Iranian Journal of Pharmaceutical Research (2003) 103-105
Received: September 2002
Accepted: May 2003

Original Article

Quantity and Composition of the SDE Prepared Essential Oil of *Nepeta macrosiphon* Boiss

Alireza Ghannadi*[a], Fatemeh Aghazari[a], Mitra Mehrabani[a], Abdolali Mohagheghzadeh [b], Iraj Mehregan[b].

[a]*Department of Pharmacognosy, School of Pharmacy and Pharmaceutical Sciences, Isfahan University of Medical Sciences and Health services, Isfahan, Iran.* [b]*Department of Pharmacognosy, School of Pharmacy, Shiraz University of Medical Sciences and Health services, Shiraz , Iran.*

Abstract

The essential oil from flowering aerial parts of *Nepeta macrosiphon* Boiss. growing wild in Kermanshah Province, Iran, was analyzed by GC/MS. This essential oil was prepared by a modified Likens-Nickerson's simultaneous distillation-extraction (SDE) method. Forty-five compounds consisting 95.1% of the total components were identified from the oil obtained with a yield of 0.1%w/w. Among them, spathulenol (14.1%), germacrene D (9.2%) and caryophyllene oxide (8.1%) were the major components of the oil.

Keywords: *Nepeta macrosiphon*; Lamiaceae; Essential oil composition; GC/MS; Spathulenol; Germacrene D; Caryophyllene oxide.

Introduction

The genus *Nepeta,* also called *Glecho ma* and *Cataria*, is named after the ancient Italian city of Nepi (1). This genus which belongs to Stachyoideae-Nepeteae tribe, Lamiaceae family, consists of about 250 species distributed in the central and southern parts of Europe, Asia and Middle East (2, 3). Many reports on phytochemical analysis of this genus, including essential oil analysis, are found in the literature (4-27). Most oils of *Nepeta* species contain nepetalactones as the main components, but some differences in the essential oil composition were detected in several *Nepeta* oils (15-27). Antibacterial, fungicidal, antiviral and opioid analgesic activities have been attributed to nepetalactones (19, 21). *Nepeta* species are still used in the traditional medicine of many countries as diuretic, diaphoretic, vulnerary, antitussive, antispasmodic, anti-asthmatic, tonic, febrifuge, emmenagogue and sedative agents (22, 27, 28). Some of Iranian *Nepeta* species has been of great interest to Iranian folk and traditional medicines and used in the treatment of various disorders, such as some nervous, respiratory and gastrointestinal diseases (27, 29).

The Iranian flora comprises 67 species of *Nepeta* and one of them is *Nepeta macrosiphon* Boiss (2, 30). This herb distributed in different rocky western areas of Iran (2). The Persian names of the plant are "punesaye sisakhti" and "punesaye lulehboland" (30). Our literature surveys revealed that the essential oil of the aerial parts of *N. macrosiphon* has not been chemically studied to date, therefore this article deals with the detailed quantity and composition of the SDE oil prepared by GC/MS.

Experimental

Plant Material

The aerial parts of wild-growing *N. macrosiphon* were collected during the flowering period from northern slopes of Dalakhani mountain, Songhur (Kermanshah Province, Iran) at an altitude of ca. 2300 m in

* Corresponding author:
E-mail: ghannadi@pharm.mui.ac.ir

June 2001. The plant identity as *N. macrosiphon* was confirmed by the Herbarium Department of the Faculty of Pharmacy, Shiraz University of Medical Sciences, Shiraz, Iran. A voucher specimen of the plant was deposited in the Herbarium of the Department of Pharmacognosy, School of Pharmacy and Pharmaceutical Sciences, Isfahan University of Medical Sciences, Isfahan, Iran.

The air-dried aerial parts of the plant were powdered and the volatile fraction was prepared by a modified Likens-Nickerson's simultaneous distillation and extraction (SDE) method (31, 32). A microscale simultaneous distillation extraction apparatus (Ashke Shishe, Tehran, Iran) was used. Dried powdered plant was homogenized with distilled water and the homogenate subjected to SDE apparatus for 3 h using pentane (chromatography grade reagent, Merck) as solvent and then extract was concentrated with nitrogen.

