Original Article

Lipid and fatty acid profiles of *Castanea sativa* Mill. Chestnuts of 17 native Portuguese cultivarsOlga Pires Borges^a, José Soeiro Carvalho^a, Paula Reis Correia^b, Ana Paula Silva^{c,*}^a*Direcção Regional de Agricultura de Trás-os-Montes, Quinta do Valongo, Carvalhais, 5370-087 Mirandela, Portugal*^b*Escola Superior Agrária de Viseu, Quinta da Alagoa, Estrada de Nelas, Ranhados, 3500-606 Viseu, Portugal*^c*Universidade de Trás-os-Montes e Alto Douro, CECEA, Quinta dos Prados, 5000-911 Vila Real, Portugal*

Received 16 November 2005; received in revised form 5 July 2006; accepted 5 July 2006

Abstract

In recent years, chestnuts have increasingly become important in human nutrition because of their nutrient contents and potential beneficial health effects. In Portugal, Trás-os-Montes e Alto Douro is the most important region for chestnut production, generating 85% of the total national output. In 1994, three chestnut protected designation of origin (PDO) areas were created: Terra Fria, Soutos da Lapa, and Padrela within the jurisdiction area of the Regional Agricultural Services of Trás-os-Montes Region. Seventeen cultivars were selected from these PDO to evaluate crude fat content and fatty acid (FA) composition over a period of 2 years. Total fat extraction was performed according to AOAC methodology, using a Soxhlet apparatus for 16 h. Petroleum ether was used for extraction and FAs were identified and quantified by gas chromatography. The cultivars studied confirm that chestnuts have low crude fat content, low saturated fatty acids (SFA) (17%) and high unsaturated fatty acids (USFA) (83%). The major FAs are linoleic, oleic and palmitic; these accounted for more than 85% of the total FA content. From the results two potentially health-beneficial cultivars were identified: cv. Lada, with high linoleic acid content, and cv. Avelaira, with higher USFA content. With respect to FA profiles of chestnuts from the three PDO regions, Padrela chestnuts consistently contained the highest level of linoleic acid. The variation detected among cultivars and PDO areas may reflect differences in genotype and are influenced by environmental and growing conditions. The Longal chestnuts, the only traditional cv. growing on two different PDO areas, had better nutritional qualities in PDO Terra Fria, which confirm that the ecological conditions where chestnut orchards are established influences nut quality.

© 2006 Elsevier Inc. All rights reserved.

Keywords: Chestnut; *Castanea sativa* Mill.; Gas chromatograph; Protected designation of origin (PDO); Genotype; Crude fat; Fatty acid composition

1. Introduction

The sweet chestnut tree (*Castanea sativa* L.) produces chestnuts, which are an ancient tree crop in northern Portugal of great social and economic importance. They were a basic foodstuff for many families until potatoes and cereals became available. The Trás-os-Montes e Alto Douro region is one of the most favorable in terms of climatic, edaphic and ecological conditions. Nowadays, this region stands out as the most important chestnut-producing area of the country, generating 85% of the total national chestnut output, and it is one of the few regions

with a largely positive trade balance, which was about 9 million euros in 2002 and continues to show annual improvements (GPPAA, 2003).

In Trás-os-Montes e Alto Douro there are three protected designations of origin (PDO) areas for chestnuts: Terra Fria, Padrela and Soutos da Lapa. In each PDO, with different ecologies and geographical parameters, many well-adapted native cultivars are cultivated.

In recent years, chestnuts have become more important in human health because of their nutritional qualities and potential beneficial health effects. Chestnut fruits could be used as human and animal foods as extensively as potato and most of the tubers (Ferreira-Cardoso et al., 1999). Consumers have been showing an increased interest in food composition particularly with respect to fatty acid (FA)

*Corresponding author. Tel.: +351 259350450; fax: +351 259350327.

E-mail address: asilva@utad.pt (A. Paula Silva).

contents. Essential FAs are important because of their role in diverse physiological processes affecting normal health and chronic diseases, such as the regulation of plasma lipid levels, cardiovascular and immune function, insulin action and neuronal development and visual function (Benatti et al., 2004). The consumption of omega-3 FAs can slow the growth of cancerous cells, increase the efficacy of chemotherapy and reduce the side effects of the chemotherapy for cancer (Hardman, 2002). In addition chestnuts are currently used in pediatrics for treatment of gastroenteritis and as a gluten-free diet in cases of celiac disease (Pazianas et al., 2005). Chestnuts are a good source of essential FAs (Desmaison and Adrian, 1986; Senter et al., 1994; Ferreira-Cardoso et al., 1999; Künsch et al., 1999; Mello et al., 2001).

Several studies in other crops showed important correlations between FA composition and environmental conditions. Significant differences among genotypes and cultivation areas were found in the proportions of palmitic, palmitoleic and stearic FAs for almond kernels in California production regions (Abdallah et al., 1998). A strong influence of environmental and harvest factors on the synthesis of triglycerides was found in hazelnuts, namely those with at least one linoleic acid moiety in their structure (trilinolein, oleo-diolein, dilinolein-palmitin, dioleo-linolein and palmito-oleo-linolein) (Parcerisa et al., 1994). Myristic, palmitoleic, stearic and oleic acids were negatively correlated with latitude of non-glutinous rice cultivars in Japan (Kitta et al., 2005). Ferreira-Cardoso et al. (1999) suggest that the differences found in FA

composition among cultivars could be influenced by environmental and growth conditions, but more work needs to be done. Gomes et al. (1997) also detected a relationship between the protein content of chestnuts and the type of soil in which the trees had been grown.

Data on crude fat and FA composition from traditional economically important Portuguese cultivars is still limited. The purpose of this study was to evaluate the variation in crude fat and FA composition among 17 cultivars of sweet chestnut fruits, which grow in the three PDO from the Trás-os-Montes and Alto Douro region.

