



Fatty acid composition in leaf lipids of some *Carex* L. (*Cyperaceae*) species from Northeast Anatolia (Turkey)

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RESUMEN

Composición en ácidos grasos de los lípidos de las hojas de varias especies del género *Carex* L. (*Cyperaceae*) del Nordeste de Anatolia (Turquía).

Se presenta la composición en ácidos grasos de 16 especies de *Carex* (de 15 secciones). El ácido palmítico es el ácido graso dominante en los lípidos de todas las especies examinadas, seguidos por los ácidos linoleico y linolenico. El valor taxonómico y las implicaciones filogenéticas de los resultados son discutidos.

PALABRAS-CLAVE: Ácidos grasos (composición) - *Carex* L. - Hoja - Turquía.

SUMMARY

Fatty acid composition in leaf lipids of some *Carex* L. (*Cyperaceae*) species from Northeast Anatolia (Turkey).

Fatty acid composition of 16 species of *Carex* from 15 sections are reported. The palmitic acid is the dominant fatty acid in lipid of all species and sections surveyed. The linoleic and linolenic acids were the second most abundant fatty acids. Taxonomic value and phylogenetic implications of results are discussed.

KEY-WORDS: *Carex* L. - Fatty acid (composition) - Leaf - Turkey.

1. INTRODUCTION

The genus *Carex* L., with about 2000 species, is the largest genus in *Cyperaceae* family and one of the most widespread and ecologically important genera of vascular plants (Reznicek, 1990). The genus is widely distributed and the species occur in a wide range of habitats from tropical areas to the high arctic. Some are particularly important in wetlands (Bernard, 1990). Species of *Carex* are among the most important forage plants for livestock in certain parts of the world, such as the rangelands of western North America, Northern Europe, Russia and Iceland (Catling *et al.*, 1990). For example, investigations on forage quality of *Carex macrochaeta*, has determined that it is a highly nutritious forage with high nitrogen and sodium values (Fox, 1991).

The genus *Carex* has taxonomic complexity (Catling *et al.*, 1990). Numerical (Bruederle *et al.*, 1989; Cayouette, 1990), micromorphological (Standley, 1985; Olgun and Beyazoglu, 1997), cytological

(Manhart, 1986; Luceno and Castroviejo, 1991) and anatomical (Toivonen, 1980; Standley, 1987) studies have been done to solve taxonomical problems of the genus. Manhart (1990) has reported that there have been few chemosystematic investigations on *Carex* genus. These investigations usually dealt with flavonoid (Catling *et al.*, 1989) and isozyme (Whitkus, 1992; Ford *et al.* 1991) studies. Lethonen (1990), has studied the amount of total water-soluble lipids in *Carex* peats. To our knowledge there is no other chemosystematic study on fatty acid composition in *Carex* genus. However, a few investigations were carried out to determine fatty acid composition in the tubers (Eteshola and Oraedu, 1996) and fruits (Kapseu *et al.*, 1997) of some species from other genera of *Cyperaceae* family.

The value of fatty acid patterns to establish systematic relationships among plants is becoming increasingly important. There are many studies in the literature dealing with phylogenetic and taxonomical aspects in relation with fatty acid composition in higher plants (Shorland, 1963; Harborne and Turner, 1984; Hegnauer, 1989).

This paper examines whether chemotaxonomical data can contribute to a phylogenetic classification within *Carex* and examines the uses of chemotaxonomical data in understanding the relationships of species and sections.

2. MATERIALS AND METHODS

The *Carex* species studied are *Carex hartmanii* Cajander, *C. pendula* Hudson, *C. rostrata* Stokes, *C. depressa* Link, *C. brevicollis* DC., *C. sylvatica* Hudson, *C. pallescens* L., *C. halleriana* Asso, *C. flacca* Schreber, *C. digitata* L., *C. liparocarpus* Gaudin, *C. pontica* Albov, *C. orbicularis* Boott., *C. punctata* Gaudin, *C. cilicica* Boiss and *C. paniculata* L. collected in Northeast Anatolia between June and August of 1996. Identifications of the specimens were made with the aid of Flora of Turkey (Nilsson, 1985), Flora of the USSR (Kreczetovicz, 1935), Flora Europaea (Chater, 1980). Specimens are kept in the Herbarium of Karadeniz Technical University, Department of

Biology (KTUB). Leaves were dried overnight under vacuo at 60°C and kept in a freezer (-20°C) until lipid extraction.

Lipid Extraction and Preparation of Fatty Acid Methyl Esters

Leaves were ground into a fine powder with a Waring blender and lipids were extracted with chloroform:methanol (2:1, v:v) according to Bligh and Dyer (1959). The extract was filtered through a sintered funnel, followed by 4 washes with the solvent mixture. The pooled filtrate was subjected to Folch washing (Folch and Sloane-Stanley, 1957) using 0.9% sodium chloride one fourth of the total filtrate to remove water-soluble impurities. The chloroform:methanol phase was subsequently evaporated to near dryness in a rotary vacuum evaporator, and the residue was again dissolved in 10 ml of chloroform. The lipids were saponified and fatty acids methylated according to Folch and Sloane-Stanley (1957). Lipid extractions were performed from dry leaf samples in triplicate. One g dry leaf sample was used for each extraction. Fatty acid methyl esters were also prepared from these three separate lipid extracts.

