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Short Communication

Variation in the essential oil composition of *Artemisia annua* L. of different growth stages cultivated in Iran

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Variation in the quantity and quality of the essential oil of *Artemisia annua L.* (Asteraceae) at different developmental growth stages including pre-flowering, flowering and post-flowering, are reported. The oils were obtained by hydro distillation of the air-dried samples. The yields of oils (w/w %) in different stages were in the order of: pre-flowering (0.97%), flowering (1.23%) and flowering (0.87%). The oils were analyzed by gas chromatography-mass spectrometry. In total, 32, 35 and 33 constituents were identified and quantified in the oil of pre-flowering, flowering and post-flowering plants, representing 97.67, 92 and 92.4% of the oils, respectively. Camphor, 1,8-cineole, camphene, spathunelol, α–pinene and artemisia ketone were the main compounds in all samples. Monoterpenes were the main group of compounds in pre-flowering (69.96%), flowering (72.44%) and post flowering (70.96%) stages.

Key words: *Artemisia annua L.*, essential oil, GC/MS, variation.

INTRODUCTION

Artemisia is one of the largest genera of the Asteraceae family (Bertea et al., 2005). This genus belongs to a useful group of aromatic and medical plants comprising about 300 species which are distributed throughout the world (Bertea et al., 2005). There are approximately 34 native *Artemisia* spp. in Iran (Mozaffarian, 1996).

Artemisia annua L. (Asteraceae) called annual or sweet wormwood or Qinghao is an annual herb native of Asia and has been used for many centuries in the treatment of fever and malaria (Brown et al., 2003). *A. annua* L. is a source of both essential oil (1.4 – 4.0 %) depending on chemotype, and other substances such as sesquiterpene lactones, flavonoids, polyalkynes and coumarins. The essential oil composition has been studied thoroughly and about 60 components have been identified; camphor, artemisia ketone, germacrene D and 1,8-cineole, are usually the main components (Ahmad et al., 1994; Kata-youn et al., 2005; Ma et al., 2007; Marco et al., 1998).

As a part of our studies on the chemical composition of the essential oils and screening programme for bioactive compounds from plants that grow in Iran, the present study describes the essential oil composition of aerial parts of *A. annua* L. in different growth stages.

MATERIALS AND METHODS

Plant materials

The aerial parts of *A. annua L.* were collected during three periods of pre-flowering on the 6th April, flowering on the 1st May, and post-flowering on the 15th of June of 2006 from the Gorgan province, north of Iran. A voucher specimen was deposited at the Herbarium of Faculty of Pharmacy, Tehran University of Medical Sciences.

Isolation of the essential oil

The aerial parts (100 g) were dried at 25°C in the shade and subjected to hydro distillation, using a Clevenger-type apparatus for 4 h. The oil was dried with anhydrous sodium sulphate, weighed and stored at 4 – 6°C in dark until use.

Gas chromatography/ mass spectrometry

(GC-MS). A Hewlett-Packard 6890 gas chromatography was used, with HP-5 capillary column (phenyl methyl siloxane of 25 m length, 0.25 mm i.d., and 0.25 µm film thickness). Carrier gas was He; split ratio was 1:25, and the detector was flame ionization. Temperature program was 60°C (2 min) rising to 240°C at 4 °C/min; injector temperature was 250°C, and detector temperature was 260°C. GC–MS was a Hewlett-Packard 6859 equipped with a quadruple detector, on a HP-5 column (see GC), operating at 70 eV ionization energy, using the same temperature program and carrier gas as above. Retention indices were calculated by using retention times of C8-

Table 1. The chemical composition of the essential oil of *A. annua L.* in different growth stages.

No.	Components	RI	Pre-flowering stage	Flowering stage	Post-flowering stage
1	Tricyclene	914	0.63	0.53	0.42
2	α – Thujene	919	0.28	0.20	0.30
3	α – Pinene	926	3.13	2.50	3.53
4	Camphene	941	6.98	1.70	3.24
5	Sabinene	967	0.16	0.20	0.15
6	β – Pinene	974	-	-	0.15
7	Dehydro – 1,8- cineole	981	0.25	0.45	0.36
8	Delta-3- carene	997	0.65	0.50	0.34
9	α - Terpinene	1008	0.21	0.30	0.60
10	Cymol	1017	1.31	1.27	1.65
11	1,8-Cineole	1024	9.39	13.90	12.00
12	γ-Terpinene	1049	0.39	0.42	0.20
13	Artemisia ketone	1055	2.68	3.37	5.45
14	*cis*- Sabinene hydrate	1058	0.29	0.45	0.56
15	Terpinolene	1078	1.27	2.70	3.70
16	Artemisia alcohol	1082	-	0.23	0.40
17	*trans*-Sabinene hydrate	1094	0.47	0.67	0.32
18	Camphor	1148	48.00	43.51	36.75
19	Pinocarvone	1151	-	2.12	1.76
20	Chrysanthenol	1155	-	0.30	0.40
21	Borneol	1162	2.53	1.79	2.67
22	Terpinene-4-ol	1170	1.40	0.30	0.29
23	Myrtenal	1184	0.78	0.10	0.17
24	Myrtenol	1187	0.93	0.20	0.40
25	*trans*-Carveol	1209	0.79	0.63	1.12
26	*cis*-Carveol	1219	0.42	0.53	0.41
27	Pregeijerene	1282	0.53	0.54	0.43
28	Eugenol	1342	0.18	-	-
29	Benzyl 2-methyl butyrate	1372	0.52	0.23	-
30	*trans*-Caryophyllene	1481	0.41	0.25	0.42
31	α – Neoclovene	1442	0.12	-	-
32	Farnesene	1459	1.25	0.15	0.12
33	Germacrene D	1462	-	0.30	-
34	β – Selinene	1468	1.01	-	-
35	Germacrene A	1507	-	0.73	0.60
36	γ-Cadinene	1518	-	0.10	0.12
37	Spathulenol	1564	4.89	3.73	4.50
38	Ledenoxid	1611	3.07	2.26	1.53
39	*Epi*-Cubenol	1615	1.84	1.20	-

