

Fatty acid composition of hempseed oils from different locations in Turkey

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Abstract

Recent interest in hempseed as a source of food has largely focused on its oil content and fatty acid profile. The oil content and fatty acid composition (15 fatty acids) of twenty one different hempseed samples of domestic origin from north-western Turkey were monitored. The samples were obtained from seed wholesalers and local spice shops and are of unknown genetic origin. The oil content of the hempseeds ranged between 29.6 to 36.5%. Out of the 15 detected fatty acids, the omega-6 linoleic acid (18:2n-6) was predominant and fluctuated from 55.4 to 56.9%, while the omega-3 α -linolenic (18:3n-3) acid ranged from 16.5 to 20.4% and the omega-9 oleic acid (18:1n-9) ranged from 11.4 to 15.9%. Of the minor fatty acids, the highest concentrations were found for γ -linolenic acid (18:3n-6), range 0.6-1.1%, followed by stearidonic acid (18:4n-3), range 0.3-0.5%. These results show that hempseed grown in north-western Turkey provides a well balanced and rich source of dietary omega-6 and -3 essential fatty acids and appears to be a potentially valuable source of food.

Additional key words: EFA, essential fatty acid, hemp, linoleic acid, linolenic acid, seed oil.

Resumen

Composición de ácidos grasos en aceites de semillas de cáñamo de diferentes localidades de Turquía

El reciente interés por las semillas de cáñamo como alimento se ha enfocado en su contenido en aceite y su perfil de ácidos grasos. En este estudio se analizó el contenido en aceite y composición de ácidos grasos de 21 muestras diferentes de semillas de cáñamo de origen genético desconocido en el noroeste de Turquía. El contenido en aceite de las semillas varió entre 29,6 y 36,5%. De los 15 ácidos grasos detectados, el ácido linoleico (18:2n-6) (omega-6) fue predominante y fluctuó entre 55,4 y 56,9%; el ácido α -linolénico (18:3n-3) (omega-3) entre 16,5 y 20,4%; y el ácido oleico (18:1n-9) (omega-9) entre 11,4 y 15,9%. Entre los ácidos grasos minoritarios, se encontraron las mayores concentraciones (entre 0,6 y 1,1%) para el ácido γ -linolénico (18:3n-6), seguido del ácido estearidónico (18:4n-3), entre 0,3 y 0,5%. Estos resultados muestran que las semillas de cáñamo que se cultivan en el noroeste de Turquía pueden ser un alimento potencialmente valioso y constituirían para la dieta una fuente rica y equilibrada de ácidos grasos omega-6 y -3.

Palabras clave adicionales: aceite de semillas, ácido linoleico, ácido linolénico, ácidos grasos esenciales, cáñamo, EFA.

Introduction

Hemp (non-drug varieties of *Cannabis sativa* L.), considered to originate from Western and Central Asia, has long been widely cultivated in many parts of the

world for mainly fiber and oil. It has received increasing attention during the last few decades because of its wide range of possible uses; particularly for the nutritional and healing properties of hempseed and its oil to humans.

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Abbreviations used: ALA (alpha-linolenic acid), DHA (docosahexaenoic acid), EFA (essential fatty acid), EPA (eicosapentaenoic acid), FID (flame ionization detector), GLA (gamma-linolenic acid), LA (linoleic acid), MUFA (monounsaturated fatty acid), PUFA (polyunsaturated fatty acid), SDA (stearidonic acid), SE (standard error), SFA (saturated fatty acid), UFA (unsaturated fatty acid).

Hempseed has unique nutritional properties and has commonly been claimed to be one of the most nutritionally complete foods and is truly one of nature's super foods (Osburn, 1992; Deferne and Pate, 1996). Hempseeds contain all the essential amino acids and essential fatty acids necessary to maintain healthy human life. No other industrial plant seed has such a full amino acid spectrum in such an easily digestible form with the two essential fatty acids (EFAs) in ratio that benefits human nutritional needs (Callaway *et al.*, 1996). Hempseed contains 20-25% protein, 20-30% carbohydrates, 25-35% oil and 10-15% insoluble fiber and a rich array of minerals (Deferne and Pate, 1996; Pate, 1999).