Gas Chromatography Analysis of Fatty Acid Methyl Esters

The methylated sample solutions were analyzed with a Varian 3300 gas chromatograph (GC) equipped with a flame ionization detector (FID) and a HP-1 silica capillary column (crosslinked methyl silicone gum, 0.17 µm film thickness, 25 m, 0.32 mm i.d.). Hydrogen was used as the carrier gas at a flow rate of ca. 40 cm/s, and the column oven temperature was programmed from 100°C to 290°C at 6°C/min heating rate. The injector and detector temperatures were held at 260°C and 290°C, respectively. Peak areas were measured with a Merck-Hitachi D-2000 integrator. Peaks were identified by comparison with standard fatty acid methyl esters.

Statistical analysis

All extractions and determinations were conducted three times independently. Analysis of variance of the data were evaluated by the Statistical Analysis System (STATGRAPH Version 5.0). Duncan's Multiple Range Test was employed to determine the statistical significance of differences among the means ($P=0.05$).

Phenetic Analysis

A phenetic study of 16 species of *Carex* was made using fatty acid composition (Table I). STATISTICA

package was used to produce a phenogram using the unweighted pair-group method with an arithmetic average (UPGMA).

3. RESULTS AND DISCUSSION

Total leaf lipid fatty acids composition is shown in Table I. A total of 9 fatty acids were identified in 16 species of *Carex* genus belonging to 15 sections.

The presence of saturated (C 14:0, C 16:0, C 18:0, C 20:0, C 22:0 and C 24:0) and unsaturated (C 18:1, C 18:2 and C 18:3) fatty acids were determined, with C 16:0, C 18:2 and C 18:3 being the major fatty acids. The values of C 16:0 acid varied between 51.87% in *C. liparocarpus* and 77.64% in *C. rostrata*. The values of C 18:2 varied between 3.06 and 26.36%, for *C. punctata* and *C. cilicica*, respectively.

The values for C 14:0, C 20:0, C 22:0 and C 24:0 were generally found lower varying between 2.02 and 4.91% for C 14:0, < 0.1 and 5.04% for C 20:0, < 0.1 and 1.92% for C 22:0 and 1.32 and 2.80% for C 24:0, respectively.

To sum up, *Carex* fatty acid composition is characterized by the presence of high levels of C 16:0, C 18:2, and C 18:3.

Several works have been undertaken on fatty acid composition in oils from different *Cyperaceae* species. In *Cyperus esculentus* L. tubers, C 14:0 acid was the main saturated acid and C 18:1 the predominant unsaturated one. C 18:2 acid was determined in the tubers with a percentage of 8.8% (Eteshola and Oraedu, 1996). Kapseu *et al.* (1997) recently reported some fatty acids of fruit oils in

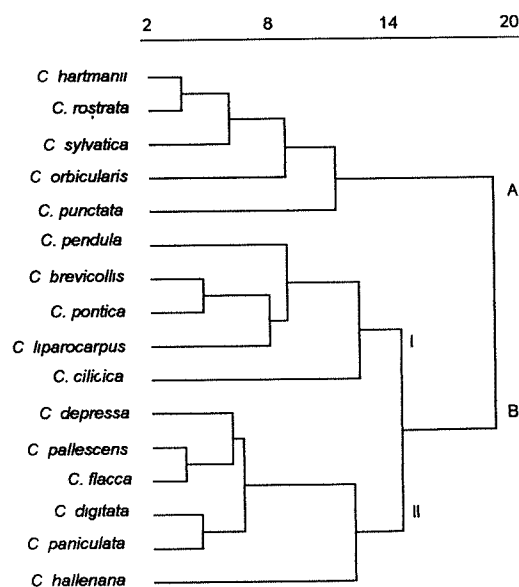


Figure 1
UPGMA phenogram of *Carex* species clustered on the basis of fatty acid composition

Table I
Fatty acid composition of *Carex* species studied (Means of three different extraction and determinations. Values with same letter are not significantly different at $P=0.05$. Means were compared within each column of the data, not rows)

