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Essential Oil Compounds of Three *Nepeta* L. Taxa From Turkey and Their Chemotaxonomy

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The essential oil aerial parts of *Nepeta baytopii* Hedge and Lamond., *Nepeta cataria* L. and *Nepeta fissa* C.A. Mey. were investigated by GC and GC-MS. The yield of oils are *ca*. 0.40, 0.45 and 0.50 mL/100 g, respectively. Forty six, fourty seven and forty nine compounds were identified representing 92.4, 91.2 and 92.5 % of the oil, respectively. 1,8-Cineole (23.2 %) and nepetalactone (12.8 %) in *N. baytopii*, nepetalactone (27.5 %) - 1,8-cineole (10.8 %) and germacrene D (9.2 %) in *N. cataria*, 1,8-cineole (24.3 %) and nepetalactone (17.6 %) were identified as major components in *N. fissa*. The chemical distribution of the essential oil compounds in the genus pattern were discussed in means of chemotaxonomy.

Key Words: *Nepeta*, Lamiaceae, Essential oil, Chemotaxonomy.

INTRODUCTION

The genus *Nepeta* L. belongs to the family Lamiaceae, rarely annual, perennial and often pleasantly aromatic herbs found in temperate Europe, Asia, North Africa, in mountains of tropical Africa and comprises of approximately 250 species[1]. *Nepeta* represented in Turkey by 40 taxa, 16 of them are endemic (*ca*. 40 %)[2-4]. Stems erect or procumbent, eglandular or glandular.

External nutlet characters are very important in the Iranian and Afghan species, but of limited taxonomic value in Turkey. However, detailed anatomical investigation of the pericarp might well yield useful new information. The existing infra-generic classifications are extremely unsatisfactory. In this account *Nepeta* have not recognised any sections but have placed the species in three informal groups (designated A, B and C) based largely on flower colour and inflorescence characters. *N. cataria* belongs to group A, *N. baytopii* and *N. fissa* are belongs to group B[2]. *Nepeta baytopii* is an endemic species with limited distribution and included in the lower risk and least concern category in the red data book of Turkey[5].

Some of these species are well known for their medicinal properties and biological activities. They are used in the folk medicine for their diuretic, diaphoretic, antitussive, antispasmodic, anti-asthmatic, febrifuge, emmenagogue, sedative and stomachic properties. Pharmacological and biological effects are usually attributed to nepetalactones, especially found in *Nepeta* oils. Considering their essential oil compositions, *Nepeta* species can be divided into two groups, *i.e.*, nepetalactone-containing and nepetalactone-less species[6]. In the literature, there are chemical studies of essential oils of some *Nepeta* taxa (Table-2). In continuation of chemical composition of essential oils obtained from various *Nepeta* taxa, we now report on the chemical composition of three species of *Nepeta* and their chemotaxonomy.

EXPERIMENTAL

Samples were collected from their natural habitats. *N. cataria* (ÖK-3360) was collected in an island which behind the Atatürk dam wall, from Adiyaman/Turkey, on June 2011 at an altidude of 1100-1200 m. *N. baytopii* (BIN-4) was collected from Bingöl, south of Genç, Samdagi on september 2011 at an altitude of 1600-2100 m. *N. fissa* (BIN- 55) was collected from 15-20 km west of Bingöl-Asagiköy steppe, on August 2011, at an altitude of 1400-1500 m. The voucher specimens have been deposited at the Herbarium of department of Biology, Bingol and Firat University.

Isolation of the essential oil: Air-dried aerial parts of the plant materials were subjected to hydrodistillation using a Clevenger-type apparatus for 3 h.

Gas chromatographic (GC) analysis: The essential oil was analyzed using HP 6890 GC equipped with and FID detector and an HP-5 MS column (30 m × 0.25 mm i.d., film tickness 0.25 μm) capillary column was used. The column and analysis conditions were the same as in GC-MS. The percentage composition of the essential oils was computed from GCFID peak areas without correction factors.