2. Materials and methods

Crude fat and FA profiles were determined for 17 sweet chestnut cultivars in two crop years (2001 and 2002). The selection of the cultivars was made within the three protected designation of origin (PDO) areas for chestnut tree production in the Trás-os-Montes e Alto Douro region: Terra Fria, Padrela and Soutos da Lapa. Each PDO area has specific ecological conditions (Table 1 and Fig. 1). From each PDO were selected a representative sample, consisting of three trees from the most common traditional cultivars: Redonda, Aveleira, Rebordã, Longal, Trigueira, Boaventura, Lamela and Zeive from Terra Fria; Judia, Lada, and Cota from Padrela; Demanda, Martainha, Ferreirinha, Cancela, Carreiró, Longal and Riscada from Soutos da Lapa. Mature chestnuts were harvested and three replications of 1 kg were randomly collected for each genotype.

Table 1
Portuguese chestnuts: geographic and soil characterization of protected designation of origin (PDO)

PDO	Altitude (m)	Latitude	Longitude	Soil
Terra Fria	650–900	41°40′–41°55′N	6°50′–7°10′W	District cambisols from schists
Soutos da Lapa	700–800	40°59′–41°05′N	7°25′–7°50′W	District cambisols from granites
Padrela	800–900	41°35′–41°35′N	7°24′–7°30′W	Humic umbrisols from granites

Source: Soil map region and correspondence to the world reference base for soil resources (FAO, 1998).

Fig. 1. Chestnut growing areas.

2.1. Sample preparation

Samples were stored for 1 week in a refrigerator (2 °C) until analyses were performed. Nut samples were pre-dehydrated at 40 °C in a model FD 115 BINDER ventilated drying chamber for 24 h. After drying they were peeled and the nuts rapidly cut into small pieces, in order to promote milling operations. They were further dried at 42 °C under the same conditions for 48 h, and after drying they were milled in a model SK 100 Cross Beater RETSCH mill to a uniformly fine powder.

2.2. Determination of total fat and FA composition

Total fat extraction was performed according to the published AOAC (2000) methods, using a Soxhlet apparatus for 16 h with petroleum ether as the extraction solvent. The solvent was removed by rotary evaporation and the residue was dried for 1 h at 105 °C.

FA profiles were determined according the EU Regulation 2568/91 (European Union Commission, 1991) method, i.e., conversion of the free FAs released to their methyl esters (FAME) following the method of Hartman and Lago (1973). The sample preparation for injection was carried out as follow: 1 g of total lipids, 10 mL of *n*-heptane, p.a. and 0.5 mL of methanol solution of KOH 2 N. This mixture was shaken and centrifuged. After derivatization a 0.45 µL sample of each FAME preparation was injected into the gas chromatograph (GC). All samples were run in duplicate. The chromatography separation was performed using a Perkin-Elmer Auto-system GC equipped with a flame ionization detector, with a capillary column, SUPELCO sptm-2380, 60 m length, 0.25 mm i.d. and 0.20 µm film thickness. The gas chromatography conditions: oven temperature program –175 °C for 25 min and 220 °C for 10 min at a rate of 5 °C/min. The injector temperature was 240 °C and the detector temperature was 250 °C. The head pressure of the carrier gas helium was maintained at 20 psi; the column flow rate was 5 mL/min. Identification and quantification of the detected FAME was carried out against external standards after adjustment through regression lines. The FA methyl esters were identified by a comparison of retention times to the FAME standards (Reg. CEE 1989/08–10). The results are expressed in relative percentage for each FA, calculated by internal normalization of the chromatographic peak area.

The FAs composition for each cultivar was examined in terms of saturated acids: myristic acid (C14:0), pentadecanoic (C15:0), palmitic (C16:0), margaric (C17:0), stearic (C18:0), arachidic (C20:0), behenic (C22:0) and lignoceric (C24:0) and unsaturated acids: palmitoleic (C16:1), margaroleic (C17:1), oleic (C18:1), linoleic (C18:2), linolenic (C18:3), and gadoleic (C20:1).

FA composition was expressed as the percentage of total FA. Values presented in the tables are the average values of three individual samples, *n* = 3, each sample analyzed in triplicate.

2.3. Statistical analysis

The analyses of data were performed as analysis of variance using the Super ANOVA software (1.1, Abacus Concepts Inc., 1991). Mean separations were made using Fisher's Protected LSD Test (*P* = 0.05), designed to allow all possible linear combination of group means to be tested.

3. Results and discussion

The results of the total crude fat content of 17 chestnut cultivars are presented in Table 2. The crude fat content was significantly affected by cultivar and crop year but did not show significant differences among PDO areas. Crude fat content was higher in 2002 (2.52%) than in 2001

Table 2
Crude fat of 17 chestnuts cultivars from the three PDO areas from 2 separate years

