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Phytochemical of constituents of essential oil from *Myrtus communis* L. by GC-MS analysis

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ABSTRACT

Background and aims: Humans always have been considered the use of medicinal plants in the treatment of diseases because of fearing the side effects of chemical drugs. The current study aimed to identify the essential oil of *Myrtus communis* L. collected from natural habitats province of Khuzestan and Kohkiluyeh and Boyer-Ahmad.

Methods: In this experimental study, *Myrtus communis* L. was collected from Khuzestan and Kohkiluyeh and Boyer-Ahmad in July 2015. Plant material was identified by Ramin Agriculture and Natural Resources University of Khuzestan. 50 gm of cleaned and dried plant material was powdered and pestle and placed in a round bottom flask fitted with condenser hydro distilled for 3hrs at atmospheric pressure and constant temperature. The strongly aromatic oil was separated from the water layer using n-Hexane and the solvent was removed by boiling. The component identification was achieved by the GC-MS analysis.Total GC running time was 75 min.

Results: Phytochemical analyses of the essential oil collected from samples were contains 31 combinations. Secondary compounds in essential oils were including: α-Pinene, 1,8-Cineole, Limonene, Linalool L, α-Terpineol, Linalyl acetate, Geranyl acetate. α-Pinene and 1,8-Cineole were highest percentage of compositions. α-Terpinene combination was only existed in Kohkiluyeh and Boyer-Ahmad samples and Limonene combination was only existed in Khuzestan samples.

Conclusion: The α-Pinene and 1,8-Cineole are two dominant component in the essential oil of *Myrtus communis* L. in studied samples. Considering the effect of environmental factors on metabolites in medicinal plants identifying the metabolites in this medicinal and commercial plants will increase cost-effectiveness.

Keywords: 1,8-Cineole, α-Pinene, Essential oil, *Myrtus communis* L.

INTRODUCTION

Humans always have been considered the use of medicinal plants in the treatment of diseases because of fearing the side effects of chemical drugs.[1]

Researchers have been identified 145 genera of Myrtle that 16 genera of Myrtle found in Middle East and Asia.[2,3] *Myrtus communis* L. is one herb that is

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used traditionally in medicinal and nutritional purposes.[1] Myrtus species are rich resource of volatile oils, phenolic acids, tannins, anthocyanin pigments, and fatty acids.[3-6] In addition to food and industrial applications, several effects including anti-carcinoma and anti-inflammatory effect are been attributed to the compounds in the *Myrtus communis* L.[7-9]

Gas Chromatography-Mass Spectrometry (GC-MS) is the most popular method for the determination of essential oil composition. Components existing in the essential oil can be identified by comparison of their relative retention time or indices and their mass spectra (MS).[10]

Because of the weather and climatic conditions are likely effects on compounds in the plants. Therefore, the current study was aimed to identify the essential oil of *Myrtus communis* L. collected from the natural habitats province of Khuzestan (Mollasani) and Kohkiluyeh and Boyer-Ahmad (Ghachsaran).

METHODS

The geographical coordinates of the places where they took samples are shown in Table 1.

Table 1: Geographical Profile the region

Region Name	Latitude	Longitude	Height
Mollasani	31º 35′	48º 53′	25 m
Gachsaran	30º 15′	50º 45′	970 m

Due to determine the soil characteristics of the seed source, one kilogram of soil at a depth of thirty centimeters was taken randomly from each location. Samples were transported to the laboratory in Khuzestan Ramin Agriculture and Natural Resources University to evaluate some soil physical and chemical characteristics. Measurements of soil characteristics are shown in Table 2.

Table 2: Soil characteristics of the region

Region Name	E.C dSm^2	pH	O.C %	Sand%	Silt%	Clay%
Mollasani	6	8	0.55	14	69	34
Gachsaran	2.59	7.68	3.19	26	57	27

In this experimental study, *Myrtus communis* L. was collected from Khuzestan (Mollasani) and Kohkiluyeh and Boyer-Ahmad (Ghachsaran), in July 2015. Plant material was identified by Ramin Agriculture and Natural Resources University of Khuzestan and transported to Analytical chemistry laboratory for essential oil extraction.

