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Original Article

Study of the Chemical Composition of Essential Oils of *Perovskia abrotanoides* Karel at the Different Stage and Distillation by Gas Chromatography

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Abstract

Medicinal plante Borazambol with the scientific name of *Perovskia abrotanoides* Karel. belongs to the family Lamiaceae. It is growing wild in the margin of mountainous roads of arid and cold climate of north Iran. It is for a long time that indigenous people by different methods in traditional medicine use its products in preventing and curing diseases. In this research, essential oils of *Perovskia abrotanoides* Karel, extracted and measured at the time of flowering stage and vegetative stage by different methods of distillation and then were analyzed by GC and GC/MS. The essential oil yield at vegetative stage with hydro-distillation was 2.2%, water & steam distillation (Kyzer & Long) 5.7% and steam distillation 2.5%, respectively. Major component identificated by water distillation (Clavanger) were α- terpineol (32%), n-octanol (22.5%), myrcene (7.2%), and by water and water & steam distillation (Kyzer & Long) were α- terpineol (30.3%), n-octanol (20.1%), myrcene (7.2%), and by steam distillation were α- terpineol (26.2%), n-octanol (17.4%), n-pentadecane (8.2%). The essential oil yield at flowering stage with hydro-distillation was 1.9%, water & steam distillation (Kyzer & Long) 1.5% and steam distillation 1%, respectively. Major component identificated by water distillation (Clavanger) were n-octanol (23.3%); α- terpineol (21.9%) and (Z)-β- ocimene (15.5%), and by water and steam distillation (Kyzer & Long) were α- terpineol (19.9%), n-octanol (19.9%), (Z)-β- ocimene (13.6%), and by steam distillation were n-octanol (16.6%), α- terpineol (15.3%), and (Z)-β- ocimene (13.5%).

Key words: Essential oil, *Perovskia abrotanoides* Karel , flowering stage, vegetative stage, GC, GC/MS.

Introduction

The genus *Perovskia*, which belongs to the tribe stachyo ideae-Nepeteae, family Lamiaceae, is distributed in various regions of Asia, Afghanistan, Himalaja, Turkestan, Pakistan and Tibet. The Iranian flora includes four species of *Perovskia* which one of them is *Perovskia abrotanoides* Karel [1]. Some other members of this genus are *P. atriplicifolia Benth., P. scrophulariifolia* and *P. angustifolia* [2-5].

Perovskia abrotanoides Karel. is one of the valuable medicinal species in the north of Iran and it is growing wild in the margin of mountainous roads of arid and cold climate of Golestan and north Khorasan. *P. abrotanoides* is an herb used to treat leishmaniasis in Iranian folk-medicine tradition. Thus, villagers in the Isfahan province of Iran apply a poultice, made of crushed roots of the plant, water, sesame oil, and wax, on lesions caused by cutaneous leishmaniasis [6]. The essential oil of this plant from Pakistan possessed antibacterial activity against salmonella typhi, which was

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comparable to that of chloromphenicol but was much less than that of streptomycin [7]. A new triterpene, perovskone with a novel carbon skeleton was isolated from the whole plant of *P. abrotanoides* [8], a novel triterpene, peradione was isolated from the Pakistani medicinal plant *P. abrotanoides* [9], and a quinoid diterpene with a nor-abietane skeleton, and three new natural products were isolated from roots of *P. abrotanoides*, have leishmanicidal, antiplasmodial , and cytotoxic activity [6].

In this communication, we report on the volatile oil contents and composition of different plant organs of *P. abrotanoides* Karel at flowering stage and vegetative stage by different methods of distillation. To our knowledge the oil of *P. abrotanoides* Karel from Mazanderan provience (area kyasar, Sary) has not been studied previously.

Materials and Methods

Plant material

The plant material of *Perovskia abrotanoides Karel.* (Persian name: brazambel) were collected on August 2012 from Mazanderan provience (area kyasar, Sary) Iran, and were dried in the shade at room temperature. The specimen is deposited in Central Herbarium of Iran (TARI). (see: Holmgren, Index Herbariorum).

Isolation of the essential oil

100 gr. of dried airal parts of *Perovskia abrotanoides Karel.* were extracted by different methods of distillation (hydro-distillation (Clavanger type), water and steam distillation (Kyzer & Long), and steam distillation), The essential oil yield at the time of eruption with hydro-distillation were 2.2%, water & steam distillation (Kyzer & Long) 5.7% and steam distillation 2.5%, respectively. The essential oil yield at flowering time with hydro-distillation was 1.9%, water & steam distillation (Kyzer & Long) 1.5% and steam distillation 1%, respectively. The quantitative and qualitative analyses of the oils were performed by GC and GC-MS, respectively.