The value of hempseed oil from the point of view of the primary components is its fatty acid composition. Hemp is of high nutritional quality because it contains the two dietary essential fatty acids, namely the omega-6 linoleic acid (LA; 18:2n6, 50-60%) and the omega-3 alpha-linolenic acid (ALA; 18:3n3, 20-25%) in the ratio of 3:1, which has been claimed ideal for human nutrition (Simopoulos, 1991, 2002). These two essential fatty acids, comprise up to 80% of total fatty acids of hempseed oil, serve mainly as raw materials for cell structure and as precursors for biosynthesis of many functional metabolites. Hempseed oil is also one of the few botanical sources of gamma-linolenic acid (GLA; 18:3n6) and stearidonic acid (SDA; 18:4n3), which are of increasing pharmaceutical interest, making the nutritional value of hempseed superior to other seed oils (Leizer *et al.*, 2000). Both GLA and SDA act as precursors for the rapid synthesis of longer chain fatty acids, such as eicosanoids, in the human body (Callaway *et al.*, 1996; Guil-Guerrero, 2007).

Hempseed, in addition its nutritional value, has demonstrated positive health benefits (Callaway *et al.*, 2005; Schwab *et al.*, 2006) and has traditionally been consumed in folk and folk medicinal preparations, employed as bird and poultry feed, or as a functional food (Oomah *et al.*, 2002). Roasted hempseed, Turkish name is *çedene*, mixed with roasted bread wheat (Turkish name is *kavurga*) has long been traditionally used as a snack in countryside of Turkey. The interest in hemp for cosmetic products, particularly skin care, is due to the high content of favorable unsaturated fatty acids in seed of hemp, particularly linoleic, α -linolenic, and γ -linolenic acids, which influence several important cell membrane functions (Vogl *et al.*, 2004).

It has been reported that the origin of the seed seems to influence oil content and particularly the fatty acid

composition (Ross *et al.*, 1996; Mölleken and Theimer, 1997). In the literature, there are a number of studies indicating appreciable differences among fatty acid compositions of the hempseed oils from variable regions or countries (Leizer *et al.*, 2000; Kriese *et al.*, 2004), or even among samples from the same country (Orhan *et al.*, 2000; Bagci *et al.*, 2003; Anwar *et al.*, 2006). Therefore, the full characterization of hempseed oils of diverse origins still appears to be a sound research priority to obtain a reliable data on this valuable crop. However, studies on oil content and fatty acid composition of hempseed from Turkey are very limited. We thus analyzed twenty one hempseed samples that were produced in north-western Turkey, formerly the most important region for hempseed production.

Material and methods

Materials

Hempseed samples of domestic origin were obtained from seed wholesalers and local spice shops in north-western Turkey (Taşköprü, Vezirköprü, Ordu, Çorum, Gümüşhacıköy, Samsun and Ereğli provinces or towns) between Lat. 40°30'-41°30'N and Long. 31°25'-37°52'E. This is the most important region for hemp production in Turkey, where there is no registration of hemp varieties, but just local landraces. Then, the genetic origin of samples is unknown. For oil and fatty acid analysis, three different seed samples (a total of 21 seed samples) of 300 g were taken in each of the seven sites. After removing the seed impurities, the seed samples were stored at dark and cooling place until they were analyzed.

Methods

The seeds (approximately 1 g) were ground and added to 75 mL of hexane. The mixture was then placed in FOSS Soxtec 2055 Apparatus for oil extraction. To determine the fatty acid fraction of each oil sample, fatty acids were esterified to methyl esters (AOAC, 1990) and the resulting solutions were analyzed by Shimadzu (Kyoto, Japan) gas chromatograph equipped with DB-23 capillary column (30 m \times 0.25 μ m) and FID (flame ionization detector). The carrier gas was helium; at a flow rate of 1.0 mL min⁻¹. Injector and detector temperatures were 230 and 240°C, respectively. Column

temperature was kept at 190°C for 30 min. Samples of 1.0 µL were injected by hand with a split mode (1:80). The fatty acid identification was performed by retention time comparisons with the corresponding fatty acid methyl ester standards. Individual reference methyl ester standards (miristic acid (C14:0), palmitic acid (C16:0), stearic acid (C18:0), oleic acid (C18:1), linoleic acid (C18:2), arachidic acid (C20:0), gadoleic acid (C20:1), behenic acid (C22:0) and lignoceric acid (C24:0) and as well as fatty acid methyl ester mix (37 components FAME mix) were purchased from Sigma Chemical Co. (Sigma-Aldrich GmbH, Sternheim, Germany).

