## FATTY ACID COMPOSITION OF THE SEEDS OF FIVE Nepeta SPECIES FROM TURKEY

N. Akpinar, M. A. Akpinar, S. Gorgun, T. Dirmenci, and A. Aktumsek

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The genus *Nepeta* (Lamiaceae) comprises about 300 species, most of which grow wild in Central and Southern Europe, North Africa, and Central and Southern Asia [1–3]. Recent studies have indicated that *Nepeta* species in Turkish flora are represented by 44 taxa, of which 22 are endemic to Turkey, and endemic and non-endemic species mostly grow in East Anatolia and the Taurus Mount [4].

Nepeta species appears to be widely used in folk medicine [5], and analgesic, antifungal, and antiviral activities from Nepeta species have been shown [6–8]. Moreover, oil from Nepeta species has toxic effects on insects [9]. At the same time, one important feature of Nepeta species is that they may also exhibit antimicrobial activity against Helicobacter pylori [10].

In spite of many works on the chemical constituents of the genus *Nepeta*, there is no available data on the fatty acid composition of this genus. Consequently the fatty acid dynamics of this genus is not well known, and this work aims to establish the fatty acid patterns of some *Nepeta* species from Turkish flora.

The *Nepeta* species used in this study were *N. transcaucasica* Grossh., *N. italica* L., *N. isaurica* Boiss. & Heldr. (endemic to Turkish flora), *N. caesarea* Boiss. (endemic to Turkish flora), and *N. nuda* L. ssp *albiflora* Boiss. The species were collected from various locations in Turkey.

The air-dried seed material was ground. From each of the milled samples, 1 g was taken and stored in chloroform/methanol (2/1, v/v) for 48 h at 4°C. Weighed samples were homogenized in chloroform-methanol (2:1, v/v) using an Ultra-Turrax T25 homogenizer in an ice bath, and isolation of total lipids from seeds was carried out [11]. The total lipids obtained were saponified by refluxing with methanol (50%) containing 5% sodium hydroxide for 1 h. The saponifiable lipids were converted to their methyl esters using the standard boron trifluoride-methanol (BF<sub>3</sub>) method [12].

The resultant mixture of fatty acid methyl esters (FAMEs) in hexane–chloroform (4:1, v/v) was injected onto a HP (Hewlett Packard) Agilent 6890 N model gas chromatograph (GC), equipped with a flame ionization detector (FID) and fitted with a DB-23 capillary column (60 m, 0.25 mm i.d. and 0.25  $\mu$ m). Injector and detector temperatures were 270 and 280°C, respectively. Column temperature program was 190°C for 35 min, then increasing at 30°C/min up to 220°C, where it was maintained for 5 min. Carrier gas was helium (2 mL/min) and split ratio was 30:1.

Identification of normal fatty acids was carried out by comparing the peak relative retention times of the sample FAMEs with those obtained for Alltech standards (Lexington, USA). Results were expressed as FID response area relative percentages. Each reported result is the average value of three GC analyses.

The statistical analyses were performed using a commercial statistical program (SPSS 8.0). All analytical determinations were performed in triplicate, and the mean values were reported. The statistical analyses of percentages of FAMEs were tested by analysis of variance (ANOVA), and comparisons between means were performed with Tukey's test. Differences between means were evaluated as significant if  $p \le 0.05$ .

The fatty acid compositions of different species of *Nepeta* are presented in Table 1. Seven fatty acid methyl esters were identified from the seeds of *N. italica* and *N. isaurica*, while from the seeds of *N. transcaucasica*, *N. caesarea*, and *N. nuda* ssp *albiflora* six FAMEs were identified.

<sup>1)</sup> Cumhuriyet University, Science and Art Faculty, Department of Biology, 58140, Sivas, Turkey, e-mail: sgorgun@cumhuriyet.edu.tr; 2) Balikesir University, Necatibey Education Faculty, Department of Biology Education, Balikesir, Turkey; 3) Selcuk University, Science and Art Faculty, Department of Biology, 42075, Konya, Turkey. Published in Khimiya Prirodnykh Soedinenii, No. 1, pp. 71-72, January-February, 2008. Original article submitted October 24, 2006.