Gas Chromatography

GC analyses were performed using a Shimadzu-9A gas chromaph equipped with a flame ionization detector, and quantitation was carried out on Euro Chrom 2000 from Knauer by the area normalization method neglecting response factors. The analysis was carried out using a DB-5 fused-silica column (30m x 0.25 mm, film thickness 0.25 μm, J & W Scientific Inc., Rancho Cordova, CA, USA). The operating conditions were as follows: injector and detector temperature, 250 °C and 265 °C, respectively; carrier gas,Helium. Oven temperature programme was 40 °- 250 °C at the rate of 4 °C/min.

Gas Chromatography - Mass Spectrometry

The GC/MS unit consisted of a Varian Model 3400 gas chromatograph coupled to a Saturn II ion trap detector was used. The column was same as GC, and the GC conditions were as above. Mass spectrometer conditions were: ionization potential 70 eV; electron multiplier energy 2000 V.

The identity of the oil components was established from their GC retention indices, relative to C_7- C_{25} n-alkanes, by comparison of their MS spectra with those reported in the literature [10-12], and by computer matching with the Wiley 5 mass spectra library, whenever possible, by co-injection with standards available in the laboratories.

Results

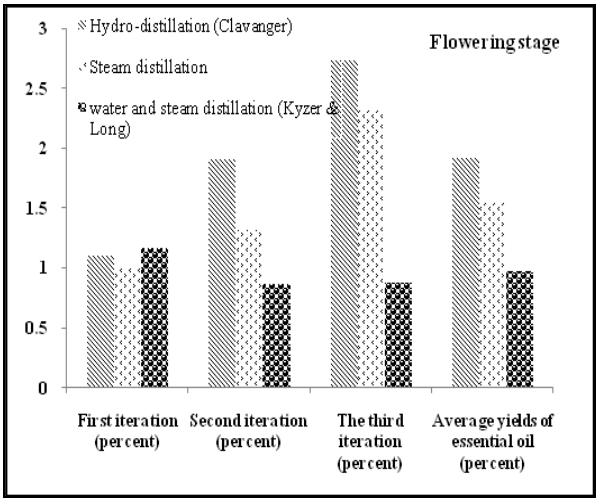

Fig. 1 A comparison diagram of essential oil from *Perovskia abrotanoides* Karel in flowering stage in triplicate

Fig. 1 show a comparison diagram of essential oil from *Perovskia abrotanoides* Karel in flowering stage in triplicate and Fig. 2 show a comparison diagram of essential oil from *Perovskia abrotanoides* Karel in vegetative stage in triplicate.

Table l Essential oils percentage of perovskia abrotanoides karel at flowering stage and vegetative stage by different methods of distillation

		Flowering stage			Vegetative stage		
Compound	R.I.	Clavanger	Kyzer & Long	Steam	Clavanger	Kyzer & Long	Steam
Camphene	956	7.0	8.4	7.8	4.8	5.3	4.8
Sabinene	973	3.7	3.8	3.3	3.2	3.2	3.0
Myrcene	989	0.9	1.0	1.0	7.2	7.4	6.9
δ-2-careen	1010	0.9	1.0	0.9	-	-	-
(Z)-β-ocimene	1041	15.5	13.6	13.5	2.5	2.2	2.3
(E)-β-ocimene	1051	1.2	-	-	0.5	-	0.3
γ-terpinene	1056	2.0	0.5	1.7	1.2	0.5	1.0
N-octanol	1066	23.3	19.9	16.6	22.5	20.1	17.4
Terpinolene	1085	0.8	0.3	-	0.7	0.5	-
Trans-thujone	1113	0.6	0.6	0.5	0.4	0.3	0.3
α-terpineol	1184	21.9	19.9	15.3	32.0	30.3	26.2
Trans-carveol	1217	2.3	2.1	1.4	3.1	3.1	2.5
Cis-carveol	1228	0.3	0.3	---	0.4	0.9	0.3
Methyl decanoate	1321	1.7	1.7	1.6	1.4	1.3	1.4
α-copaene	1371	1.5	1.4	1.1	1.2	1.2	1.3
β-copaene	1431	0.9	1.4	2.0	0.9	1.4	1.5
γ-gurjunene	1475	1.2	1.8	2.1	1.1	1.5	1.8
N-pentadecane	1500	4.0	6.3	6.7	4.7	6.7	8.2
Trans-calamenene	1529	2.4	4.6	5.6	3.7	4.9	6.4
Elemicin	1553	0.4	0.6	1.9	1.0	1.3	1.8
Spathulenol	1576	3.0	4.7	6.7	3.5	4.4	6.4
Caryophyllene oxide	1582	-	-	-	0.5	0.6	0.9
Z,E-farnesol	1713	0.2	0.3	0.7	-	-	-
(E,E)-farnesyl cetate	1727	0.3	0.3	1.0	-	-	0.5
(E,Z)-farnesol	1741	1.5	1.9	2.5	1.6	1.0	2.6
Benzyl salicylate	1864	-	-	0.5	-	-	-
1-eicosene	1975	0.1	0.2	0.6	-	-	-