Cultivar	PDO	Year	Crude fat (%DM)
Aveleira	TF	2001	2.57 ± 0.06 f
Boaventura	TF	2001	2.40 ± 0.10 e
Lamela	TF	2001	3.13 ± 0.06 I
Longal	TF	2001	2.53 ± 0.06 ef
Rebordã	TF	2001	1.73 ± 0.06 a
Redonda	TF	2001	2.73 ± 0.06 gh
Trigueira	TF	2001	2.60 ± 0.00 fg
Zeive	TF	2001	2.73 ± 0.06 gh
Cancela	SL	2001	2.77 ± 0.15 h
Carreiró	SL	2001	2.03 ± 0.15 c
Demanda	SL	2001	3.10 ± 0.10 I
Ferreirinha	SL	2001	2.77 ± 0.15 h
Longal	SL	2001	1.97 ± 0.06 bc
Martainha	SL	2001	2.50 ± 0.10 ef
Riscada	SL	2001	2.20 ± 0.17 b
Cota	P	2001	1.83 ± 0.06 ab
Judia	P	2001	2.63 ± 0.06 fgh
Lada	P	2001	1.77 ± 0.06 a
Aveleira	TF	2002	3.50 ± 0.27 k
Boaventura	TF	2002	2.57 ± 0.12 fg
Lamela	TF	2002	3.37 ± 0.06 kl
Longal	TF	2002	2.43 ± 0.12 ef
Rebordã	TF	2002	2.13 ± 0.06 cd
Redonda	TF	2002	2.40 ± 0.00 def
Trigueira	TF	2002	3.07 ± 0.06 ij
Zeive	TF	2002	2.73 ± 0.06 gh
Cancela	SL	2002	2.80 ± 0.10 ghi
Carreiró	SL	2002	2.10 ± 0.36 bc
Demanda	SL	2002	3.13 ± 0.06 j
Ferreirinha	SL	2002	2.87 ± 0.06 hij
Longal	SL	2002	2.20 ± 0.00 cde
Martainha	SL	2002	2.60 ± 0.17 fgh
Riscada	SL	2002	2.20 ± 0.20 cde
Cota	P	2002	1.83 ± 0.15 ab
Judia	P	2002	2.57 ± 0.40 fg
Lada	P	2002	1.67 ± 0.06 a
<i>P</i> > 0.001	<i>P</i> > 0.05	<i>P</i> > 0.01	

Different letters within one column denote statistically significant differences (*P* < 0.05) by ANOVA and Fisher's LSD test.

Values are average of three individual samples each analysed in triplicate ± standard deviation.

(2.40%), with ranges from 1.67% for cv. Lada (PDO Padrela) in 2002 to 3.50% for cv. Aveleira (PDO Terra Fria) in the same year. These results are higher than the values reported by [Ferreira-Cardoso \(2002\)](#) with an average of 1.3% but lower than the values of 3.55% reported by [Desmaison and Adrian \(1986\)](#) and 3.6% reported by [McCarthy and Meredith \(1988\)](#) for European chestnuts. These authors also presented data for other chestnut sources with crude fat values of 2.34% for American chestnuts and 1.98% for Chinese chestnuts. [Üstün et al. \(1999\)](#), studying 55 different types of chestnuts grown in Turkey, reported values between 0.66% and 3.08% for crude fat content. Our results confirm that chestnuts have low crude fat content compared with almonds, with values of between 36% and 53% ([Abdallah et al., 1998](#)), and hazelnuts, with values of between 46.7% and 76.8% ([Parcerisa et al., 1993](#)). Some of these differences may reflect the influence of environmental conditions and orchard management techniques.

The analysis of variance for the individual FAs ([Table 3](#)) showed significant differences for cultivar, PDO, year and the interaction of cultivar $\times$ year for the majority of samples. The source of variation “year” did not significantly affect the content of pentadecanoic, palmitic, margaric, stearic or lignoceric acids. The variation in pentadecanoic and lignoceric acids was independent of the PDO area. The content of total FA of all the cultivars studied from the three PDO areas in the 2 years is shown in [Table 4](#). The cv. Boaventura showed the lowest value (97.25 g/100 g), while cv. Zeive (99.87 g/100 g) had the

highest. These minimum and maximum values were found for genotypes within PDO Terra Fria. The second year of this study showed that samples from within this PDO had the highest values of all three PDO.

Chestnuts samples analyzed contained 17% saturated fatty acids (SFA), 83% unsaturated fatty acids (USFA) consisting of 31% monounsaturated fatty acids (MUFA) and 52% of polyunsaturated fatty acids (PUFA) ([Fig. 2](#)). The proportions of USFA are similar to those reported by [Ferreira-Cardoso \(2002\)](#), [Senter et al. \(1994\)](#) and [Desmaison and Adrian \(1986\)](#) with reported USFA of 80.5%, 83% and 75%, respectively.

The results presented in [Tables 4 and 5](#) show that chestnut crude fat consists primarily of three FAs: linoleic, oleic and palmitic. These accounted for more than 85% of the total FA content, slightly higher than the 85% reported by [Desmaison and Adrian \(1986\)](#).

The effect of cultivar and locations (PDO) on total SFA content was statistically significant as shown in [Table 3](#). The cv. Aveleira had the lowest content (14.08 g/100 g) in 2001 and cv. Carreiró (PDO Soutos da Lapa) in 2002 the highest content (18.64 g/100 g) ([Table 4](#)). The PDO Padrela had the lowest value (15.45 g/100 g), and PDO Soutos da Lapa (16.91 g/100 g) the highest value in the same year ([Table 6](#)). These values did not change significantly with harvest years. The SFA give the fat more stability, but they are considered harmful to the heart and blood vessels, so with respect to the SFA content, the cv. Aveleira is potentially the most beneficial to the human diet because it had the lowest SFA value.

The most important SFA was palmitic acid, with values between 12.54 and 16.80 g/100 g for cv. Aveleira in 2001 and cv. Carreiró in 2002, respectively ([Table 4](#)). These values were consistently higher than the levels for the other saturated acids, which had relatively low values. These results are in agreement with those reported by [Desmaison and Adrian \(1986\)](#) and [Künsch et al. \(1999\)](#) for five Swiss cultivars and one Italian cultivar. Also, in other important crops in Mediterranean countries, such as olives, palmitic acid was the predominant SFA ([Aguilera et al., 2005](#)). Palmitic acid has been recognized as a major contributor to the build-up of serum cholesterol ([Groff et al., 1996](#)).

