

Volatile Constituents of Essential Oils Isolated from Flowers and Leaves of *M.officinalis* L.

Mohsen Kazemi^{1*} and Farshid Esmaili¹

¹Department of Horticultural Science, Faculty of Agricultural Science and Natural Resources, Science and Research Branch, Islamic Azad University, Tehran, IRAN

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ABSTRACT

Volatile components of leaves of *Melissa officinalis* L. (*Lamiaceae* family) from Iran were investigated. Essential oils were isolated by hydrodistillation from leaves of *Melissa officinalis*. Twenty-one constituents in the leaves oil were identified. Major constituents of the leaves oil were: trans-b-ocimene (4.5%), citronellal (3.21%), cadinol isomer (3.45%), neral (4.32%), geranial (8%), b-caryophyllene (16%), b-cubebene (10.21), calarene (4.32%), b-cadinene (4%), sesquiterpene alcohol (10.43%) and a-cadinol (9.05%). However, there were significant differences among the rates of those reported components. In conclusion, it is worthwhile to screen the commonly used plants from the local flora for different biological activities because they might present a new alternative source for possible bioactive substances.

Key Words: *Melissa officinalis*, *Lamiaceae*, Essential oil composition, b-Caryophyllene

INTRODUCTION

Lemon balm (*Melissa officinalis* L.) belongs to the family *Lamiaceae* and grows widely in central and southern Europe and in Asia minor (Zargari 1990). Lemon balm, one of the important medicinal plant species mainly grown in natural flora -especially in the north, north-west and western parts Iran. In Iran, this plant is known locally by the names Badranjbooye (Anon 2002). Main components *Melissa officinalis* are citronellal, citral a, and citral b, as well as other monoterpenes and sesquiterpenes. Other ingredients are tannins unique to the *Lamiaceae*, such as triterpenylic acid, bitter principles, and flavonoids (Mulkens and Kapetanidis 1987; Dimitrova *et al.* 1993). It is used in the Iranian traditional system of medicine for the treatment of headaches, flatulence, indigestion, colic, nausea, nervousness, anaemia, vertigo, syncope, malaise, asthma, bronchitis, amenorrhea, cardiac failure, arrhythmias, insomnia, epilepsy, depression, psychosis, hysteria, ulcers and wounds (Zargari, 1990; Anon, 2002). The present work was undertaken to determine the chemical composition of essential oils from *Melissa officinalis* wild in several locations of north, Iran.

MATERIALS AND METHODS

Plant material and oil isolation

The plant materials were collected from the several locations of north -Iran in 2012- 2013. The *Melissa officinalis* leaves were ground and the resulting powder was subjected to hydrodistillation for 3 hours in an all glass Clevenger-type apparatus according to the method recommended by the European Pharmacopoeia (1975). The obtained essential oils were dried over anhydrous sodium sulphate and after filtration, stored at +4 °C until tested and analysed.

Essential oil analysis

The GC/MS analyses were executed on a Hewlett–Packard 5973N gas chromatograph equipped with a column HP-5MS (30 m length × 0.25 mm i.d., film thickness 0.25 μm) coupled with a Hewlett–Packard 5973N mass spectrometer. The column temperature was programmed at 50 °C as an initial temperature, holding for 6 min, with 3 °C increases per minute to the temperature of 240 °C, followed by a temperature enhancement of 15 °C per minute up to 300 °C, holding at the mentioned temperature for 3 min. Injector port temperature was 290 °C

* Corresponding author: kazemimohsen85@hotmail.com

and helium used as carrier gas at a flow rate 1.5 ml/min. Ionization voltage of mass spectrometer in the EI-mode was equal to 70 eV and ionization source temperature was 250 °C. Linear retention indices for all components were determined by coinjection of the samples with a solution containing homologous series of C8-C22 *n*-alkanes and comparing them and their mass spectra with those of authentic samples or with available library data of the GC/MS system (WILEY 2001 data software) and Adams libraries spectra (Adams 2001).

