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Chemical compositions of peppermint (*Mentha piperita* L.) grown in Isfahan province, Iran

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Abstract

Peppermint is scientific named *Mentha peperita* L. belongs to Lamiaceae family. The Mediterranean region can be described as the center of the genus. Peppermint is one of most important medicinal plants that was used in food, cosmetics hygienic industries in most of the developed countries. *Mentha* species have strong anti-spasmodic, anti-sickness, anti-helminthic, carminative and stomachic activities. Breeding programs need to identify the best land races as base population. Existence sufficient genetic diversity also is important in selection the superior cultivars. *Mentha piperita* L. or peppermint oil is one of the most popular and widely used essential oils, mostly because of its main components menthol and menthone. This study was conducted to determine the major components of peppermint cultivated in Isfahan province. The aerial parts of plants were collected in Isfahan (Semiroom) Province in 2014. The aerial parts of plants analyzed by using GC/MS in Islamic Azad University Isfahan (Khorasgan). Twenty five compounds were identified in aerial parts of peppermint. The results obtained in the study showed that major components of peppermint were Camphane (15.203%), Menthone (12.013%), Menthol (11.406%), β -Pinene (7.62%), Pulegone (6.42%), β -Cubebene (4.95%), α -Pinene (4.012%), γ -Terpinene (4.081%), Carane (3.82%) and Piperiton (3.05%). This land race was collected from Isfahan province having high amount of Camphane, Menthone and Menthol so is promising population for breeding programs via selection to improve medicinal value of this important medicinal plant.

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Introduction

Peppermint (*Mentha piperita* L.) from Lamiaceae family is one of the most important medicinal plants, used in food, sanitary and cosmetic industries. The leaves are strongly scented due to the presence of essential oils. Peppermint is widely used for its medicinal properties such as anti-spasmodic, anti-sickness, anti-helminthic, carminative, stomachic and others. Peppermint cultivated in the temperate, Mediterranean and subtropical regions of the world (Nostro *et al.*, 2000; Ormancey *et al.*, 2001). Peppermint (*Mentha piperita* L.) which is tetraploid (2n=72), is a sterile natural hybrid of *M. aquatica* L. (2n=96) and *M. spicata* L. (2n=48) (Tucker, 1992). The plant is a perennial, 50-60 cm high. The square stems are usually reddish-purple and smooth. The leaves are short, oblong-ovate and serrate. The flowers are purple-pinkish and appear in the summer months. The plant has runners above and below ground (Mozaffarian, 1996; Yazdani *et al.*, 2003). The essential oil of peppermint (up to 2.5% in the dried leaves) is mostly made up from Menthol (ca. 50%), Menthone (10 to 30%), Menthyl esters (up to 10%) and further Monoterpene derivatives (Pulegone, Piperitone, Menthofurane) (Murray, 1995). Derwich *et al.* (2010) found the twenty nine compounds were identified in leaves oil representing 58.61% of the total oil composition. The yield of essential oil of *Mentha piperita* was 1.02% and the major compound in aerial parts was: Menthone (29.01%), followed by Menthol (5.58%), Menthyl acetate (3.34%), Menthofuran (3.01%), 1,8-cineole (2.40%), Isomenthone (2.12%), Limonene (2.10%), α -pinene (1.56%), Germacrene-D (1.50%), β -pinene (1.25%), Sabinene (1.13%) and Pulegone (1.12%). report on major constituents of *M. piperita* essential oil: Menthol (18 mg/g) and Neo-menthol (0.72 mg/g) (Ka *et al.*, 2005), as well as Menthol (28-42 %), Menthone (19-27 %), and 1,8-cineole (4-5 %) (Iskan *et al.*, 2002). The aim of this study was to identify the chemical diversity and components of peppermint (*Mentha piperita* L.) cultivated in Iran province.

Materials and Methods

Plant materials

The aerial parts of peppermint (*Mentha piperita* L.) were collected from the center of Iran, Semirom city, Isfahan province (35° 18' N and 52° 18' E, 1965 m above sea level) in 2014. The aerial parts of plants analyzed by using GC/MS in Islamic Azad University, Isfahan (Khorasgan) Branch.