Concerning the total of MUFA, this varied significantly with cultivars, PDO and year ([Table 3](#)). The cv. Cota (PDO Padrela) had the lowest MUFA value (22.46 g/100 g) in 2001, and cv. Trigueira (PDO Terra Fria) the highest value (39.29 g/100 g) in 2002 ([Table 5](#)). The major MUFA in all genotypes tested was oleic acid, with ranges from 20.66 g/100 g for cv. Cota in 2001 to 37.60 g/100 g for cv. Trigueira in the 2002. The MUFA also varied significantly with locations: the PDO Padrela had the lowest MUFA value (25.94 g/100 g) while PDO Terra Fria had the highest (35.13 g/100 g) ([Table 6](#)). Recently it has been reported that MUFA were as effective as PUFA in the reduction of low-density-lipoprotein cholesterol in humans ([Mensink and Katan, 1989](#)).

Table 3

Summary of analysis of variance for fatty acid composition for year, cultivar, PDO and interaction year $\times$ cultivar

	Cultivar	PDO	Year	Cultivar $\times$ year
C14:0	***	**	**	***
C15:0	***	Ns	Ns	*
C16:0	***	***	Ns	***
C17:0	***	***	Ns	*
C17:1	***	***	***	Ns
C18:0	***	**	Ns	***
C20:0	***	***	*	***
C22:0	***	**	***	***
C24:0	***	Ns	Ns	Ns
C16:1	***	**	***	**
C18:1	***	***	***	***
C18:2	***	***	***	***
C18:3	***	***	***	***
C20:1	***	***	**	*
Σ SFA	***	***	Ns	***
MUFA	***	***	***	***
PUFA	***	***	***	***
USFA	***	**	***	***
MUFA/PUFA	***	***	***	***
C18:1/C18:2	***	***	***	***
Total	***	Ns	***	***

***, ** and * significant at $P < 0.001$, 0.01 or 0.05, respectively. Ns = not significant ($P > 0.05$).

Table 4
Total fatty acids and SFA composition (%) of 17 chestnuts cultivars from the three PDO areas in the 2 cropping years