Species	Fatty Acids (%)								
	14:0	16:0	18:0	18:1	18:2	18:3	20:0	22:0	24:0
1. Sect. Atratae <i>C. hartmanii</i> Cajander	3.46 ^e	76.38 ^k	2.27 ^a	5.1 ^{ef}	6.72 ^{cd}	3.42 ^b	1.15 ^a	tr*	1.48 ^{bc}
2. Sect. Rhyncocystis <i>C. pendula</i> Hudson	3.33 ^e	59.14 ^e	4.37 ^c	3.74 ^{cd}	12.86 ^g	14.03 ^k	tr	tr	2.53 ^e
3. Sect. Carex <i>C. rostrata</i> Stokes	3.19 ^d	77.64 ^l	3.54 ^{ab}	4.22 ^d	3.87 ^b	4.07 ^b	1.47 ^{ab}	tr	2.10 ^{cd}
4. Sect. Mitratae <i>C. depressa</i> Link	2.35 ^{ab}	59.79 ^{ef}	6.24 ^e	4.89 ^e	11.64 ^{fg}	6.18 ^{de}	5.04 ^e	tr	2.05 ^{cd}
5. Sect. Ventricosae <i>C. brevicollis</i> DC	2.04 ^a	56.19 ^d	4.41 ^c	4.23 ^d	18.03 ⁱ	11.4 ^j	1.45 ^{ab}	0.92 ^a	1.32 ^b
6. Sect. Strigosae <i>C. sylvatica</i> Hudson	2.71 ^c	72.37 ^j	5.66 ^d	4.47 ^{de}	6.33 ^c	4.74 ^c	1.18 ^a	0.92 ^a	1.63 ^{bc}
7. Sect. Porocystis <i>C. pallescens</i> L.	2.11 ^a	63.77 ^g	3.76 ^{abc}	4.38 ^{de}	11.74 ^{fg}	8.29 ^g	1.51 ^{ab}	1.92 ^c	2.80 ^{ef}
8. Sect. Hallerianae <i>C. halleriana</i> Asso	2.47 ^b	53.03 ^b	8.06 ^{fg}	6.66 ^h	7.19 ^d	8.49 ^g	4.6 ^{de}	1.75 ^c	2.74 ^{ef}
9. Sect. Glaucuae <i>C. flacca</i> Schreber	3.39 ^e	62.7 ^f	4.98 ^{cd}	6.01 ^g	12.55 ^g	6.65 ^e	1.35 ^{ab}	1.14 ^{ab}	1.42 ^{bc}
10. Sect. Digitatae <i>C. digitata</i> L.	2.33 ^{ab}	63.97 ^g	7.55 ^f	4.14 ^d	9.48 ^f	7.19 ^f	3.69 ^c	tr	1.64 ^{bc}
11. Sect. Lamprochlaenae <i>C. liparocarpus</i> Gaudin	2.02 ^a	51.87 ^a	5.12 ^{cd}	5.02 ^{ef}	15.42 ^h	15.44 ^l	2.02 ^b	1.16 ^{ab}	1.95 ^c
12. Sect. Aulocystis <i>C. pontica</i> Albov	2.32 ^{ab}	54.98 ^{cd}	2.77 ^a	4.1 ^d	22.04 ^j	10.82 ⁱ	0.97 ^a	0.65 ^a	1.36 ^b
13. Sect. Phacocystis <i>C. orbicularis</i> Boott.	2.24 ^{ab}	69.96 ⁱ	3.37 ^{ab}	3.22 ^{bc}	8.72 ^e	9.23 ^h	1.09 ^a	1.06 ^{ab}	1.09 ^a
14. Sect. Fulvellae <i>C. punctata</i> Gaudin <i>C. cilicica</i> Boiss	3.90 ^{ef} 2.89 ^{cd}	70.54 ⁱ 54.58 ^c	11.46 ^h 3.25 ^{ab}	3.49 ^{bc} 2.03 ^a	3.06 ^a 26.36 ^k	1.83 ^a 5.99 ^d	4.4 ^d 2.21 ^b	tr 1.20 ^b	1.32 ^b 1.58 ^{bc}
15. Sect. Heleoglochis <i>C. paniculata</i> L.	4.91 ^f	65.82 ^h	7.71 ^f	2.89 ^b	7.87 ^{de}	5.57 ^{cd}	2.51 ^{bc}	1.1 ^{ab}	1.61 ^{bc}

*: trace amounts (< 0.1%).

Cyperus esculentus. C 18:0 acid was identified as the main fatty acid with 33.5%. C 16:0 and C 18:0 acid were the second most abundant acids constituting 17% and 43.6% of total fatty acid composition.

Thus, as in the tuber of other *Cyperaceae* species, in the leaves of *Carex* species C 16:0 was identified as the main fatty acid while C 18:2 acid was the second most abundant fatty acid.

The cluster analysis (Fig. 1) indicates the existence of two groups. Group A consists of the sections with a high level C 16:0 acid. Group B consists of the sections with a high level C 18:2 and

C 18:3 acids. *C. punctata* and *C. cilicica* from the section *Fulvellae* occur in different groups.

From a chemotaxonomic point of view, the present results explain the alliances among species of the genus *Carex* suggesting that fatty acid composition may be useful in the chemotaxonomy of *Carex* genus.

Further studies which could include a larger number of *Carex* species, as well as the analysis of additional lipidic fractions, will be useful in enlarging the chemical characterization of this genus and also gaining a better understanding of current taxonomic relationships in *Carex*.

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