50 gm of cleaned and dried plant material was powdered using metal mortar and pestle and placed in a round bottom flask fitted with condenser hydro distilled for 3h at atmospheric pressure and constant temperature. The strongly aromatic oil was separated from the water layer using n-Hexane and the solvent was removed by boiling.

The component identification was achieved by the GC-MS analysis using Agilent Technologist 7890 series GC equipped with mass selective detector (MSD), 5975 series in Centrallab of Agriculture and Natural Resources University of Khuzestan. Helium was used as the carrier gas at a constant flow of 0.8 ml/min and an injection volume of 1µl was employed, injector temperature 290°C; Ion-source temperature 280°C. The oven temperature was programmed from 50°C (isothermal for 5 min), with an increase of 3°C/min, to 240°C and with an increase of 15°C/min, to 280°C held for 10 min. Total GC running time was 75 min.

The components of essential oil were identified on the basis of comparison of their retention time and mass spectra with published data and computer matching with Wiley 7n and National Institute of Standards and Technology (NIST5.0) libraries provided with computer controlling the GC-MS system, and Adams book in Analytical Chemistry laboratory of Agriculture and Natural Resources University of Khuzestan. The spectrum of the unknown component was compared with the spectrum of the known components stored in the library. The name of the components of the test materials was ascertained.[11]

RESULTS

The retention times and chemical composition of phytocomponents present in *Myrtus communis* L. essential oil are presented in Table 3, chromatogram essential oil Mollasani Figure 1, and chromatogram of essential oil in Gachsaran Figure 2.

Table 3: Phytocomponents identified in *Myrtus communis* L. essential oils

Component	KI	Component %	
		Mollasani	**Gachsaran**
α-Thujene	0930	0.10	0.16
α-Pinene	0939	42.66	34.43
Camphene	0954	0.11	0.08
ß-Pinene	0979	0.26	0.33
Myrcene	0991	0.20	0.21
α-Terpinene	1017	-	0.09
O-Cymene	1026	0.99	0.94
Limonene	1029	2.69	-
1,8-Cineole	1031	23.15	30.45
(E)-ß-Ocimene	1037	0.11	0.10
γ-Terpinene	1060	0.12	0.19
(E)-Linalool oxcide	1073	0.12	0.09
Terpinolene	1089	0.12	0.17
Linalool L	1097	6.49	6.99
α-Campholenal	1126	0.51	0.47
(E)-Pinocarveol	1139	1.09	0.82
(Z)-Verbenol	1141	0.13	0.12
Delta-Terpineol	1166	0.14	0.18
Terpineol-4-ol	1177	0.31	0.34
α-Terpineol	1189	4.57	5.39
Estragol	1196	0.17	0.47
(E)-Carveol	1217	0.42	0.36
Nerol	1230	0.28	0.28
Linalyl acetate	1257	2.15	1.80
Thymol	1290	0.07	0.11
Carvacrol	1299	0.09	0.11
Neryle acetate	1362	0.37	0.30
Geranyl acetate	1381	1.86	2.41
(E)-Caryophyllene	1419	0.08	0.16
α-Humulene	1455	0.08	0.15
Humulene epoxide II	1608	0.54	0.50
Caryophyllene oxide	2200	0.82	0.92