Gas chromatography/mass spectrometry (GC-MS) analysis: The oils were analyzed by GC, GC-MS, using a Hewlett Packard system. HP-Agilent 5973 N GC-MS system with 6890 GC in Plant Products and Biotechnology Research Laboratory (BUBAL) in Firat University. HP-5 MS column (30 m × 0.25 mm i.d., film tickness (0.25 µm) was used with helium as the carrier gas. Injector temperature was 250 ºC, split flow was 1 mL/min. The GC oven temperature was kept at 70 ºC for 2 min and programmed to 150 ºC at a rate of 10 ºC/min and then kept constant at 150 ºC for 15 min to 240 ºC at a rate of 5 ºC/min. Alkanes were used as reference points in the calculation of relative retention indices (RRI). MS were taken at 70 eV and a mass range of 35-425. Component identification was carried out using spectrometric electronic libraries (WILEY, NIST).

RESULTS AND DISCUSSION

The chemical composition essential oil of dried aerial parts of *N. baytopii, N. cataria* and *N. fissa* were analyzed by GC and GC-MS. 46, 47 and 49 compounds were identified in *N. baytopii, N. cataria* and *N. fissa*, respectively, accounting from 92.4, 90.2-92.5 % of the whole oil. Altogether, 59 compounds have been identified. The yield of oils are *ca*. 0.40, 0.50 and 0.45 mL/100 g, respectively. 1,8-Cineole (23.2 %), nepetalactone (12.8 %) in *N. baytopii*, nepetalactone (27.5 %), 1,8-cineole (10.8 %) and germacrene D (9.2 %) in *N. cataria*, while 1,8-cineole (24.3 %)-nepetalactone (17.6 %), were identified as major components in *N. fissa*. The chemical composition of the essential oils of the three *Nepeta* species were reported in Table-1 and altogether 59 compounds have been identified and the main constituents of *Nepeta* taxa from literature and studied samples were listed in Table-2. Among the sesquiterpenes, β-caryophyllene was found principal constituents of *N. curviflora* (41.6 %) and *N. oxyodonta* (12.6 %). On the other hand, β-caryophyllene determined low amounts or not detected in many samples of *Nepeta* taxa (Table-2). Among the sesquiterpenes germacrene D was found high percentage of *N. govaniana* (11.5 %) and *N. involucrata*. Whereas this compound was not detected in *N. menthoides*, *N. crispa, N. nuda* subsp. *albiflora*, *N. denudata, N. cephalotes*, *N. floccosa* and *N. frachonitica*. Nepetalactone detected as the main compound in the essential oil of *N. cephalotes* (35.1 %), *N. govaniana* (25.9 %), *N. fissa* (17.6 %), *N. baytopii* (12.8 %) and *N. cataria* (27.5 %). Whereas nepetalactone was not determined in the oil of *N. nuda* subsp. *nuda*, *N. crispa, N. menthoides*, *N. mahanensis*, *N. ispahanica, N. denudata*, *N. floccosa*, *N. discolor, N. oxyodonta, N. satureioides*, *N. frachonitica* and *N. involucrata.* Moreover; nepetalactone was reported minor amount in *N. eremophila*, *N. rivularis, N. curviflora* and *N. nuda* subsp. *albiflora*. *N. satureioides* was characterized by high content of linalool (23.8 %) and no percentages of germacrene D and nepetalactone. It is noteworthy that linalool was detected high amount (23.8 %) only *N. satureioides* than other twenty four *Nepeta* taxa.

Among monoterpenes 1,8-cineole was found high percentage of almost all *Nepeta* taxa, except *N. curviflora*, *N. nuda*

TABLE-1
CHEMICAL PROFILES OF *Nepeta* TAXA. (%)

