diarrhea, abdominal pains and cardiac diseases. The plants from this genus are also used due to their diuretic and insecticidalactivities (Baytop, 1999; Ezer and Arısan, 2006; Fujita *et al*., 1995;Honda *et al*., 1996; Sarper *et al*., 2009; Yesilada *et al*., 1993).Yarrow is one of the most important genera belonging to the citrus family (*Asteraceae*). The genus contains 19 species of herbaceous perennial, which are often aromatic (Rechinger, 1986).*A. millefolium*L. subsp. *millefolium*is an important species of the genus*Achillea*. This specie, in addition to drug use, is used as ornamental plant or covering in some parts of the world (Halevy, 1999).*A. eriophora* is one of *Achillea* species that is exclusive to Iranian plateau and is part of Endemic plants in Iran (Ghahreman, 1989; Ghahreman, 1998).

There are a number of interesting essential oils used in biological activities reported obtained from various Achillea species, for use as antifungal(Kordali *et al*., 2009; Ristic *et al*., 2004; Tuberoso *et al*., 2005;),antibacterial (Barel and Yashphe, 1989; Filippi *et al*., 2006; Simic*et al*., 2005; Skocibusic *et al*., 2004), antimicrobial (Barel *et al*., 1991;Baser, 2002a; Bezic *et al*., 2003; Candan *et al*., 2003; Demirci *et al*.,2009; Ghasemi *et al*., 2008; Senatore *et al*., 2005;Sökmen *et al*.,2003, 2004; Ünlü *et al*., 2002), genotoxicity (Sant'Anna *et al*., 2009),herbicidal (Kordali *et al*., 2009), insecticidal (Calmasur *et al*., 2006;Khani and Asghari, 2012; Tabanca *et al*., 2011; Tozlu *et al*., 2011)and antiradical (Candan *et al*., 2003; Demirci *et al*., 2009; Sökmen*et al*., 2004; Tuberoso *et al*., 2005) activities.

The present study aimed to investigate the chemicalcomposition of theessential oils of two Achillea species, namely *A. millefolium*L and *A. eriophora*growing in Khorasan, Iran.

Material and methods

Collection and preparation of the test plants

The aerial parts of *A. millefolium*L and *A. eriophora*werewere collected from different locations inKhorasan, Iranin July, 2012,during the flowering period.

Plant material

Samples of Achillea species were obtained from the KhorasanRazavi Agricultural Research Center of Iran. Thefresh plant samples were air dried in the shade for 7 days at environmentaltemperature (28-32°C day time). The dried parts were then powdered mechanically by using an electric blender, then sievedthrough a mesh size of 0.5 mm. The resulting fine powders were kept in tightly closed dry bags until use for the extractionof the essential oils.

Hydrodistillation

Powdered samples, 500 g each, from the tested plant specieswere subjected to hydrodistillation using a modified Clevengertypeapparatus to produce the plant oils. The extraction conditionwas as follows: 50 g powders; 500 ml distilled water, and 5 h distillation.

Anhydrous sodium sulphate was utilized to remove waterafter extraction. The oil yield (% w/w) was calculated on a dry weightbasis. The extracted oils were stored in a refrigerator at 4°C untilanalysis and testing.

GC/MS analysis

Agilent 7890 gas chromatograph with mass spectrometry detector 5975C inert MSD (Agilent Corporation, USA) was applied for sample analysis.

Chromatography conditions were set according to AOAC method No. 963/22 (27), and Capillary column of DB-35 MS, at length of 30 meters and diameter 0.25 mm, while thePolar Silica had thickness at 0.25 micrometers.The injector was set at 250°C, ion source 200°C as chromatography temperature parameters. The oven was programmed to operate at initial temperature of 80°C for 20s, 80°C to 240°C at 4°C

min⁻¹, and 240°C for 10 min. Helium gas was used as carrier gas, having column flow at 1.4 ml.min^{-1} and split rate at 1:30. 1.0 mm^{3}of the sample was injected. The energy for the EI source of the Agilent mass spectrometer was 70 eV. The mass unit was monitored to range from 30 to 450 m/z. Identification of components in the oil was based on retention indices, relative to n-alkanes and computer matching with the WILLEY 275.L library, as well as by making a comparison between the fragmentation patterns of mass spectra and those reported in the literature (Adams, 1995).