Essential oil extraction

The fresh aerial parts of Denaian thyme was dried inside for six days at room temperature (25 \pm 5 °C), and the ground to fine a powder using Moulinex food processor. The essential oil was extracted from 100 g of ground tissue in 1 L of water contained in a 2 L flask and heated by heating jacket at 100 °C for 2 h in a Clevenger-type apparatus, according to producers outlined British Pharmacopoeia. The collected essential oil was dried over anhydrous sodium sulphate and stored at 4 °C until analysed.

Identification of the oil components

The GC/MS analysis was carried out with a 20 Agilent 5975 GC-MSD system. HP-5MS column (30m \times 0.25mm, 0.25 μ m film thickness) 20 was used with helium as carrier gas (1.5 mL / min). GC oven temperature was kept 20 at 50 C2 BoC for 4 min and programmed to 280 C2 BoC at a rate of 5 C2 BoC/min, and kept 20 constant at 280 C2 BoC for 5 min, at split less mode. The injector temperature was at 20 280 C2 BoC. Transfer 20 line temperature 280 C2 BoC. MS were taken at 70 20 eV. Mass range was from m/z 35 to 450.

Results and Discussion

GC-MS analysis

The twenty five compound were identified in aerial parts of peppermint. The results obtained in the study showed that major components of peppermint were Camphane (15.203%), Menthone (12.013%), Menthol (11.406%), β -Pinene (7.62%), Pulegone (6.42%), β -Cubebene (4.95%), α -Pinene (4.012%), γ -Terpinene (4.081%), Carane (3.82%) and Piperiton (3.05%)

(Table 1 and Fig. 1). However, 25 components were identified in aerial parts of peppermint in this study, only a few of the main components are discussed here.

Table 1. Chemical composition of *Mentha piperita* L. dried aerial parts.

Compound ^a	RT ^b	Content %
α-Thujen	6.069	0.802
α-Pinene	6.182	4.012
β-Pinene	7.374	7.621
(+)-4-Carene	8.450	0.405
γ-Terpinene	9.876	4.081
α-Terpinolene	10.591	0.625
Menthone	12.932	12.013
α-Terpineol	14.835	1.491
Pulegone	15.590	6.415
Piperiton	15.928	3.047
Carane	16.270	3.816
Camphane	16.916	15.203
Menthyl acetate	17.138	1.640
Camphene	17.701	1.087
α-Cubebene	17.987	0.430
α-Copaene	18.655	1.293
β-Caryophyllene	18.919	3.422
Menthol	19.929	11.406
Germacrene-D	20.051	1.952
β-Cubebene	20.337	1.116
α-Caryophyllene	20.575	2.351
β-Farnesene	20.645	1.560
β-Cubebene	21.291	4.950
α-Amorphene	21.629	2.625
delta-Cadinene	22.166	0.988

^a Compounds listed in order of elution

^b Rt (retention time)

The components of peppermint oil vary slightly from year to year. This may be mostly due to changes in weather conditions and the effect of weather on chemotypes of mints. Yazdani *et al.* (2003) reported the comparative essential oil and Menthol of *Mentha piperita* L. different origin cultivated in Iran, the highest and lowest of Menthol content in essential oil was found to be 56.4% (Sari Province) and 34.4%

(Tafresh, Markazi Province) respectively. The highest and lowest of Menthol content in the dried leaves was found to be 1.49% (Kerend-e Gharb, Kermanshah Province) and 0.76% (Tafresh, Markazi Province), respectively.

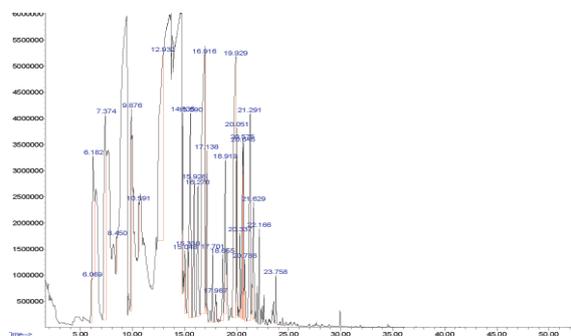


Fig. 1. Chromatogram of *Mentha piperita* L. dried aerial parts compounds.