Cultivar	PDO	Year	Total fatty acids	C14:0	C15:0	C16:0	C17:0	C18:0	C20:0	C22:0	C24:0	ΣSFA
Aveleira	TF	2001	99.51±0.29 hi	0.12±0.01 ab	0.08±0.01 a	12.54±0.10 a	0.10±0.01 a	0.72±0.02 a	0.25±0.01 cde	0.18±0.00 c	0.08±0.01 bc	14.08±0.07 a
Boaventura	TF	2001	97.25±0.21 a	0.16±0.03 bcd	0.21±0.03 j	15.37±0.33 hi	0.14±0.02 bcd	0.85±0.13 cd	0.25±0.03 bc	0.22±0.02 fgh	0.15±0.03 f	17.35±0.18 hi
Lamela	TF	2001	99.12±0.36 fg	0.13±0.04 ab	0.13±0.01 cdef	15.59±0.10 i	0.14±0.01 bcd	1.02±0.05 i	0.28±0.02 efg	0.15±0.03 b	0.07±0.01 ab	17.52±0.11 hi
Longal	TF	2001	99.20±0.31 fgh	0.12±0.00 ab	0.09±0.01 abc	13.72±0.12 cd	0.13±0.00 b	0.88±0.01 def	0.28±0.01 efg	0.18±0.01 c	0.07±0.01 ab	15.48±0.13 cde
Rebordá	TF	2001	99.24±0.34 gh	0.25±0.06 f	0.18±0.05 ghi	14.26±0.04 e	0.13±0.01 bc	0.83±0.01 bcd	0.25±0.01 bc	0.19±0.01 cdef	0.09±0.01 bc	16.17±0.15 f
Redonda	TF	2001	99.23±0.40 gh	0.20±0.12 de	0.12±0.0 bcde	13.69±0.01 cd	0.10±0.00 a	0.79±0.01 bc	0.27±0.01 cdef	0.19±0.01 cd	0.07±0.01 ab	15.43±0.01 bcd
Trigueira	TF	2001	98.08±0.10 b	0.11±0.01 a	0.08±0.03 a	15.52±0.20 hi	0.14±0.00 bcd	0.85±0.01 cde	0.25±0.01 cd	0.19±0.01 cd	0.08±0.01 bc	17.22±0.17 h
Zeive	TF	2001	98.58±0.17 de	0.24±0.04 f	0.16±0.01 fgh	14.68±0.66 f	0.14±0.02 bcd	0.92±0.01 efg	0.27±0.02 defg	0.23±0.01 ghi	0.19±0.00 g	16.83±0.65 g
Cancela	SL	2001	98.86±0.07 ef	0.17±0.03 cde	0.11±0.02 bcd	14.84±0.10 fg	0.15±0.01 de	0.99±0.01 ghi	0.29±0.01 gh	0.20±0.01 cdefg	0.08±0.00 bc	16.84±0.11 g
Carreiró	SL	2001	99.70±0.03 i	0.30±0.01 g	0.19±0.02 hij	16.73±0.10 j	0.17±0.01 e	0.77±0.01 ab	0.23±0.02 b	0.12±0.01 a	0.05±0.01 a	18.57±0.10 j
Demanda	SL	2001	98.47±0.08 cd	0.14±0.01 abc	0.10±0.01 abc	15.18±0.03 gh	0.14±0.00 bcd	1.06±0.01 i	0.31±0.01 h	0.21±0.00 defgh	0.09±0.01 bc	17.23±0.01 h
Ferreirinha	SL	2001	98.17±0.03 bc	0.30±0.01 g	0.20±0.01 ij	15.29±0.01 hi	0.20±0.01 f	1.01±0.05 hi	0.28±0.01 fg	0.20±0.01 cdefg	0.11±0.02 de	17.61±0.44 i
Longal	SL	2001	98.20±0.13 efg	0.14±0.01 abc	0.14±0.01 def	13.81±0.11 d	0.14±0.01 b	0.99±0.06 ghi	0.26±0.01 cde	0.21±0.01 efg	0.09±0.01 bcd	15.80±0.86 e
Martinha	SL	2001	98.5±0.06 cd	0.14±0.01 abc	0.10±0.00 bc	15.18±0.03 gh	0.14±0.00 bcd	1.06±0.06 i	0.31±0.01 h	0.21±0.01 defgh	0.09±0.02 bc	17.23±0.01 h
Riscada	SL	2001	99.10±1.19 fg	0.14±0.01 abc	0.15±0.02 efg	13.26±0.35 b	0.15±0.03 bcd	0.80±0.10 bc	0.25±0.03 bc	0.23±0.04 hi	0.12±0.03 e	15.10±0.40 b
Cota	P	2001	97.99±0.79 b	0.23±0.04 ef	0.18±0.02 ghi	13.73±0.31 cd	0.14±0.00 bcd	0.70±0.01 a	0.20±0.01 a	0.19±0.01 cde	0.07±0.02 ab	15.45±0.56 bcde
Judia	P	2001	99.50±0.03 hi	0.11±0.01 a	0.14±0.03 ef	13.40±0.05 bc	0.13±0.00 b	0.84±0.01 bcd	0.26±0.01 cdef	0.18±0.01 c	0.10±0.01 cd	15.17±0.25 bc
Lada	P	2001	99.17±0.17 fgh	0.20±0.00 de	0.15±0.01 fg	13.51±0.32 bcd	0.15±0.00 cde	0.94±0.02 fgh	0.26±0.02 cde	0.24±0.03 i	0.11±0.01 de	15.56±0.23 de
Aveleira	TF	2002	99.84±0.08 hi	0.13±0.01 a	0.09±0.01 a	13.02±0.08 ab	0.12±0.01 abc	0.80±0.03 bc	0.31±0.08 gh	0.27±0.04 cde	0.11±0.02 bc	14.85±0.08 ab
Boaventura	TF	2002	99.35±0.44 cdef	0.14±0.01 abcd	0.17±0.04 gh	15.96±0.39 hi	0.15±0.01 defg	1.00±0.01 f	0.31±0.05 gh	0.24±0.01 bcd	0.14±0.03 c	18.14±0.44 hi
Lamela	TF	2002	99.73±0.32 efg	0.13±0.01 abc	0.13±0.02 cdef	15.60±0.28 gh	0.14±0.01 defg	1.00±0.05 f	0.27±0.01 cdefg	0.19±0.02 ab	0.08±0.02 ab	17.55±0.90 g
Longal	TF	2002	99.75±0.20 fghi	0.12±0.01 a	0.10±0.01 ab	12.67±0.16 a	0.12±0.02 abc	0.85±0.04 d	0.30±0.02 efg	0.42±0.05 f	0.09±0.01 ab	14.67±0.20 a
Rebordá	TF	2002	99.40±0.42 cdefg	0.16±0.02 cde	0.12±0.02 abcd	13.96±0.40 d	0.10±0.02 a	0.80±0.01 b	0.24±0.02 abcd	0.32±0.05 e	0.11±0.02 bc	15.81±0.42 d
Redonda	TF	2002	99.44±0.20 cdefgh	0.12±0.01 a	0.10±0.01 ab	13.32±0.13 bc	0.11±0.03 ab	0.78±0.01 b	0.30±0.01 efg	0.25±0.03 bcd	0.07±0.0 ab	15.05±0.20 abc
Trigueira	TF	2002	99.58±0.26 defghi	0.12±0.01 ab	0.11±0.02 abc	16.13±0.18 i	0.15±0.02 defg	1.12±0.03 i	0.34±0.01 h	0.24±0.06 bcd	0.09±0.02 abc	18.31±0.26 i
Zeive	TF	2002	99.87±0.40 i	0.18±0.01 ef	0.23±0.01 h	15.23±0.32 fg	0.16±0.01 gh	1.08±0.03 hi	0.30±0.01 efg	0.33±0.08 e	0.19±0.03 d	17.69±0.40 gh
Cancela	SL	2002	98.92±0.03 b	0.18±0.02 ef	0.11±0.02 abc	14.83±0.02 ef	0.16±0.02 fg	0.99±0.03 ef	0.29±0.02 defg	0.20±0.00 bc	0.08±0.00 ab	16.84±0.03 ef
Carreiró	SL	2002	99.76±0.05 ghi	0.31±0.03 h	0.19±0.02 efg	16.80±0.04 j	0.18±0.01 h	0.77±0.02 b	0.22±0.02 ab	0.12±0.00 a	0.05±0.00 a	18.64±0.05 i
Demanda	SL	2002	98.48±0.02 a	0.14±0.01 abcd	0.11±0.02 ab	15.19±0.02 efg	0.14±0.00 cdef	1.06±0.03 gh	0.31±0.01 fgh	0.21±0.01 bcd	0.08±0.01 ab	17.24±0.02 fg
Ferreirinha	SL	2002	98.17±0.03 a	0.31±0.03 h	0.20±0.00 gh	15.29±0.02 fg	0.20±0.01 i	1.01±0.03 fg	0.29±0.01 defg	0.21±0.02 bcd	0.11±0.02 bc	17.63±0.03 gh
Longal	SL	2002	99.32±0.99 bcde	0.13±0.01 abc	0.14±0.02 abcde	13.33±1.06 bc	0.13±0.01 bcd	0.87±0.05 d	0.25±0.02 bcde	0.24±0.07 bcd	0.09±0.02 abc	15.19±0.99 abc
Martinha	SL	2002	99.31±0.57 bcd	0.17±0.02 de	0.12±0.02 abcde	14.64±0.47 e	0.13±0.01 bcd	0.86±0.03 d	0.23±0.04 abc	0.22±0.06 bcd	0.07±0.03 ab	16.44±0.57 e
Riscada	SL	2002	99.45±0.63 cdefgh	0.15±0.03 bcd	0.14±0.01 bcdef	13.27±0.49 bc	0.14±0.01 defg	0.79±0.04 bc	0.25±0.03 bcde	0.23±0.05 bcd	0.13±0.07 c	15.12±0.63 abc
Cota	P	2002	99.65±0.10 defghi	0.23±0.02 g	0.17±0.01 efg	13.70±0.10 cd	0.14±0.00 cdef	0.70±0.03 a	0.20±0.02 a	0.19±0.03 ab	0.07±0.02 ab	15.40±0.10 bcd
Judia	P	2002	99.51±0.11 cdefghi	0.12±0.01 a	0.14±0.01 bcdef	13.41±0.09 bc	0.13±0.01 bcd	0.84±0.00 cd	0.26±0.02 bcdef	0.18±0.00 ab	0.10±0.00 bc	15.17±0.11 abc
Lada	P	2002	99.16±0.09 bc	0.19±0.02 f	0.16±0.03 cdefg	13.48±0.21 bcd	0.15±0.01 efg	0.94±0.01 e	0.26±0.04 bcde	0.24±0.02 bcd	0.12±0.01 bc	15.54±0.09 cd