Compounds	RRI	*N. baytopii*	*N. cataria*	*N. fissa*
α-Thujene	925	–	0.4	0.2
α-Pinene	935	3.1	2.5	2.2
β-Pinene	970	3.8	2.1	3.2
Myrcene	985	0.2	0.1	–
α-Terpinene	1008	0.3	–	0.4
p-Cymene	1015	1.2	0.7	0.5
1,8-Cineole	1025	23.2	10.8	24.3
β-Ocimene	1036	–	0.2	0.3
cis-Sabinene hydrate	1039	0.2	0.1	–
γ-Terpinene	1045	0.4	0.3	0.5
Sabinene	1055	2.6	–	3.2
Linalool	1080	0.1	3.6	0.2
Nonanal	1098	–	0.1	0.3
trans-Pinocarveol	1128	1.2	–	1.4
Pinocarvone	1132	–	0.3	0.4
Borneol	1146	0.1	0.3	–
Terpinen-4-ol	1165	0.4	–	0.6
Myrtenal	1170	0.1	0.2	0.3
Myrtenol	1175	–	2.1	1.4
α-Terpineol	1185	3.1	5.3	2.8
Camphor	1186	–	1.3	0.6
Verbenone	1208	0.2	–	0.4
Pulegone	1240	0.2	0.3	0.1
2-Cyclohexen-1-one	1250	0.9	1.1	–
Geraniol	1265	–	0.2	–
Nepetalactone	1320	12.8	27.5	17.6
Bicycloelemene	1335	–	1.3	0.5
Geranylacetate	1342	1.2	–	1.3
β-Elemene	1350	0.5	2.5	1.0
α-Copaene	1365	–	0.4	0.3
β-Bourbonene	1366	0.1	0.2	–
β-Cedrene	1419	1.1	–	0.3
β-Caryophyllene	1420	5.6	5.5	9.2
β-Gurjunene	1428	2.5	1.2	–
Germacrene D	1435	4.5	9.2	5.4
(*Z*)-β-Farnesene	1449	–	2.6	3.1
α-Humulene	1450	0.1	–	0.3
Spathulenol	1455	3.9	2.8	3.0
Aromadendrene	1459	0.4	0.2	–
δ-Cadinene	1458	–	0.1	0.2
β-Copaene	1470	0.2	–	0.4
B-Sesquiphellandrene	1475	0.3	1.1	0.4
Bicyclogermacrene	1480	0.8	0.5	0.3
β-Selinene	1484	0.4	0.3	–
Caryophyllene oxide	1498	3.2	2.4	6.5
β-Bisabolene	1510	0.6	0.2	0.3
Muurolene	1512	0.2	1.6	0.7
γ-Cadinene	1514	1.8	0.3	1.3
Nerolidol	1520	0.2	–	0.6
δ-Cadinene	1524	1.5	0.6	1.9
α-Farnesene	1544	–	0.4	–
Ledol	1566	0.7	–	0.2
Caryophyllene oxide	1580	1.2	0.6	1.7
Longifolene	1585	2.2	1.3	0.5
Globulol	1590	1.1	0.4	0.1
β-Eudesmol	1610	0.8	0.3	0.2
Cadalene	1650	–	0.1	0.2
α-Cadinol	1660	3.1	0.9	1.4
Farnesol	1676	0.1	–	0.3
Total		92.4	91.2	92.5

TABLE-2
MAIN CONSTITUENTS OF *Nepeta* TAXA FROM LITERATURE AND STUDIED SAMPLES (%)

Main constituents	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
β-Caryophyllene	0.2	–	–	t	1.0	–	0.1	t	41.6	1.6	–	–	0.5	0.9	2.5	3.0	12.6	0.3	6.6	–	0.1	5.6	5.5	19.2
Germacrene D	0.6	–	–	–	6.5	0.5	0.1	1.9	0.7	–	–	–	–	0.4	20.5	0.4	7.4	0.9	–	7.4	15.1	4.5	8.2	5.4
Nepetalactone	–	–	–	10.3	–	–	2.6	2.4	5.7	3.6	–	35.1	–.	–	25.9	3.0	–	1.9	–	–	–	12.8	27.5	17.6
Caryophyllene oxide	–	–	–	t	3.4	0.8	–	–	9.5	6.9	–	–	0.6	1.3	0.5	0.4	5.3	0.8	6.4	1.1	2.2	3.2	2.4	6.5
Sabinene	2.8	–	2.4	0.7	–	0.4	0.2	14.8	–	–	–	2.0	0.7	0.6	–	t	1.6	0.6	–	4.1	6.7	2.6	–	3.2
Linalool	–	1.4	0.9	1.6	–	–	–	2.8	2.2	–	–	–	3.2	6.3	1.6	0.5	0.3	0.3	23.8	0.7	0.1	0.1	3.6	0.2
Spathulenol	–	–	0.1	–	–	0.5	–	t	3.9	2.7	–	–	–	–	–	–	8.5	–	1.0	22.2	2.3	3.9	2.8	3.0
Mrycene	–	–	–	–	–	–	–	–	–	–	–	–	0.2	10.7	1.9	t	0.2	0.3	t	–	1.4	0.2	0.1	–
α-Terpineol	–	4.1	5.7	3.3	1.4	1.0	0.5	3.6	0.8	t	1.4	–	0.9	0.5	–	0.4	0.3	0.6	2.5	0.6	3.3	3.1	5.3	2.8
α-Pinene	0.6	1.8	0.2	1.2	1.6	1.2	0.3	2.5	–	–	1.7	2.1	3.4	18.5	3.6	1.3	3.2	0.2	–	0.5	4.9	3.1	2.5	2.2
1,8-Cineole	21.0	71.0	41.1	62.8	27.2	71.7	13.1	38.5	0.1	2.1	48.0	11.4	–	3.0	t	75.0	3.3	39.8	0.3	–	23.1	23.2	10.8	24.3
β-Pinene	0.5	5.0	5.6	3.6	4.3	4.2	1.2	10.7	–	–	4.6	18.2	2.0	12.6	t	1.0	1.4	1.2	–	1.9	12.2	3.8	2.1	3.2

