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Original Article

Effects of different drying methods on the yield and the composition of essential oil from herb *Mentha longifolia* (L.) Hudson

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***Abstract*:**
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This paper discusses the impact of different methods of drying on the content and chemical composition of the essential oil from the herb *Mentha longifolia* (L.) Hudson. Drying of plant material was carried out naturally in the shade of draughty place, in the laboratory oven at the temperature 45°C and absorptive low temperature condensation drying oven at 35°C (low temperature drying). Isolation of essential oil from dried samples in three different ways was conducted by hydrodistillation, whilst chemical analysis was carried out by GC/FID and GC/MS methods. The highest yield of the essential oil was obtained from the herb which was dried at low temperature (1.1%) and the lowest from that dried in the laboratory oven (0.6%). The biggest content of the dominant component of essential oils, piperitone, was recorded in the oil from low temperature dried herb (71.7%), while those isolated from naturally dried drug and in from the laboratory oven contained piperitone in lower concentrations (50.8% and 43.1%, respectively).

Key words: drying, essential oil, *Mentha longifolia* L., piperiton

Introduction

Mentha longifolia (L) Hudson, (wild mint, English horsemint) belongs to the genus *Mentha*, Lamiaceae family. Inhabits wet habitats, the rivers, lakes, ponds and channels, wet meadows, forested terrain, and even drier places along the roads and arable land. It is widespread in Serbia except in Vojvodina, where it can be found only on Fruška Gora.

In South Serbia it is widespread from the regions of lowlands, where it can be found along the roads and on the banks of rivers, to the hilly, near the streams in the community Junco-Menthetum longifolia. It is widespread throughout the Mediterranean, Central and Northern Europe, Asia Minor, Africa (Janković, 1974; Stamenković, 1995).

Wild mint is an aromatic and melliferous plant. It is used in the pharmaceutical, tobacco, food industry (in the development of various liqueurs and sweets), and especially in cosmetology. Essential oil of English horsemint has pleasant and refreshing odour. According to the PDR exhibits carminative

and stimulant properties of the gastrointestinal tract. Relieves colds, respiratory inflammation, headaches, pain in muscles and joints. Internally it is used in the form of infusion, and externally as bath additive (PDR, 2004). There are phytopreparations on the basis of the active ingredients of essential oils of mint (menthol, menthone, pinene, and cineole) in the form of cream, the solution of rubbing (alcohol, oil), syrup, capsule, coated tablet (Rote Liste, 1997).

Chemical composition of the essential oil of wild mint herb is very variable depending on the habitat and climate where the species grow. Forty-five constituents were identified in the essential oil of *M. longifolia* from Turkey, with the cis-epoxy piperitone, pulegone and piperitenone oxide as main components, and studied oil exhibits strong antimicrobial activity (Gulluce et al., 2007). Analysis of essential oil of Moroccan *M. longifolia* showed interesting relative quantities of piperitenone oxide and piperitone oxide (Ghoulami et al., 2001). In the essential oil of wild mint from South Africa, 31 components were identified. Menthone (50.9%), pulegone (19.3%) and 1,8-cineole (11.9%) were the main ingredients of the oil (Oyedeji and Afolayan, 2006). Analysis of oil of *M. longifolia* from Italy and Israel revealed piperitenone oxide as the main component, while the essential oil from Sinai contained 1,8-cineole (28.8%), piperitone oxide (15.4%) and piperitone (13.8%) (Maffei, 1988; Fleisher & Fleisher, 1998). There is more then 70% of pulegone in the oil that grows in the desert of Jordan (Fleisher & Fleisher, 1991). The dominant components of essential oil of wild mint herb in Vojvodina are menthone, isomenthone and 1,8-cineole, and the oil exhibits a strong antimicrobial and significant fungicidal effect (Mimica et al., 2003). Wild mint from Croatia contains carvone, piperitenone oxide, limonene and β-caryophyllene as the main ingredients (Mastelić & Jerkovic, 2002). The main components of oil types which were collected on Zlatar are cis- and trans-dihidrocarvone (15.9% and 30.6%) (Matović & Lavadinović, 1999). Piperitone oxide was found as the main component in nine populations of *M. longifolia* essential oils from Greece (Kokkini & Papageorgiou, 1988).