SFA = saturated fatty acid.

Different letters within one column denote statistically significant differences ($P<0.05$) by ANOVA and Fisher's LSD test.

Values are average of three individual samples each analysed in triplicate ± standard deviation.

Fig. 2. Portuguese chestnuts: proportion of saturated (SFA), monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids.

The total PUFA content varied significantly with cultivars, PDO and year (Table 3). The cv. Trigueira had the lowest PUFA value (41.98 g/100 g) in 2002 and cv. Cota the highest value (60.11 g/100 g) in the same year (Table 5). Significant differences were found among PDO regions: Terra Fria had the lowest PUFA values (48.02 g/100 g) while Padrela had the highest values (57.61 g/100 g) in the same year (Table 6). Linoleic acid, the most important PUFA, had a value of 37.57 g/100 g in cv. Trigueira in 2002 and 50.93 g/100 g in cv. Lada in the same year (Table 5). Also, in other nut crops such as hazelnut and almond, the most important PUFA is linoleic acid, with values between 5.7 g/100 g and 22.2 g/100 g in hazelnuts (Abdallah et al., 1998) and between 15.4 g/100 g and 22.2 g/100 g in almonds (Parcerisa et al., 1993). It is generally accepted that the most important FA in chestnut fruit is linoleic acid (Ferreira-Cardoso, 2002; Desmaison and Adrian, 1986; Künsch et al., 1999; Senter et al., 1994) which, being an essential FA, has clear effects on human health. Consumers are especially interested in essential FAs, with emphasis on the health potential of PUFA. It is considered that these FAs play a natural preventive role in cardiovascular disease (Melgarejo and Artes, 2000). From these studies two potential health-promoting cultivars were identified: cv. Lada, with higher linoleic acid, and cv. Aveleira, with more SFA. With respect to the PDO areas, the Padrela

consistently had the highest values of linoleic acid and SFA.

The ratio value of MUFA to PUFA was less than 1, which means a predominance of PUFA, in contrast to almond which presents low PUFA and high MUFA (Abdallah et al., 1998). This ratio varied significantly ($P < 0.001$) among genotypes, years and production regions (Table 3), with the minimum ratio value being 0.37 for cv. Cota in 2001 and the maximum value of 0.94 in cv. Trigueira in 2002 (Table 7). With respect to the locations, the average values for the ratio was within the range 0.45–0.46 for PDO Padrela, 0.53–0.57 for PDO Soutos da Lapa and 0.69–0.74 for PDO Terra Fria in the years 2001 and 2002, respectively (Table 6).

Like the MUFA:PUFA ratio, the oleic:linoleic ratio was very low with significant differences among cultivar, PDO and year (Table 3). This ratio was lowest for cv. Cota (0.41) in 2001 and highest for cv. Trigueira (1.00) in 2002 (Table 7). The samples from PDO Padrela had the lowest average value (0.50) and Terra Fria the highest (0.80) (Table 6).

Thus the pool of oleic acid appears to be controlled by its conversion to linoleic, probably as a result of the enzymatic activity of oleic desaturase. According to Garcia et al. (1992) this enzyme controls the variation in FA composition in pistachio.

Table 5
Unsaturated fatty acid (USFA) composition (%), of 17 chestnuts cultivars from the three PDO areas on 2 years