Jaimand *et al.* (2001) reported the essential oils composition of two cultivars of *Mentha piperita* L. main components from (sample 1) Karaj city, Kalak representing 93.55% of total oil were, Neo-menthol (42.62%), 1,8-Cineole (16.51%) and piperitone (12.25%), and for second sample from Kamoo village 70 Km away from Kashan city, main components are L-menthol (37.55%), L-menthone (19.13%), 1, 8 - Cineole (11.48%) and Menthofuran (4.45%). Soltani *et al.* (2009) reported the results showed that essential oil composition of *M. piperita* in two harvests was different. In first harvest, the main constituents of the oil were Menthone, Menthol, Menthofuran, Pulegone, 1,8-Cineole and menthyl acetate, respectively whereas the main constituents of the oil in second harvest were Menthol, Menthone, Neo-Menthone, 1,8-Cineole and Menthyl acetate respectively. Mirza *et al.* (2011) reported the major constituents of essential oils in *M.piperita* at early, full and after flowering stages were Menthol (27.7%, 26.9%, 27.0%), Menthon (37.0%, 21.9%, 17.2%), and Menthofuran (16.0%, 22.0%, 25.3%), respectively.

Peppermint of different origins contains almost the same components, and differences are found in the percentage of components. The Menthol content in peppermint of Chinese origin was lower and limonene

was higher than that of USA origin (Aflatuni *et al.*, 2000). The major components of *Mentha piperita* essential oils analyzed in Serbia were Menthol (37.4%), Menthyl acetate (17.4%) and Menthone (12.7%) (Iscan *et al.*, 2002). Menthol and Menthone were the main components of *Mentha piperita* (Sokovic *et al.*, 2009). Menthol (64.0%), Menthyl acetate (9.2%) and Menthofuran were dominant in *Mentha piperita* study in Italy by (Ashok *et al.*, 1999). Also, Menthanol (36.24%) and Menthone (32.42%) were the major compounds of the *Mentha piperita* essential oil study in Iran (Behnam *et al.*, 2006). Menthon (44.1%), Menthol (29.5%), Menthylacetate (3.8%) and Menthofuron (0.9%) were the major compounds of % *Mentha piperita* from Turkey (Arldogan *et al.*, 2002).

Contrary it's different to the composition of essential oil of leaves of *Mentha piperita* study in Korea which the major component were Linalyl acetate (28.2%), Menthol (33.4%), 1,8-cineole (46.1%), Limonene (64.5 and 94.2%), and P-menth-2-en-ol (34.5%) (Seun-Ah *et al.*, 2010). The main components of the oils of *M. piperita* were α -terpene (20.11%), Pipertitinone oxide (17.10%) and Trans-carveol (19.48%) (Eteghad *et al.*, 2009). The Chemical composition of *M. piperita* L essential oil study in Iran, contained α -terpinene (19.7%), Isomenthone (10.3%), Trans-carveol (14.5%), Pipertitinone oxide (19.3%), and β -caryophyllene (7.6%) as the major compounds (Yadegarinia *et al.*, 2006). Intensive research on the chemical characteristics has been conducted on this species (Carla and Decorti, 2009). Different medicinal plant species show a marked variation in active ingredients during different seasons; these have been widely attributed to variations in environmental variables such as temperature and rainfall (Ahmad *et al.*, 2009).

Conclusion

Comparison between these results and the results of the other reports showed differences, probably due to that plant varieties or sites, as well as the time of harvesting. Differences observed may be due to the

different environmental and genetic factors, different chemotypes and the nutritional status of the plants or any other factors that can influence the oil composition. Therefore, this land race that has collected from Isfahan province could be used as the base population in breeding programs. Selection of the best individual had the genetic efficiency to improve peppermint as one of the most important and valuable medicinal plant.

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