Table 1. Components present within the oil obtained from the aerial parts of *Nepeta macrosiphon.*

No.	Compound	%	RI
1	trans-2-hexenal	trace	850
2	alpha-pinene	0.3	936
3	camphene	0.1	951
4	beta-pinene	0.1	978
5	1,8-cineole	0.2	1030
6	trans-beta-ocimene	0.3	1048
7	gamma-terpinene	1.0	1058
8	cis-sabinene hydrate	0.1	1065
9	linalool oxide B	0.1	1071
10	linalool oxide A	0.4	1086
11	alpha-pinene oxide	0.3	1094
12	linalool	4.1	1097
13	trans-pinocarveol	0.4	1137
14	trans-verbenol	1.0	1142
15	borneol	0.2	1165
16	terpinene-4-ol	0.5	1176
17	myrtenol	2.0	1194
18	verbenone	4.4	1204
19	geraniol	0.5	1253
20	bornyl acetate	1.1	1283
21	citronellyl acetate	3.8	1350
22	alpha-copaene	0.4	1375
23	beta-bourbonene	1.3	1384
24	longifolene	1.0	1401
25	aromadendrene	4.8	1438
26	germacrene D	9.2	1481
27	bicyclogermacrene	5.7	1495
28	alpha-muurolene	6.0	1497
29	germacrene A	2.4	1501
30	gamma-cadinene	1.0	1512
31	delta-cadinene	2.3	1523
32	alpha-cadinene	3.0	1538
33	spathulenol	14.1	1580
34	caryophyllene oxide	8.1	1581
35	gamma-eudesmol	3.9	1633
36	torreyol	2.0	1642
37	alpha-cadinol	4.9	1657
38	cadalene	0.4	1679
39	caryophyllene acetate	0.4	1705
40	khusimol	0.2	1740
41	gamma-eudesmol acetate	1.0	1782
42	n-octadecane	0.5	1804
43	pentadecanoic acid	0.3	1869
44	`n-nonadecane	0.2	1907
45	methyl octadecanoate	1.1	2139

GC/MS Analysis

The oil was analyzed by GC/MS using a Hewlett Packard 6890 mass selective detector coupled with a Hewlett Packard 6890 gas chromatograph, equipped with a cross-linked 5% PH ME siloxane HP-5MS capillary column (30 m × 0.25 mm, film thickness 0.25 μm). Operating conditions were as follows: carrier gas, helium with a flow rate of 2 ml/min; column temperature, 60-275ºC at 4ºC/min; injector and detector temperatures, 280ºC; volume injected, 0.1 μl of the oil; split ratio, 1:50.

The MS operating parameters were as follows: ionization potential, 70 ev; ionization current, 2 A; ion source temperature, 200ºC; resolution, 1000.

Identification of components in the oil was based on retention indices relative to *n*-alkanes and computer matching with the WILEY 275.L library, as well as by comparison of the fragmentation patterns of the mass spectra with those reported in the literature (33-36).

Results and Discussion

Aerial parts of *N. macrosiphon* yielded 0.1% (w/w *vs* dried material) of a pale yellowish oil with a strong pleasant aroma. Forty-five components were characterized, representing 95.1% of the total oil compounds detected. These are listed in Table 1 with their percentage composition. The major constituents of the sesquiterpene-rich oil of *N. macrosiphon* were spathulenol (14.1%), germacrene D (9.2%), caryophyllene oxide (8.1%), alpha-muurolene (6.0%) and bicyclogermacrene (5.7%). Other components were present in amounts less than 5%. Many of the unidentified compounds were present in trace amounts.