Cultivar	PDO	Year	C16:1	C17:1	C18:1	C20:1	ΣMUFA	C18:2	C18:3	ΣPUFA	ΣUSFA
Aveira	TF	2001	0.74±0.00 a	0.09±0.01 ef	34.16±0.07 m	0.73±0.01 j	35.71±0.72 k	44.33±0.03 cd	5.38±0.01 d	49.71±0.04 c	85.42±0.06 m
Boaventura	TF	2001	1.19±0.27 e	0.10±0.01 gh	35.55±0.30 n	0.55±0.04 cde	37.38±0.13 l	37.83±0.21 a	4.68±0.08 b	42.51±0.29 a	79.90±0.17 a
Lamela	TF	2001	0.89±0.02 bc	0.08±0.01 bcd	36.90±0.35 o	0.65±0.01 i	38.51±0.32 m	38.52±0.45 b	4.57±0.09 a	43.09±0.53 b	81.60±0.30 ef
Longal	TF	2001	0.80±0.01 ab	0.08±0.01 def	31.51±0.08 l	0.56±0.00 def	32.96±0.80 j	45.05±0.08 f	5.71±0.01 ef	50.76±0.09 e	83.72±0.16 jk
Rebordá	TF	2001	0.92±0.02 bc	0.07±0.00 b	28.21±0.05 h	0.64±0.01 hi	29.83±0.03 g	46.28±0.24 h	6.96±0.01 i	53.23±0.23 i	83.07±0.20 i
Redonda	TF	2001	0.81±0.01 ab	0.07±0.01 bc	31.33±0.04 l	0.65±0.01 I	32.86±0.04 j	44.68±0.03 e	6.26±0.02 h	50.94±0.02 ef	83.80±0.03 jk
Trigueira	TF	2001	1.26±0.01 ef	0.08±0.01 def	28.72±0.05 i	0.54±0.02 cd	30.60±0.06 h	45.26±0.06 f	5.00±0.01 c	50.26±0.06 d	80.86±0.11 bc
Zeive	TF	2001	1.01±0.02 cd	0.08±0.00 cde	27.41±0.26 f	0.73±0.05 j	29.23±0.29 f	46.79±0.30 i	5.74±0.06 f	52.52±0.33 h	81.75±0.62 fg
Cancela	SL	2001	0.93±0.02 bc	0.09±0.00 f	29.21±0.02 j	0.50±0.01 b	30.73±0.02 h	44.20±0.04 c	7.10±0.01 k	51.30±0.05 g	82.03±0.06 g
Carreiró	SL	2001	0.96±0.01 c	0.10±0.01 gh	25.95±0.12 d	0.44±0.02 a	27.46±0.14 d	46.57±0.04 hi	7.10±0.06 k	53.67±0.06 j	81.13±0.12 cd
Demanda	SL	2001	0.99±0.02 cd	0.09±0.01 f	28.31±0.02 h	0.58±0.00 efg	29.98±0.01 g	45.63±0.11 g	5.64±0.01 e	51.26±0.12 fg	81.24±0.08 cde
Ferreirinha	SL	2001	1.38±0.01 h	0.11±0.00 h	27.89±0.02 g	0.56±0.01 cdef	29.94±0.01 g	44.60±0.02 de	6.02±0.02 g	50.62±0.01 e	80.56±0.02 b
Longal	SL	2001	0.98±0.03 c	0.08±0.01 def	21.50±0.08 b	0.52±0.00 bc	23.08±0.03 b	50.62±0.08 m	9.42±0.01 n	60.04±0.10 l	83.12±0.07 i
Martainha	SL	2001	0.99±0.01 cd	0.08±0.01 cde	28.31±0.02 h	0.58±0.00 def	29.88±0.02 d	45.63±0.04 g	5.67±0.01 f	51.30±0.04 g	81.27±0.06 de
Riscada	SL	2001	1.02±0.18 cd	0.10±0.01 g	26.95±0.40 e	0.56±0.05 def	28.63±0.55 e	48.18±0.44 j	7.19±0.05 l	55.37±0.48 k	82.00±0.80 kl
Cota	P	2001	1.14±0.12 de	0.06±0.00 a	20.66±0.11 a	0.61±0.04 gh	22.46±0.24 a	50.07±0.21 l	10.00±0.12 o	60.07±0.30 l	84.53±0.46 h
Judia	P	2001	0.73±0.02 a	0.08±0.00 cde	30.02±0.02 k	0.59±0.01 fg	31.42±0.04 i	46.31±0.01 h	6.60±0.01 i	52.90±0.02 l	84.32±0.04 l
Lada	P	2001	0.87±0.06 abc	0.09±0.00 f	22.27±0.07 c	0.58±0.04 efg	23.82±0.05 c	50.93±0.35 m	8.86±0.08 m	59.79±0.39 l	83.61±0.43 j
Aveira	TF	2002	0.70±0.02 ab	0.10±0.01 gh	37.40±0.14 j	0.74±0.01 f	38.95±0.15 j	41.01±0.30 c	5.04±0.19 c	46.04±0.49 d	85.00±0.34 gh
Boaventura	TF	2002	1.25±0.10 l	0.12±0.01 i	35.43±0.19 i	0.58±0.04 cd	37.37±0.29 i	39.04±0.53 b	4.80±0.08 b	43.83±0.58 c	81.21±0.87 b
Lamela	TF	2002	0.88±0.09 defgh	0.08±0.01 bcde	37.36±0.38 j	0.63±0.01 de	38.95±0.28 j	38.74±0.58 b	4.49±0.095 a	43.23±0.66 b	82.18±0.41 c
Longal	TF	2002	0.62±0.06 a	0.08±0.01 bcd	31.18±0.39 g	0.62±0.03 de	32.50±0.38 g	45.64±0.20 g	6.95±0.05 g	52.59±0.39 ij	85.09±0.24 h
Rebordá	TF	2002	0.78±0.04 bcd	0.08±0.01 bc	28.96±0.50 e	0.57±0.06 cd	30.38±0.54 de	45.92±0.43 gh	7.29±0.09 hi	53.21±0.51 jk	83.59±0.75 e
Redonda	TF	2002	0.70±0.05 ab	0.07±0.00 ab	31.09±0.48 g	0.67±0.01 e	32.54±0.44 g	44.66±0.38 ef	7.20±0.02 hi	51.85±0.39 h	84.39±0.38 fg
Trigueira	TF	2002	0.98±0.02 hi	0.09±0.06 efg	37.60±0.30 j	0.62±0.02 de	39.29±0.31 j	37.57±0.37 a	4.40±0.07 a	41.98±0.39 a	81.27±0.22 b
Zeive	TF	2002	0.79±0.03 bcde	0.10±0.01 fg	30.48±0.18 f	0.81±0.02 g	32.18±0.18 g	44.87±0.47 f	5.13±0.17 c	50.00±0.55 f	82.17±0.48 c
Cancela	SL	2002	0.94±0.03 fghi	0.09±0.0 def	29.22±0.03 e	0.50±0.02 ab	30.75±0.05 e	44.24±0.06 e	7.10±0.02 gh	51.33±0.05 l	82.08±0.07 c
Carreiró	SL	2002	0.97±0.02 ghi	0.10±0.00 fgh	25.94±0.10 b	0.44±0.02 a	27.45±0.09 b	46.57±0.03 i	7.09±0.08 gh	53.66±0.08 k	81.11±0.04 ab
Demanda	SL	2002	0.99±0.02 ij	0.09±0.01 efg	28.31±0.03 d	0.58±0.00 cd	29.98±0.02 d	45.63±0.08 g	5.64±0.01 d	51.27±0.09 h	81.25±0.07 b
Ferreirinha	SL	2002	1.38±0.01 m	0.11±0.00 hi	27.89±0.02 d	0.55±0.03 bc	29.92±0.03 d	44.60±0.02 ef	6.02±0.01 e	50.62±0.03 g	80.54±0.02 a
Longal	SL	2002	0.84±0.01 cdef	0.09±0.01 efg	27.04±0.03 c	0.62±0.03 cd	28.55±0.04 c	48.22±0.31 j	7.37±0.05 i	55.59±0.36 l	84.13±0.39 ef
Martainha	SL	2002	0.88±0.11 efgh	0.09±0.01 efg	32.61±0.28 h	0.62±0.03 de	34.20±0.03 h	43.57±0.09 d	5.10±0.24 c	48.67±0.32 e	82.87±0.47 d
Riscada	SL	2002	1.09±0.19 jk	0.11±0.02 hi	26.96±0.48 c	0.56±0.09 cd	28.71±0.38 c	48.43±0.51 j	7.19±0.08 hi	55.62±0.44 l	84.23±0.71 h
Cota	P	2002	1.15±0.01 kl	0.06±0.01 a	22.32±0.10 a	0.60±0.04 cd	24.14±0.29 a	50.09±0.07 k	10.02±0.07 k	60.11±0.05 m	84.25±0.17 f
Judia	P	2002	0.75±0.01 bc	0.08±0.00 bcd	30.03±0.55 f	0.59±0.01 cd	31.44±0.53 f	46.30±0.03 gh	6.60±0.03 f	52.90±0.06 hi	84.34±0.49 h
Lada	P	2002	0.87±0.06 defg	0.09±0.01 def	22.25±0.10 a	0.60±0.10 cd	23.81±0.17 a	50.93±0.44 l	8.87±0.11 j	59.81±0.37 m	83.62±0.32 e