Results and discussion

In this study twenty one different hempseed samples obtained from seven provinces and/or vicinities in north-western Turkey were subjected to chemical analysis for their oil content and fatty acid composition. The oil contents of the hempseeds, given as means \pm SD for the three samples obtained from each site, are presented in Table 1.

The oil contents of the hempseed samples ranged from 29.61 to 36.47%, with the average of 32.13%. The highest oil content was measured in the samples received from the town of Gümüşhacıköy, followed by Taşköprü (34.24%), which are temperate regions with a cold winter and hot summer. On the other hand, the lowest value was recorded in the samples from the district of Ereğli, which is a wet region with mild climate. The vicinity of Gümüşhacıköy has previously been well-known for hempseed production of good quality.

The values obtained in this study for oil content of the hempseeds in north-western Turkey were within the expected range reported previously for hempseed

(Deferne and Pate, 1996; Oomah *et al.*, 2002; Kriese *et al.*, 2004). The average oil content (32.13%) was similar to that (31.79%) reported earlier from Turkey (Bagci *et al.*, 2003), but higher than the value (28.87%) reported for hempseeds grown in different agro-ecological regions of Pakistan (Anwar *et al.*, 2006). The oil content values, however, ranged between 35.27 and 36.88% in a study undertaken in north-western Turkey (Ozdemir, 1993). Since the oil content and composition of hemp seeds is largely affected by environmental factors (Ross *et al.*, 1996; Kriese *et al.*, 2004), along with processing and storage methods, differences in oil content and, to a lesser extent, the fatty acid composition of the seeds may be observed among samples even from the same country.

The fifteen fatty acids such as myristic, palmitic, palmitoleic, stearic, oleic, linoleic, gamma-linolenic, alpha-linolenic, stearidonic, arachidic, gadoleic, eicosadienoic, behenic, erucic and lignoceric acid were identified in the oil of the hempseeds monitored. The fatty acid breakdown of the hempseed oils are shown in Table 2.

The principal saturated fatty acid in the hempseed samples was palmitic acid (C16:0), one of the most common saturated fatty acids found in animals and plants, ranging between 6.08% and 6.82%. It was followed by stearic acid (C18:0), varying from 2.34% to 2.67%. The amounts of the other saturated fatty acids were below 1%. Arachidic (0.60-0.76%), behenic (0.21-0.25%), lignoceric (0.06-0.12%) and myristic acid (0.03-0.04%) were saturated fatty acids found in trace amounts in all seed samples. The saturated fatty acid fraction represents 9.37-10.57% of the total fatty acids present in hempseeds. This low level of saturation was comparable to those reported in the literature (Ross *et al.*, 1996; Mölleken and Theimer, 1997; Oomah *et al.*, 2002; Bagci *et al.*, 2003; Anwar *et al.*, 2006).

Concentrations of palmitic and stearic acid from hempseed oil were within the range reported by Bagci *et al.* (2003), Parker *et al.* (2003) and Kriese *et al.* (2004). Stearic acid, in contrast with palmitic acid, was shown not to raise cholesterol concentration (Grundy, 1994). Oils containing higher stearic acid contents have more desirable physical and chemical properties for food industry (Fernández-Moya *et al.*, 2005; Mensink, 2005) and might be used in place of those high in palmitic acid in cholesterol-lowering diets. However, French *et al.* (2002) showed that palmitic acid had no hypercholesterolaemic effect if intake of linoleic acid was greater than 4.5% of energy.

Table 1. Oil contents of hemp seeds (%), mean \pm SE

Samples	Oil content (%)
Taşköprü	34.24 \pm 0.36
Vezirköprü	30.95 \pm 0.62
Çorum	31.55 \pm 0.64
Ereğli	29.61 \pm 0.01
Gümüşhacıköy	36.47 \pm 0.01
Ordu	30.78 \pm 0.54
Samsun	31.29 \pm 1.67

SE: standard error.