1. *N. nuda* subsp. *nuda*[7], 2. *N. crispa*, 3. *N. menthoides*[8], 4. *N. crispa*, 5. *N. mahanensis*, 6. *N. ispahanica*, 7. *N. eremophila*, 8. *N. rivularis*[9], 9. *N. curviflora*, 10. *N. nuda* subsp. *albiflora*[10], 11. *N. denudata*, 12. *N. cephalotes*[11], 13. *N. floccosa*, 14. *N. discolor*, 15. *N. govaniana*, 16. *N. royleana*[12], 17. *N. oxyodonta*[13], 18. *N. argolica* subsp. a*rgolica*[6], 19. *N. satureioides*[14], 20. *N. frachonitica*[15], 21. *N. involucrata*[16], 22. *N. baytopii*, 23. *N. cataria*, 24. *N. fissa* (studied samples).

subsp. *albiflora, N. floccosa*, *N. discolor*, *N. govaniana*, *N. satureioides*, *N. frachonitica* (Table-2). This compound was determined high amount in our samples with *N. baytopii* (23.2 %), *N. cataria* (10.8 %) and *N. fissa* (24.3 %) (Table-1). It is noteworthy that spathulenol was detected high amount only in *N. frachonitica* (22.2 %) (Table-2). *N. baytopii* and *N. fissa* are belongs to group B[2], in this study their major components (1,8-cineole and nepetalactone) were similar, so we can say that our results corralate with groups differences. Whereas *N. cataria* which belongs to group A, showed different behaviour from *N. baytopii* and *N. fissa*: its essential oil resulted composed by a high percentage of germacrene D (9.2 %).

Conclusion

These species synthesized many similar compounds in their essential oils that could be justified by the similar ecological conditions of their habitat (biochemical convergence). However, taking into account the differences referred to some constituents, also the taxonomic distance of these species could be confirmed by our chemical data. The comparison between the three taxa evidenced a similarity, at least with reference to the presence of the main constituents: in fact 1,8-cineole, nepetalactone and germacrene D were among the principal one in both studied taxa. The percentages of some components were comparable. The comparison between three species evidenced a similarity, at least with reference to the presence of main constituents *i.e.*, nepetalactone, 1,8-cineole, β-caryophyllene and germacrene D. Also the percentages of some constituents were comparable. This study demonstrates the occurrence of 1,8-cineole/nepetalactone chemotype in *N. baytopii*, *N. cataria* and *N. fissa* from Eastern Anatolian region of Turkey. Some of the *Nepeta* species showed different chemotype, like β-caryophyllene in *N. curviflora,* germacrene D and 1,8-cineole in *N. involucrata,* carvacrol in *N. glomerata*[17], germacrene D and nepetalactone in *N. govaniana,* 1,8-cineole in *N. nuda* subsp. *nuda*, *N. crispa, N. menthoides, N. mahanensis, N. ispahanica, N. eremophila*, *N. rivularis, N. curviflora*, *N. nuda* subsp. *albiflora, N. denudata, N. royleana* (Table-2).

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