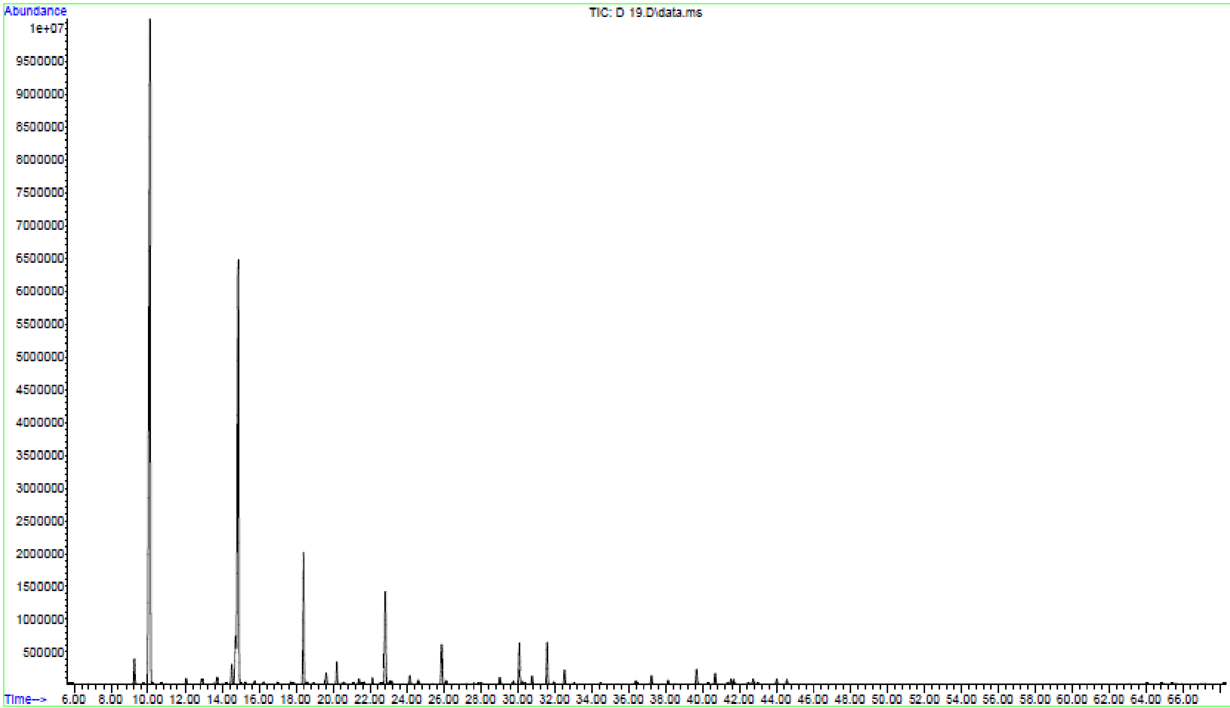

Figure 1: Chromatogram of essential oil in Mollasani

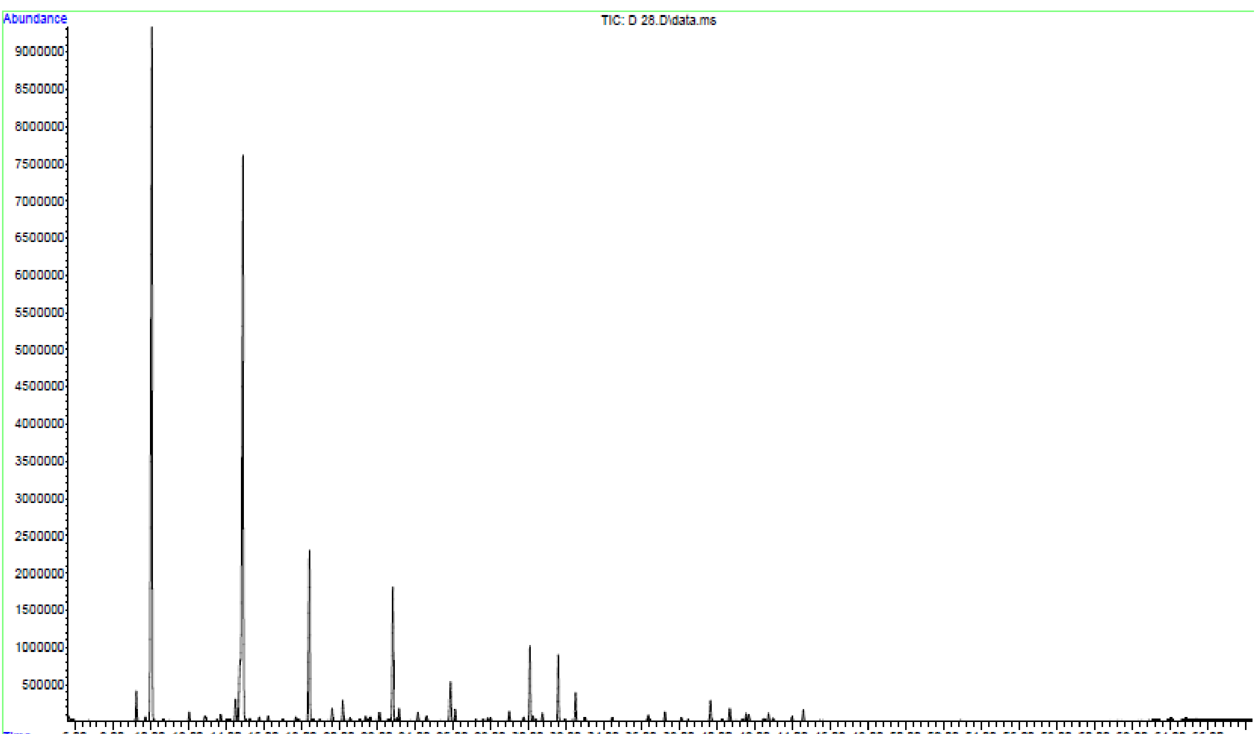

Figure 2: Chromatogram of essential oil in Gachsaran

Secondary compounds that were identified in essential oils, including: α-Pinene, 1,8-Cineole, Limonene, Linalool L, α-Terpineol, Linalyl acetate, Geranyl acetate. The α-Pinene (Figure 3) and 1,8-Cineole (Figure 4) were highest percentage of compositions. The α-Terpinene combination was only existed in Kohkiluyeh and Boyer-Ahmad samples, while, Limonene combination was only existed in Khuzestan samples.

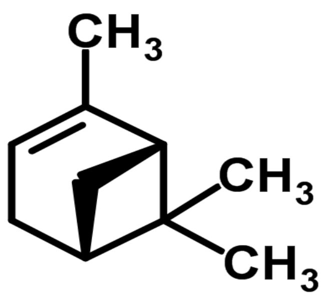

Figure 3: Structure of α-Pinene

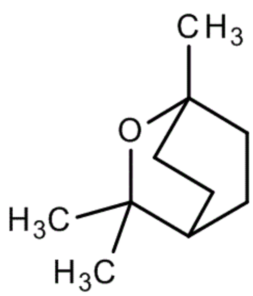

Figure 4: Structure of 1,8-Cineole

DISCUSSION

Based the results of the present study, α-Pinene and 1, 8-Cineole were highest percentage of compositions. This result is in line with previous studies.[12,13]

We well know that α-Pinene with C10H16 formula is a 2 rings hydrocarbon monoterpenes which is very important in commercial. It is used in combination with ß-Pinene in the preparation of extracts, condiments, food and polymers.[7] Recently a study investigated the anti-tumor effect of α-Pinene on human hepatoma cell and reported excellent results. The results of this study showed that α-Pinene has a liver cancer cell growth inhibitory effect including inhibitory rate of 79.3% *in vitro* and 69.1% *in vivo*.[8]

Today, an important anti-inflammatory property of 1, 8-Cineole have been identified. A study performed at 2012 reported that 1,8-cineole has analgesic and anti-inflammatory effects possibly due to its inhibition of TRPA1.[9]

It was found in the current study that the α-Terpinene combination was only existed in Gachsaran samples. However, the Limonene combination was only existed in Mollasani samples. The researchers believe that these differences in the type and amount of substances can be attributed to various climate conditions. Although the productions of secondary metabolites are controlled by genes, but their production will be significantly affected by environmental conditions. Physical and chemical properties of soil, micronutrients and macronutrients are the most important factors.[13] In an Iranian study declared that latitude and longitude are the most important factors that affect the synthesis of secondary metabolites particularly essential in plants.[14]

CONCLUSIONS

The α-Pinene and 1,8-Cineole are two dominant components in the essential oil of *Myrtus communis* L. in studied samples. Considering the effect of environmental factors on metabolites in medicinal plants identify the metabolites in this medicinal and commercial plants will increase cost-effectiveness.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interests.

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