RESULTS AND DISCUSSION

The GC/MS of oil shows the presence of 21 compounds and 91.96% of essential oil has been identified (Table 1). The yield of essential oil obtained from leaves of plants was 0.7% (v/w). The major constituents of oil were trans-*b*-ocimene (4.5%), cadinol isomer (3.45%), citronellal (3.21%), neral (4.32%), geranial (8%), *b*-caryophyllene (16%), *b*-cubebene (10.21), calarene (4.32%), *b*-cadinene (4%), sesquiterpene alcohol (10.43%) and *a*-cadinol (9.05%). Other components were amounts less than 3%. Lemon balm contains the flavonoids quercitrin and rhamnocitrin, and the 7-glucosides of apigenin, kaempferol, quercetin, and luteolin; phenolic acids and tannins, chiefly rosmarinic acid (up to 4%), and glycosidically bound caffeic and chlorogenic acids; triterpenes (ursolic, oleanolic acids); volatile oil (0.05–0.375%), of which the monoterpene citronellal is 30–40%, geranial (citral *a*) and neral (citral *b*) are 10–30%; and sesquiterpenes (*b*-caryophyllene, germacrene D) (Wichtl and Bisset 1994; Bruneton 1995; Leung and Foster 1996). In agreement with our result Mimica-Dukic *et al.* (2004) reported that major chemical compounds are citrals (geranial and neral), citronellal accompanied by *b*-caryophyllene. These oil components are also responsible for the antibacterial and antifungal properties of the dried drug. The chemical composition of the essential oil of the lemon balm leaf (0.02-0.3% dry weight) has been studied. The major compounds were citronellal (2-40%) and citral (neral and geranial: 10-30%), accompanied by *b*-caryophyllene, germacrene D, ocimene and citronellol (Sarer and Kiikdil 1991; Adzet, *et al.* 1992; Kreis and Mosandl 1994). Many researchers have reported that the main components of lemon balm are neral and geranial. Neral and geranial rates in the oil were reported respectively as 15% and 14.5% by Hefendehl (1970), 19.6-36.1% and 25.3-47.5% by Tittel *et al.* (1982), 19.5% and 31.6% by Werker *et al.* (1985). Although all of discussed studies in above reported that neral and geranial were the main components of the oil, Kirimer *et al.* (1995) found that the main component of the lemon balm oil they studied was carvacrol (60%). Essential oil obtained from a few different populations of *Melissa officinalis* L. cultivated in Poland had been investigated by Patora *et al.* (2003). In their study, the content of essential oil in the leaves and herb were recorded as 0.08 to 0.25 ml/100 g and 0.06 to 0.167 ml/100 g, respectively. However, there were significant differences among the rates of those reported components. In conclusion, it is worthwhile to screen the commonly used plants from the local flora for different biological activities because they might present a new alternative source for possible bioactive substances.

Table 1. The percentage composition of identified compounds in the total oil from the *M. officinalis*.

| Compound | % |
|-------------------------|--------------|
| 1-Hepten-3-ol | 1 |
| 6-Methyl-5-hepten-2-one | 0.5 |
| Cis-b-ocimene | 0.4 |
| Trans-b-ocimene | 4.5 |
| Linalool | 2 |
| Nonanal | 1.56 |
| Citronellal | 3.21 |
| Neral (citral b) | 4.32 |
| Geranial (citral a) | 8 |
| a-Copaene | 3 |
| b-Caryophyllene | 16 |
| a-Bisabolene | 1 |
| b-Cubebene | 10.21 |
| γ -Elemene | 2.22 |
| b-Cedrene | 1.56 |
| Calarene | 4.32 |
| b-Cadinene | 4 |
| Sesquiterpene alcohol | 10.43 |
| Nerolidol | 1.23 |
| Cadinol isomer | 3.45 |
| a-Cadinol | 9.05 |
| Total | 91.96 |

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