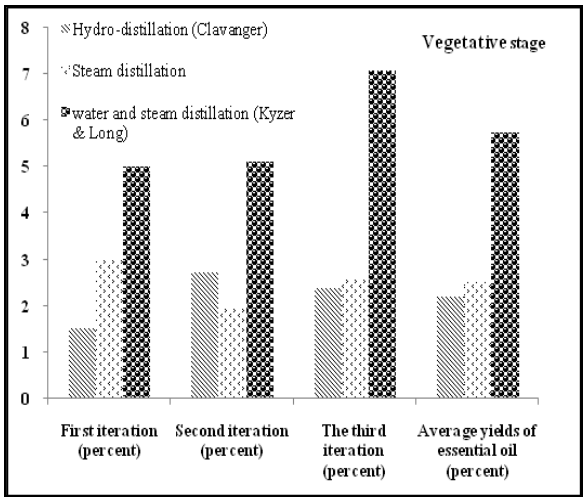

Fig. 2 A comparison diagram of essential oil from *Perovskia abrotanoides* Karel in vegetative stage in triplicate

Also in Tables 1 show retention indices and relative percentages of the oil constituents from the identified compounds in August 2012 from Mazanderan provience (area kyasar, Sary) Iran. Twenty seven components were identified by GC and GC-MS representing about 84.83-100% of the oils. Major component identificated by water distillation (Clavanger) were α- terpineol (32%), n-octanol (22.5%), myrcene (7.2%), and by water and water & steam distillation (Kyzer & Long) were α-terpineol (30.3%), n-octanol (20.1%), myrcene (7.2%), and by steam distillation were α- terpineol (26.2%), n-octanol (17.4%), n-pentadecane (8.2%). Major component identificated by water distillation (Clavanger) were n-octanol (23.3%); α-terpineol (21.9%) and (Z)-β- ocimene (15.5%), and by water and steam distillation (Kyzer & Long) were α- terpineol (19.9%), n-octanol (19.9%), (Z)-β- ocimene (13.6%), and by steam distillation were n-octanol (16.6%), α- terpineol (15.3%), and (Z)-β- ocimene (13.5%). The comparison of the results with the literature showed significant differences for oils, which can be attributed to either climatological factors or genetic differences or development stages or plant parts analyzed.

Results and Discussion

In the current research the most important compositions in the essential oil from Perovskia abrotenoides Karel were α- terpineol (32%) and including n-octanol (22.5%), by hydro-distillation (Clavanger type) method. Our results are differnts with other authors, because of condition of climate and time of collection, therefore, as it is shown in the results of the research, essential of one species in different regions and different period showed different compositions and this case is of great importance in the determination of the region for a good sample for industry. The investigations of traditional medicine showed that *P. abrotenoides* is anti-inflammatory, anti-pain and this is proved in animal model researches [14-16]. According to the reports, villagers in the Isfahan province of Iran apply a poultice, made of crushed roots of the plant, water, sesame oil and wax, on lesions caused by cutaneous leishmaniasis [13-15]. In Golestan province P.abrotenoides is used to treat leishmaniasis and dermal problems [17]. Golestan and north Khorasan provinces are located in the north east of Iran and are one of the natural habitats of different species of *Perovskia abrotanoides* Karel. and it is for a long time that this plant is used in the traditional medicine of the people in this region to prevent some diseases. In Pakistan *P.abrotenoides* is used as refrigerant [18].

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