Although the presence of nepetalactones in several *Nepeta* species in relatively high concentrations has been reported (15-27), no nepetalactones were found in this oil. The predominance of spathulenol and caryophyllene oxide has been found in essential oils of two Turkish *Nepeta* species (9, 12). These compounds and germacrene D are typical in most *Nepeta* species (4-27).

Acknowledgments

This work was supported by Research Council of the Isfahan University of Medical Sciences, Isfahan, Iran (Research project No. 80256). We are grateful to Dr. Javad Asili (Department of Pharmacognosy, Faculty of Pharmacy and Pharmaceutical Sciences, Isfahan University of Medical Sciences, Isfahan , Iran) for his assistance in preparing the plant essential oil.

References

(1) Simonovic D. *Botanical Dictionary*, Vol. 3. Institute for Serbo-Croatian Language, Belgrade (1959) No. CCCXVIII

(2) Rechinger KH. *Flora Iranica*, No. 150. Akademische Druck-u. Verlagsanstalt, Graz (1982) 2, 108-216.

(3) Evans WC. *Trease and Evans' Pharmacognosy*, 13th ed. Bailliere Tindall, London (1989) 217

(4) Baser KHC, Ozek T, Akgul A and Tumen G. Composition of the essential oil of *Nepeta racemosa* Lam. *J. Essent. Oil Res*. (1993) 5: 215-217

(5) Kokdil G, Tanker M, Kurucu S and Topcu G. Essential oil analysis of *Nepeta cilicia* Boiss. *Flavour Fragr. J.* (1997) 12: 99-101

(6) Baser KHC, Demircakmak B and Duman H. Composition and the essential oils of *Nepeta sulfuriflora* P.H. Davis. *J. Essent. Oil Res.* (1998) 10: 179-181

(7) Baser KHC, Demircakmak B, Altinas A and Duman H. Essential oil of *Nepeta flavida* Hub.-Mor. *J. Essent. Oil Res.* (1998) 10: 299-300

(8) Chalcat JC, Petrovic SD and Gorunovic MS. Quantity and composition of essential oil of the wild plant *Nepeta nuda* L. from Yugoslavia. *J. Essent. Oil Res.* (1998) 10: 423-425

(9) Kokdil G, Kurucu S and Yildiz A. Essential oil composition of *Nepeta nuda* ssp. *nuda*. *Flavour Fragr. J.* (1998) 13:233-234

(10)Sajjadi SE and Ghassemi N. Volatile constituents of *Nepeta glomerulosa* Boiss. subsp. *carmanica*. *Flavour Fragr. J.* (1999) 14: 265-267

(11)Baser KHC, Ozek T, Yildiz B, Bahcecioglu Z and Tumen G. Composition of the essential oil of *Nepeta fissa* C.A. Meyer. *J. Essent. Oil Res.* (2000) 12: 27-28

(12)Baser KHC, Ozek T, Bemirci B and Tumen G. Composition of the essential oil of *Nepeta betonicifolia* C.A. Meyer from Turkey. *J. Essent. Oil Res.* (2001) 13: 35-36

(13)Sajjadi SE and Khatamsaz M. Volatile constituents of *Nepeta heliotropifolia* Lam. *J. Essent. Oil Res.* (2001) 13: 204-205

(14)Sefidkon F. Essential oil of *Nepeta glomerulosa* Boiss. from Iran. *J. Essent. Oil Res*. (2001) 13: 422-423

(15)Matloubi Moghaddam F and Hosseini M. Composition of the essential oil from *Nepeta crassifolia* Boiss. & Buhse. *Flavour Fragr. J.* (1996) 11: 113-115

(16)Kokdil G, Kurucu S and Topcu G. Composition of the essential oil of *Nepeta nuda* L. ssp. *albiflora* (Boiss.) Gams. *Flavour Fragr. J.* (1996) 11: 167-169

(17)Kokdil G, Kurucu S and Topcu G. Chemical constituents of the essential oils of *Nepeta italica* L. and *Nepeta sulfuriflora* P.H. Davis. *Flavour Fragr. J.* (1997) 12: 33-35

