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Study of isolated constituents of *Artemisia Persica* Boiss from isfahan area by nano scale injection

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Abstract: Essential oil from aerial parts of *Artemisia persica* Boiss. was obtained by hydro-distillation to produce oil in the yield of 0.47% (w/w). The oil was analyzed by capillary gas chromatography, using mass spectrometric detection. The amount of the samples injected by nano scale included were 1.0 nL (diluted 1.0 µL of sample in 1000 ml of *n*-pentane, v/v). Twenty two bioactive and flavour molecules were identified in the oil of *Artemisia persica* with cis-Ocimenone (30.8%), trans-Ascaridol (26.3%), α-Terpinene (6.4%), cis-ß-Terpineol (6.3%), ß–Phellanderene (6.3%), and para-Cymene (5.5%) as main constituents.

Keywords: *Artemisia persica*, essential oil composition, nano scale injection, cis-Ocimenone, trans-Ascaridol, α-Terpinene, cis-ß -Terpineol, ß –Phellanderene, and para-Cymene.

Introduction

Plants have always an important role to play in medicine and public health. The knowledge of use of medicinal plants and their properties was acquired by means of trial and error and transmitted from generation to generation [1]. Essential oils are secondary plant metabolites found in leaves, stems, flowers and fruits. They frequently have different chemical composition depending on the nature of the plant and the season. The family Asteraceae or Compositae, to which *Artemisia* belongs, is known as the aster, daisy, or sunflower family. It is the largest family of flowering plants in terms of number of species. Asteraceae is a taxon of dicotyledonous flowering plants [2-3]. *Artemisia* is a large, diverse genus of mostly perennial and aromatic herbs and shrubs in the daisy family Asteraceae, characterized by alternate leaves and small flower heads. Many of the perhaps 400 species in the genus are valued for their essential oils or as ornamentals [4]. *Artemisia* plants are valued for medicinal, ornamental, culinary, and insect-repelling purposes [5,6].

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Previously, the essential oil of *Artemisia persica* from Isfahan was investigated *via* micro scale ingection [7]. To the best of our knowledge, the essential oil of the aerial parts of *Artemisia persica* in Isfahan area (Kashan) has not been considered before. The maters on hand of this study were the determination of the percentage bioactive and fragrant molecules by nano scale injection. It is clear that, in some plants such as *Artemisia persica*, the amount of essential oil is trace (less than 1 µL). Thus by this method, (dissolving in a solvent) we can inject the dissolved essential oil in G.C. or G.C./M.S. and find out the components.

Results and discussion

Air-dried aerial parts of the plant were subjected to hydrodistillation using a Clevenger-type apparatus to produce oil in the yield of 0.47% (w/w). The oil was analyzed by GC and GC/MS. Twenty two, flavour and fragrance molecules, constituting 99.4% of the total components detected, were identified in this plant and listed in Table **1** with their percentage. Constituents are listed in order of their elution from HP-5MS column. The oil was characterized by a high content of cis-Ocimenone (30.8%), trans-Ascaridol (26.3%), α-

Terpinene (6.4%), cis-ß-Terpineol (6.3%), ß – Phellanderene (6.3%), and para-Cymene (5.5%).

Table 1. Bioactive and fragrance components of the aerial parts of A. persica

Compound[a]	A, %	RI [b]	Compound[a]	A, %	RI [b]
α-Pinene	0.4	935	Menthone	0.8	1150
ß - Pinene	0.5	969	cis- Chrysanthenol	0.6	1160
Sabinene	1.7	972	Terpinene-4-ol	1.9	1172
ß -Myrcene	0.7	990	α -Terpineol	3.2	1186
α -Terpinene	6.4	1016	Citronellol	0.2	1224
para-Cymene	5.5	1024	cis-Ocimenone	30.8	1225
ß -Phellanderene	6.3	1027	Cis-Carveol	0.8	1226
1,8-Cineol	1.7	1031	Piperitone	2.5	1250
Cis- ß- Ocimene	0.2	1036	Cis-Chrysanthenyl acetate	0.9	1264
γ-Terpinene	0.4	1058	iso-Ascaridol	26.3	1301
cis-ß-Terpineol	6.3	1142	**Total identified**	**99. 4**	
Isopulegol	1.3	1148			

[a]Compounds listed in order of their RI.
[b]RI (retention index) measured relative to n-alkanes (c_8-c_{32}) on the non-polar HP-5MS column.
[c]Relative percentage obtained from peak area.