Results

Chemical composition of the tested plant oils

In the experiment, the essential oils, *A. millefolium*yielded 0.54% of chemical composition, while *A. eriophora* yielded 0.31%. Chemical composition analysis revealed that the essential oilof *A. millefolium*contained mainly Camphor(8.6%), Fragranol(7.3%), d-Limonene(5.8%), Camphene (5.6%), trans-Carveol(5.5%), Fragranyl acetate(5.3%) and Thymol(4.4%). A total of 36 compounds amounting to95% were identified from the essential oil of *A. eriophora*. Its major components were Camphor (9.9%), fragranyl acetate (8.7%), β-Pinene(8.6%), Thymol(7.5%), α-Terpineol(4.9%),β-Caryophyllene(4.8%) and α-Bisabolol (4.6%), as shown in Table 1.

Table 1. Compounds obtained from GC/MS analysis on A. millefoliumand A. eriophora essential oil

Compound	RI	*A. millefolium (%)*	*A. eriophora(%)*
2-Hexanal	856	3.1	-
Isopentyl acetate	878	0.1	-
Santolinatriene	910	0.3	0.1
α-Pinene	937	2.3	1.2
Camphene	954	5.6	1.4
Sabinene	976	2.7	5.2
β-Pinene	980	1.2	8.6
Myrcene	984	-	0.4
Yomogi alcohol	998	3.3	0.1
α-Terpinene	1017	0.7	0.4
P-Cymene	1026	1.9	1.1
d-Limonene	1030	-	0.3
1,8-Cineole	1034	5.8	3.9
γ-Terpinene	1049	-	0.4
cis-Sabinene hydrate	1055	0.2	0.1
Linalool	1088	4.1	4.4
cis-p-Ment-2-en-1-ol	1122	0.3	0.6
trans-p-Ment-2-en-1-ol	1141	2.4	-
β-Thujone	1143	3.4	3.6
Camphor	1145	8.6	9.9
Pinocarvone	1162	0.3	0.8
Borneol	1166	2.7	1.3
Terpinen-4-ol	1178	4.1	0.3
α-Terpineol	1190	2.8	4.9
Myrtenol	1194	-	0.1
Fragranol	1196	7.3	2.5
trans-Carveol	1223	5.5	2.4
cis-Chrysanthenyl acetate	1250	0.1	0.3
Bornyl acetate	1285	0.4	0.3
Thymol	1288	4.4	7.5
Fragranyl acetate	1334	5.3	8.7
Eugenol	1340	2.3	1.1
Geranyl acetate	1366	0.4	0.8
α-Copaene	1378	-	0.1
β-Caryophyllene	1415	2.2	4.8
α-Humulene	1452	-	1.1

Compound	RI	*A. millefolium (%)*	*A. eriophora(%)*
trans-β-Farnesene	1458	0.4	0.2
Germacrene D	1480	0.3	0.6
Bicyclogermacrene	1492	0.1	0.2
cis-Nerolidol	1538	0.4	0.9
Dendrolasin	1556	0.8	0.7
Spathulenol	1574	1.9	2.6
Caryophyllene oxide	1582	1.3	1.9
10-epi-γ-Eudesmol	1620	2.4	2.8
β-Eudesmol	1648	1.9	1.7
α-Bisabolol	1684	3.3	4.7
% Peaks identified		96.6	95
Total yield % (mL/100g)		0.54	0.31

The results from this study revealed that there is small variability between the chemical components of essential oils oftwo different species of Achilleain Iran.

Each organic compound from the oil samples was identified based on their retention time indices in the achieved gas chromatograph with reference to homologues series of n-alkanes. The obtained mass spectrum of each analytes was also compared with the mass fragmentation from library pattern number NIST 08. L database / ChemStation data system, and also was compared with other results from previous researches.

Table 1 presents the results of the analyses carried out in this study.

Conclusion

In this study, significant qualitative differences had been found in terms of chemical composition of the essential oils obtained from Achilleaspecies, as presented in Table 1.

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