(18)Baser KHC, Demircakmak B, Altinas A and Duman H. Composition of the essential oils of *Nepeta cadmea* Boiss. *J. Essent. Oil Res.* (1998) 10: 327-328

(19)Aydin S, Beis R, Ozturk Y and Baser KHC. Nepetalactone- a new opioid analgesic *from Nepeta caesarea* Boiss. *J. Pharm. Pharmacol.* (1998) 50: 813-817

(20)Rustaiyan A and Nadji K. Composition of the essential oils of *Nepeta ispahanica* Boiss. and *Nepeta binaludensis* Jamzad from Iran. *Flavour Fragr. J*. (1999) 14: 35-37

(21)Skaltsa HD, Lazari DM, Loukis AE and Constantinidis T. Essential oil analysis of *Nepeta argolica* Bory & Chaub. Subsp. *argolica* (Lamiaceae) growing wild in Greece. *Flavour Fragr. J.* (2000) 15: 96-99

(22)Tzakou O, Harvala C, Galati EM and Sanogo R. Essential oil composition of *Nepeta argolica* Bory et Chaub. Subsp. *argolica. Flavour Fragr. J.* (2000) 15: 115-118

(23)Rustaiyan A, Khosravi M, Larijany K and Masoudi S. Composition of the essential oil *of Nepeta racemosa* Lam. from Iran. *J. Essent. Oil Res.* (2000) 12: 151-152

(24)Chalcat JC, Gorunovic MS, Petrovic SD and Maksimovic ZA. Composition of the essential oil of *Nepeta rtanjensis* Diklic et Milojevic, Lamiaceae from Serbia. . *J. Essent. Oil Res.* (2000) 12: 238-240

(25)Rustaiyan A, Komeilizadeh H, Monfared A, Nadji K, Masoudi S and Yari M. Volatile constituents of *Nepeta denudata* Benth. and *N. cephalotes* Boiss. from Iran. *J. Essent. Oil Res.* (2000) 12: 459-461

(26)Thappa RK, Agarwal SG, Srivastava TN and Kapahi BK. Essential oils of four Himalayan *Nepeta* species. *J. Essent. Oil Res.* (2001) 13: 189-191

(27)Zargari A. *Medicinal Plants.* Vol. 4. Tehran University Publications, Tehran (1990) 106-112

(28)Rapisarda A, Galati EM, Tzakou O, Flores M and Miceli N. *Nepeta sibthorpii* Bentham (Lamiaceae)-micromorphological analysis of leaves and flowers. *Il Pharmaco* (2001) 56: 413-415

(29)Amin GR. *Popular Medicinal Plants of Iran*, Vol. 1. Ministry of Health Publications, Tehran (1991) 40-41,55

(30)Mozaffarian V. *Dictionary of Iranian Plant Names.* Farhang-e Moaser, Tehran (1996) 360-364

(31)Filek G, Bergamini M and Lindner W. Steam distillation-solvent extraction, a selective sample enrichment technique for the gas chromatographic- electron- capture detection of organochlorine compounds in milk powder and other milk products. *J. Chromatogr.* (1995) 712: 355-364

(32)Linskens HF and Jackson JF. Plant Volatile Analysis. Springer, Berlin (1997) 231-234

(33)Adams RP. Identification of Essential Oil Components by Gas Chromatography / Mass Spectroscopy. Allured Publishing Co., Carol Stream (1995)

(34)Mclafferty FW and Stauffer DB. The Important Peak Index of the Registry of Mass Spectral Data. John Wiley & Sons, Inc., New York (1991)

(35)Sandra P and Bicchi C. Capillary Gas Chromatography in Essential Oil Analysis. Dr. A. Huethig, Heidelberg (1987)

(36)Swigar AA and Silverstein RM. Monoterpenes- Infrared, Mass ,Proton-NMR, Carbon-NMR Spectra and Kovats Indices. Aldrich Chemical Company Inc., Wisconsin (1981)