In this paper, we illustrate biological properties and application of two important components from *A. persica* essential oils:

cis-Ocimenone: is a natural organic compound classified as a monoterpene and used in the perfume industry.

iso-Ascaridol: is a natural organic compound classified as a bicyclic monoterpene. It is the primary constituent of the oil of Mexican Tea [8]. It is a colorless liquid that is soluble in most organic solvents; ascaridole has been used as an anthelmintic for controlling nematodes [9]. Literature survey revealed two reports for chemical composition of the essential oils of this plant. The essential oils from aerial parts, leaves, flowers and roots of *A. persica* Boiss. from Iran was previously investigated by Mirjalili, B.F *et al.*, [10] The oils were obtained by hydrodistillation and the composition of the oils were analyzed by a combination of GC and GC/MS. The oils obtained from aerial parts were rich in (Z)-ocimenone (39.6%), ascaridole (16.0%) and α-terpinene (10.0%), which are good agreement with our results in this paper. The major components of the leaves oil were cis-sabinene hydrate (38.8%) and terpinolene (13.3%). The flowers oil was characterized by higher amounts of cis-sabinene hydrate (41.2%) and ethyl 2-nonynoate (24.4%). ß-Cedren-9-one (76.7%) was the predominant compound in the roots oil.In other study davanone was reported as the main constituent of *A. persica* steam-distilled essential oil, which was not detected in our oil result [11].

Experimental

Plant Material: Aerial parts of *A. persica* were collected in May 2009 in the center region of iran (around the Isfahan area). The voucher specimens of the plant were deposited in the herbarium of Research Institute of Forests and Rangelands, Isfahan, Iran.

Isolation of the Essential Oils: Dried aerial parts (100g) of *A. persica* were subjected to separate hydrodistillation for 3.5 h using a Clevenger-type apparatus [12]. After decanting and drying over anhydrous sodium sulfate, the sample oil which was light yellow in color, recovered from the aerial parts in yield of 0.47% (w/w).

Gas Chromatography (GC): GC analysis of the oil was performed on an Agilent HP-6890 gas chromatograph equipped with flame ionization detector (FID) and an HP-5MS capillary column (30 m × 0.25 mm i.d., film thickness, 0.25 µm). The oven temperature was held at 60˚c for 3 min and then programmed to 250˚c at a rate of 3˚c/min. Injector and detector temperatures were maintained at 220ºC and 290ºC, respectively. The amount of the sample injected was 1.0 nL (diluted 1.0 µL of sample in 1000 ml of *n*-pentane, v/v) in the splitless mode. Helium was used as carrier gas with a flow rate of 1 mL min^{-1}.

Gas Chromatography-Mass Spectrometry (GC/MS): GC-MS analysis of the oil was performed on a Agilent HP-5973 mass selective detector coupled with a Agilent HP-6890 gas chromatograph, equipped with a cross-linked 5% PH ME siloxane HP-5MS capillary column (30 m × 0.25 mm i.d, film thickness, 0.25 **µ**m)

and operating under the same conditions as above was described. The flow rate of helium as carrier gas was 1 mL min^{-1}. The MS operating parameters were as follows: ionization potential, 70 eV; ionization current, 2 A; ion source temperature, 200ºC; resolution, 1000.

Identification of components: Essential oil was analyzed by GC and GC/MS systems using a non-polar column and identification of components in the oil was based on retention indices (RI) relative to *n*-alkanes and computer matching with the WILEY 275.L library, as well as by comparison of the fragmentation pattern of the mass spectra with data published in the literature [13, 14]. The percentage composition of the sample was computed from the GC-FID peak areas without the use of correction factors.

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