TABLE 1. Fatty Acid Composition of Five Nepeta species from Turkey\*

Fatty acids	N. transcaucasica	N. italica	N. isaurica	N. caesarea	N. nuda ssp. albiflora
16:0	5.32±0.11ac	5.80±0.33a	5.13±0.53c	5.03±0.20c	4.34±0.19b
18:0	1.70±0.15a	1.39±0.11a	1.30±0.26ab	1.28±0.33ab	$0.91\pm0.22b$
20:0	-	$1.05\pm0.33$	$3.47\pm0.53$	-	-
16:1	$0.48\pm0.18a$	0.37±0.21a	$0.44\pm0.26a$	$0.33\pm0.24a$	$0.34\pm0.16a$
18:1	$12.55\pm0.24a$	14.22±0.18b	19.72±0.36c	19.85±0.15c	10.47±0.33d
18:2	$20.49\pm0.36a$	17.67±0.28b	21.93±0.24c	15.66±0.37d	22.33±0.44c
18:3	59.47±0.76a	59.51±0.81a	48.20±0.30b	57.85±0.21c	61.62±0.32d

<sup>\*</sup>The data are expressed as percentages of total fatty acids. Each value represents the mean of three experiments. t: means with the same letter in each row do not significantly differ at 0.05 level.

We identified three individual fatty acid components of the saturated form of fatty acids (SFAs) as palmitic acid (C 16:0), stearic acid (C 18:0), and eicosanoic acid (C 20:0). Among these fatty acids, C 16:0 and C 18:0 were identified in all *Nepeta* species investigated, but only two species of *Nepeta* (*N. italica* and *N. isaurica*) had C 20:0. *N. nuda* ssp *albiflora* had the lowest amounts of both C 16:0 and C 18:0 (4.34, 0.91%; respectively). The richest species in C 16:0 was *N. italica* (5.80%), while the highest amount of C 18:0 was found in *N. transcaucasica* (1.70%). The richest species in C 20:0 was *N. isaurica* (3.47%), while *N. italica* had the lowest amount (1.05%). A study conducted on *Sideritis* species (Lamiaceae) from Turkey indicated that C 16:0 and C 18:0 were the main components of the SFAs as well as C 20:0 [13].

All species of *Nepeta* in our study contained palmitoleic (C 16:1) and oleic acid (C 18:1) as monounsaturated fatty acids (MUFAs). There was not found any statistical differences in the five *Nepeta* species in the amounts of C 16:1, and the proportion of this fatty acid in all species ranged from 0.33% (*N. caesarea*) to 0.48% (*N. transcaucasica*). Amounts of C 18:1 in all species investigated were found to be between 10.47% (*N. nuda* ssp *albiflora*) and 19.85% (*N. caesarea*). A previous study on two *Satureja* species (*S. thymbra* and *S. cuneifolia*) from Lamiaceae has indicated that C 16:1 and C 18:1 were the major fatty acids of the MUFA fraction [14].

Nepeta species showed that the polyunsaturated fatty acids (PUFAs) linoleic (C 18:2) and linolenic (C 18:3) acid had the highest ratio in all the fatty acid fractions. The proportion of C 18:2 in the species was found to be between 15.66% (N. caesarea) and 22.33% (N. nuda ssp albiflora). The amount of C 18:3 in Nepeta species was substantially high, ranging from 48.20% (N. isaurica) to 61.62% (N. nuda ssp albiflora), and this shows that Nepeta species are invaluable resources of the n-3 form of PUFA. From the family Lamiaceae, there is little information on the fatty acid composition of Satureja, Sideritis, and Salvia species [13–15], and from our study it is established that the Nepeta species have unusual and the highest n-3 PUFA content among these genus from Lamiaceae. Moreover, this study provides comparative and comprehensive data on fatty acid compositions in different Nepeta species. As a result, such a favorable composition of unsaturated fatty acids of Nepeta species suggests that these species might have potential as a new oilseed crop for the food industry.

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