C22 that were injected after the oil at the same chromatographic conditions according to Van Den Dool method (Van Den Dool et al., 1963).

Identification of components

The linear retention indices for all the compounds were determined by coinjection of the sample with a solution containing the homologous series of C8–C22 *n*-alkanes. The individual constituents were identified by their identical retention indices, referring to known compounds from the literature (Adams, 1995) and also by comparing their mass spectra with either the known compounds or with the Wiley mass spectral database.

RESULTS AND DISCUSSION

The essential oil content of aerial parts of *A. anuua* L. obtained by hydro distillation, were 0.97, 1.23 and 0.87% in pre-flowering, flowering and post-flowering stages respectively, calculated on dry weight basis. Table1 shows thirty-nine components of the oils that have been identified in different growth stages of *A. anuua*. Thirty-two components accounting for 97.67% of the total composition were identified in the pre-flowering stage. The major constituents of this oil were camphor (48.00%), 1,8 – cineole (9.39%), camphene (6.98%) and spathulenol (4.695%). In the volatile of flowering stage, thirty-five compounds amounting 92% of total components were identified which included camphor (43.50%), 1,8-cineole (13.90%), spathulenol (3.73%) and artemisia ketone (3.37%) as main components. In the oil obtained from post-flowering stage, thirty-four components were characterized, which represented about 92.4 % of the total composition. Camphor (36.75%), 1,8-cineole (12.00%), spathulenol (4.50%) and α – pinene were the principal components of this oil. The oil contained 69.96, 72.44 and 70.96% monoterpene and 18.56, 18.84 and 20.83% sesquiterpene compounds in pre-flowering, flowering and post-flowering stages respectively.

Conclusion

A comparison of chemical composition of the essential oil of *A. annua L.* at three stages of development shows that there are little differences in composition and major components, camphor, 1,8-cineole, camphene, spathunelol, α–pinene and artemisia ketone were the main compounds in all samples. Thus the time of harvesting of this plant does not have a major effect on chemical composition of the essential oil but it effects on the essential oil content of the plant and the flowering stage is the best time for harvesting the plant and obtaining the essential oil because at this time the plant contains highest percent of the essential oil.

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REFERENCES

Bertea CM, Freije JR, van der Woude H, Verstappen FWA, Perk L, Marquez V, De Kraker JW, Posthumus MA, Jansen BJM, de Groot A, Franssen MCR, Bouwmeester HJ, (2005). Identificationof intermediates and enzymes involved in the early steps of artemisinin biosynthesis in *Artemisia annua*. Planta Med. 71: 40-47.

Mozaffarian V, (1996). A Dictionary of Iranian Plant Names. Farhang Moaser: Tehran, Iran, pp. 56- 58.

Brown GD, Liang Gy, Sy L, (2003). Terpenoids from the seeds of *Artemisia annua*. Phytochem. 64: 303-323.

Ahmad A, Mishra LN, (1994). Terpenoids from *Artemisia annua* and constituents of its essential oil. Phytochem. 37:183-186.

Katayoun M, Akbarzadeh M, Moshiri K, (2005). Essential oil composition of *Artemisia fragrans* Willd. from Iran. Flavour Fragr J. 20: 330 – 331.

Ma C, Wang H, Lu X, Li H, Liu B , Xu G, (2007). Analysis of *Artemisia annua* L. volatile oil by comprehensive two-dimensional gas chromatography time-of-flight mass spectrometry. J Chromatogr A. 50: 50-53.

Marco JA, Sanz-Cervera JF , Ropero FJ, (1998). Germacranolides and a monoterpene cyclic peroxide from *Artemisia fragrans*. Phytochem. 47: 1417- 1419.

Van Den Dool H, Kratz PD, (1963). A generalization of the retention index system including linear temperature programmed gasliquid partition chromatography. J Chromatogr. 11: 463–471.

Adams RP, (1995). Identification of Essential Oil Components by Gas Chromatography and Mass Spectroscopy. Allured: Carol Stream, IL.