Table 2. Fatty acid contents of hemp seed oils (% , mean \pm SE)

	Çorum	Ereğli	Gümüshacıköy	Ordu	Samsun	Taşköprü	Vezirköprü
C14:0	0.04 \pm 0.00	0.04 \pm 0.00	0.03 \pm 0.00	0.04 \pm 0.01	0.04 \pm 0.00	0.04 \pm 0.01	0.04 \pm 0.00
C16:0	6.44 \pm 0.01	6.14 \pm 0.01	6.61 \pm 0.01	6.08 \pm 0.01	6.27 \pm 0.02	6.82 \pm 0.09	6.46 \pm 0.01
C16:1	0.13 \pm 0.03	0.13 \pm 0.02	0.11 \pm 0.01	0.10 \pm 0.01	0.12 \pm 0.02	0.12 \pm 0.05	0.11 \pm 0.01
C18:0	2.49 \pm 0.01	2.46 \pm 0.01	2.67 \pm 0.01	2.34 \pm 0.01	2.42 \pm 0.00	2.64 \pm 0.01	2.44 \pm 0.00
C18:1	11.40 \pm 0.01	14.50 \pm 0.01	15.88 \pm 0.02	13.38 \pm 0.01	14.95 \pm 0.01	14.60 \pm 0.04	13.20 \pm 0.01
C18:2	56.61 \pm 0.08	56.23 \pm 0.06	55.48 \pm 0.01	56.94 \pm 0.01	56.40 \pm 0.02	55.41 \pm 0.11	56.19 \pm 0.03
C18:3 γ	0.64 \pm 0.00	1.10 \pm 0.01	0.82 \pm 0.00	1.07 \pm 0.01	1.03 \pm 0.00	0.83 \pm 0.00	0.97 \pm 0.00
C18:3 α	20.40 \pm 0.04	17.40 \pm 0.00	16.51 \pm 0.01	18.20 \pm 0.01	16.86 \pm 0.01	17.58 \pm 0.05	18.58 \pm 0.00
C18:4	0.34 \pm 0.01	0.47 \pm 0.01	0.34 \pm 0.00	0.47 \pm 0.00	0.41 \pm 0.00	0.37 \pm 0.02	0.45 \pm 0.01
C20:0	0.60 \pm 0.00	0.66 \pm 0.01	0.75 \pm 0.00	0.61 \pm 0.01	0.65 \pm 0.00	0.76 \pm 0.02	0.67 \pm 0.02
C20:1	0.39 \pm 0.01	0.37 \pm 0.00	0.36 \pm 0.01	0.32 \pm 0.00	0.36 \pm 0.00	0.38 \pm 0.04	0.37 \pm 0.01
C20:2	0.06 \pm 0.01	0.05 \pm 0.00	0.04 \pm 0.01	0.04 \pm 0.00	0.05 \pm 0.00	0.04 \pm 0.01	0.05 \pm 0.00
C22:0	0.21 \pm 0.01	0.24 \pm 0.01	0.25 \pm 0.01	0.22 \pm 0.01	0.24 \pm 0.00	0.23 \pm 0.02	0.24 \pm 0.01
C22:1	0.16 \pm 0.00	0.16 \pm 0.01	0.12 \pm 0.01	0.15 \pm 0.01	0.13 \pm 0.00	0.15 \pm 0.01	0.18 \pm 0.01
C24:0	0.12 \pm 0.04	0.10 \pm 0.01	0.06 \pm 0.00	0.08 \pm 0.01	0.09 \pm 0.00	0.08 \pm 0.01	0.09 \pm 0.01
SFAs	9.9	9.6	10.4	9.4	9.7	10.6	9.9
UFAs	90.1	90.4	89.6	90.6	90.3	89.5	90.1
MUFAs	12.1	15.2	16.4	13.9	15.6	15.3	13.9
PUFAs	78.0	75.2	73.2	76.7	74.7	74.2	76.2

SFA: saturated fatty acids. UFA: unsaturated fatty acids. MUFA: monounsaturated fatty acids. PUFA: polyunsaturated fatty acids.

The results of the present study revealed that the oils in the hempseed samples from north-western Turkey were high in polyunsaturated fatty acids (89.43-90.63%). Linoleic acid (LA; 18:2n-6), running between 55.41% and 56.94%, was the most abundant unsaturated fatty acid of hempseed oils, followed by α -linolenic (ALA; 18:3n-3) and oleic acid (18:1n-9) with the ranges of 16.51-20.40% and 11.40-15.88%, respectively. These three unsaturated fatty acids, the major fatty acids present in hempseed, comprised 87.59-88.52% of the total fatty acids. It has been reported however that such a high degree of unsaturation makes hempseed oil extremely sensitive to oxidative rancidity, so the oil has a relatively short shelf life (Deferne and Pate, 1996; Callaway, 2004).