Drying is the easiest way of preservation of raw plant material. Drying procedures are different and have an impact on the content of active substances in drugs.

In the process of drying the plant material moisture content is reduced, but the amount and composition of volatile compounds are changed, too (Moyler, 1994). The way of drying has a significant impact on qualitative and quantitative composition of essential oils of aromatic plants. This paper presents the results of the impact of different methods of drying on the yield and the composition of essential oil of *M. longifolia* herb originating from Southern Serbia.

Material and methods

Plant material

Plant material was collected in the flowering stage in July 2009 from the municipality of Prokuplje, at the Rastovnica village, on the mountain Pasjača. Samples of collected plants are deposited in the Herbarium HMD (Herbarium Moesiacum Doljevac) № 233. Plants were collected in the morning hours, in dry times and dried in three ways: 15 days in the shade of a draughty place, natural drying (ND), in the laboratory oven at the temperature of 45°C (LOD) and in the low temperature absorptive condensation dryer (NT-KS/60S) at temperature of about 35°C (LTD). The drying air used was of low humidity and a minimally heated.

Dried plant material was packed in paper bags and kept in a dry and cool place.

Isolation and determination of the content of the essential oil

Isolation of the essential oil from dried and cut *M. longifolia* herb was carried out by water distillation in the Clevenger type apparatus, according to procedure Ph. Jug. IV (Ph. Jug. IV, 1984). Distillation lasted about 2 hours at the boiling temperature. Pure essential oils were kept in dark glass ampoules at +4°C. These are colourless liquids, with the characteristic sharp odour.

Results of determination of the contents of the essential oils in the three studied samples of the herb *M. longifolia* represent the average value of three comparative analyses and refer to the dry samples.

Qualitative and quantitative analysis of the essential oils

Qualitative and quantitative analysis of essential oils was conducted by GC and GC/MS methods.

Analytical gas chromatography (GC/FID)

GC/FID analysis of the oils was carried out on a HP-5890 Series II GC apparatus [Hewlett-Packard, Waldbronn (Germany)], equipped with split-splitless injector and automatic liquid sampler (ALS), attached to HP-5 column (25 m · 0.32 mm, 0.52 μm film thickness) and fitted to flame ionisation detector (FID). Carrier gas flow rate (H_2) was 1 ml/min, split ratio 1:30, injector temperature

Table 1 The chemical composition of essential oils of herb *M. longifolia* dried by different procedures (%)

Constituents	KIE	KIL	Drying method		
			ND	LOD	LTD
α-pinene	932.8	932	0.8	0.6	0.6
sabinene	973.2	969	0.7	0.5	0.4
β-pinene	975.6	974	1.3	0.9	0.8
myrcene	993.0	988	0.7	1.2	0.5
3-octanol	1000.2	988	0.3	-	0.2
limonene	1029.0	1024	6.3	1.6	2.4
1,8-cineole	1030.9	1026	3.9	3.6	3.5
cis-β-ocimene	1039.6	1032	1.3	0.8	1.2
trans-β-ocimene	1049.2	1044	-	-	0.4
linalool	1102.8	1095	-	0.4	-
menthone	1154.6	1148	0.6	17.5	-
iso-menthone	1165.1	1158	-	8.3	-
ocimenol	1169.4	n/a	-	-	0.3
menthol	1175.3	1167	0.3	-	-
cis-isopulegone	1177.1	n/a	-	0.7	-
α-terpineol	1193.4	1186	1.1	0.3	0.5
cis-dihydro carvone	1198.2	1191	3.5	0.6	0.3
trans-dihydro carvone	1205.4	1200	-	2.3	1.1
neoiso-dihydro carveol	1231.8	1226	-	0.8	-
cis-3-hexenyl isovalerate	1238.8	1232	0.5	-	-
pulegone	1240.7	1233	-	1.4	-
carvone	1247.1	1239	20.0	2.9	5.0
piperitone	1257.7	1249	50.8	43.1	71.7
dihydroedulan I	1288.6	1289	-	0.3	-
piperitenone oxide	1369.0	1366	-	2.7	-
β-bourbonene	1385.1	1387	-	-	0.8
β-elemene	1392.9	1389	-	0.3	-
trans-caryophyllene	1420.1	1417	4.3	4.1	5.4
α-humulene	1454.1	1452	-	-	0.3
γ-muurolene	1482.3	1478	3.1	4.3	3.6
bicyclogermacrene	1497.6	1500	0.4	1.1	0.8
Sum of contents %			100.0	100.0	100.0
Number of constituents			18	24	20