The range (11.40-15.88%) of oleic acid was similar to those reported earlier (Oomah *et al.*, 2002; Carvalho *et al.*, 2006), but higher than the values reported by Callaway *et al.* (1996) and Anwar *et al.* (2006). The contents of linoleic acid in the hempseed oils from north-western Turkey were slightly higher than the values reported by Orhan *et al.* (2000), Bağcı *et al.* (2003) and Kriese *et al.* (2004), but lower than those reported by Callaway *et al.* (1996), Anwar *et al.* (2006) and Carvalho *et al.* (2006). It is interesting to note that the range of α -linolenic acid found in the present study was generally lower than those reported earlier in the literature (Callaway *et al.*, 1996; Oomah *et al.*,

2002; Kriese *et al.*, 2004; Anwar *et al.*, 2006), but well in line with that of Carvalho *et al.* (2006). Similarly, Orhan *et al.* (2000) and Bağcı *et al.* (2003) reported appreciably higher α -linolenic acid content of hempseed oil from Turkey.

The ratio of n-6 to n-3, essential fatty acids that must come from dietary sources, and the total unsaturated to the saturated fatty acids varied from 2.8 to 3.4 and 8.6 to 9.7, respectively. Omega-3 and omega 6 fatty acids are not interconvertible in the human body and are important components of all cell membranes (Simopoulos, 1991). Excessive amounts of omega-6 polyunsaturated fatty acids and a very high omega-6/omega-3 ratio promote the risk of many diseases, including cardiovascular disease, cancer, and inflammatory and autoimmune diseases, whereas increased levels of omega-3 PUFA (a low omega-6/omega-3 ratio) exert suppressive effects (Simopoulos, 2002).

The other fatty acids (palmitoleic, γ -linolenic, stearidonic, gadoleic, eicosadienoic and erucic acids) were found in small amounts. However, γ -linolenic acid (GLA; 18:3n-6) content, characteristic of hempseed oil with a pharmaceutical interest, was higher among other minor unsaturated fatty acids and varied from 0.64 to 1.10%. It has been well documented (Deferne and Pate, 1996; Mölleken and Theimer, 1997; Callaway, 2004) that the presence of both stearidonic acid (highly unsaturated omega-3) and γ -linolenic acid (highly

unsaturated omega-6) in hempseed oil ultimately makes its nutritional value superior to any other industrial oilseed crop. The γ -linolenic acid content of the hempseed in the present study was significantly lower than those reported by Callaway and Laakkonen (1996), Callaway *et al.* (1996), Mölleken and Theimer (1997) and Oomah *et al.* (2002); and was higher than that reported by Bagci *et al.* (2003), whereas was comparable to the values reported by Parker *et al.* (2003) and by Anwar *et al.* (2006). Studies have shown that hempseeds from regions with a mild or warm climate contain small amounts of γ -linolenic acid, whereas hempseeds from temperate or even cold regions have a large amount (Deferne and Pate, 1996; Ross *et al.*, 1996; Mölleken and Theimer, 1997).

In the seed samples examined, stearidonic acid (SDA, 18:4n-3) was not in such high percentages as GLA and varied from 0.34 to 0.47%. Although stearidonic acid seems to have a very limited presence in domesticated plants, it may function as an important human dietary component of hempseed oil (Callaway *et al.*, 1996), especially in combination of GLA. Stearidonic acid is a polyunsaturated fatty acid that constitutes the first metabolite of α -linolenic acid in the metabolic pathway leading to longer chain omega-3 fatty acids, eicosapentaenoic acid (EPA; C20:5n-3) and docosahexaenoic acid (DHA; C22:6n-3), which have recently gained much attention (James *et al.*, 2003; Ursin, 2003; Newell-McGloughlin, 2008). The other unsaturated fatty acids such as gadoleic, palmitoleic, eicosadienoic and erucic acids were also identified in all samples within the ranges of 0.32-0.39%, 0.10-0.13%, 0.04-0.06% and 0.12-0.18%, respectively.

In conclusion, the results of this study showed that hempseed in north-western Turkey contains a fatty acid composition of high and well balanced dietary essential fatty acids; thus, it appears to be a potentially valuable source of food. Furthermore, in accordance with the recently acquired knowledge concerning hempseed oil, these findings could be a valuable contribution in obtaining a reliable data on various occurrences of fatty acids, dietary essential fatty acids in particular, among hempseeds of various regions.

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