KIE=Kovats (retention) index experimentally determined (AMDIS), KIL=Kovats (retention) index - literature data (A d a m s , 2007), n/a=not available, ND-natural drying , LOD-laboratory drying oven, LTD-low temperature drying

was 250°C, detector temperature 300°C, while column temperature was linearly programmed from 40-260°C (at rate of 4°/min). Solutions of essential oils (~1%) were consecutively injected by ALS (1 µl, split mode). Area percent reports, obtained as result of standard processing of chromatograms, were used as base for the quantification purposes.

Gas chromatography/mass spectrometry (GC/MS)

The same analytical conditions as those mentioned for GC/FID were employed for GC/MS analysis, along with column HP-5MS (30 m · 0.25 mm, 0.25 µm film thickness), using HP G 1800C Series II GCD system [Hewlett-Packard, Palo Alto, CA (USA)]. Instead of hydrogen, helium was used as carrier gas. Transfer line was heated at 260°C. Mass spectra were acquired in EI mode (70 eV), in m/z range 40-450. Solutions of the essential oils (~1%) were injected by ALS (200 nl, split mode).

The constituents were identified by comparison of their mass spectra to those from Wiley275 and NIST/NBS libraries, using different search engines. The experimental values for retention indices were determined by the use of calibrated Automated Mass Spectral Deconvolution and Identification System software (AMDIS ver.2.1.), compared to those from available literature

(Adams, 2007), and used as additional tool to approve MS findings.

Results and disscusion

The content of the essential oil from above-ground part of *M. longifolia*, which has been dried on the three described ways, was variable. The highest yield is obtained from the herb dried at low temperature (1.1%), and then the herb which was dried naturally (0.9%), while the lowest yield was obtained in the laboratory oven (0.6%). Chemical composition of essential oils is also variable (**Tab. 1**).

Twenty-four components were identified in the essential oil obtained from wild mint herb that was dried in the laboratory oven. Content of dominant component (piperitone) was 43.1%, along with 1,8-cineole, 3.6%, limonene 1.6%, menthone 17.5%, isomenthone 8.3%, carvone 2.9%, trans-caryophyllene 4.1% and γ-muurolene 4.3%.

In the essential oil obtained from the naturally dried herb 18 components were identified. Content of piperitone was 50.8%, limonene 6.3%, 1,8-cineole 3.9%, menthone only 0.6%, carvone 20.0 %, trans-caryophyllene 4.3%, γ-muurolene 3.1%. Isomenthone was not recorded.

In the essential oil from herb dried in low temperature oven 20 components were identified. Content of piperitone was 71.7%, 1,8-cineole 3.5%, limonene 2.4%, carvone 5.0%, trans-caryophyllene 5.4% and γ-muurolene 3.6%. Menthone and isomentone were not registered.

Changes in the concentrations of volatile compounds during drying depend on several factors, such as drying methods and classes of plants. Mint belongs to the family Lamiaceae, which is known to have storage of essential oil on or near the surface of the leaf (Moyler, 1994).

Pulegone has been proved to be a powerful hepatotoxin, even at low concentrations. It is metabolized by the liver to menthofuran, which is highly reactive metabolite, and it has adverse effect on the liver (Chen et al., 2001, Gordon et al., 1987), and it can also destroy the rat cytochrome P450 (Moorthi, 1991). In our tests we can find pulegone only in the oil which was isolated from wild mint herb dried in laboratory oven.

Conclusion

From the results it can be concluded that the highest essential oil content was found in plant material dried at low temperature, then dried naturally, and the lowest content was found in the plant material dried in laboratory oven. The content of the major constituent, piperitone, is reduced in the same order (71.7%, 50.8%. 43.1%). Pharmacologically active menthol, cineole, limonene and pinene are the most represented in the oil from the naturally dried herb.

So, drying of plant material for isolation of essential oils at low temperatures is could be marked as the best method. Simultaneously, draying in the natural way is quite acceptable. Pulegone is present only in the oil obtained from the dried herb in a laboratory oven, and from the security aspects this method of drying should be avoided.

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