Different letters within one column denote statistically significant differences ($P<0.05$) by ANOVA and Fisher's LSD test.

Values are average of three individual samples each analysed in triplicate ± standard deviation.

MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids.

Table 6

SFA, USFA, MUFA, PUFA, ratios MUFA to PUFA and C18:1 (oleic acid) to C18:2 (linoleic acid) and crude fat content, of the three PDO areas on 2 years ($n = 3$; mean $\pm$ SD)

PDO	Year	SFA	USFA	MUFA	PUFA	MUFA/PUFA	C18:1/C18:2	Crude fat (% DM)
TF	2001	16.36 $\pm$ 1.19 a	82.48 $\pm$ 1.73 ab	33.31 $\pm$ 3.34 b	49.18 $\pm$ 3.83 a	0.69 $\pm$ 0.13 b	0.74 $\pm$ 0.14 b	2.52 $\pm$ 0.80 a
SL	2001	17.14 $\pm$ 1.19 b	81.79 $\pm$ 1.35 a	28.11 $\pm$ 2.45 a	53.68 $\pm$ 3.14 b	0.53 $\pm$ 0.07 a	0.57 $\pm$ 0.07 a	2.52 $\pm$ 0.70 a
P	2001	15.55 $\pm$ 0.39 a	83.41 $\pm$ 0.83 b	25.82 $\pm$ 4.18 a	57.59 $\pm$ 3.52 c	0.45 $\pm$ 0.10 a	0.50 $\pm$ 0.01 a	2.60 $\pm$ 0.16 a
TF	2002	16.56 $\pm$ 1.54 b	83.15 $\pm$ 3.92 a	35.13 $\pm$ 3.55 c	48.02 $\pm$ 4.58 a	0.74 $\pm$ 0.13 c	0.80 $\pm$ 0.13 c	2.38 $\pm$ 0.12 a
SL	2002	16.91 $\pm$ 1.19 b	82.21 $\pm$ 1.45 a	29.84 $\pm$ 2.08 b	52.37 $\pm$ 2.55 b	0.57 $\pm$ 0.07 b	0.62 $\pm$ 0.69 b	2.36 $\pm$ 0.08 a
P	2002	15.45 $\pm$ 0.19 a	83.55 $\pm$ 0.95 a	25.94 $\pm$ 4.36 a	57.61 $\pm$ 3.54 c	0.46 $\pm$ 0.11 a	0.50 $\pm$ 0.12 a	2.50 $\pm$ 0.17 a

MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; SFA = saturated fatty acids; USFA = unsaturated fatty acids. Different letters within one column denote statistically significant differences ($P < 0.05$) by ANOVA and Fisher's LSD test.

Table 7

Ratios MUFA to PUFA and C18:1 to C18:2

Cultivar	PDO	Year	MUFA/PUFA	C18:1/C18:2
Aveleira	TF	2001	0.72 $\pm$ 0.00 i	0.77 $\pm$ 0.02 l
Boaventura	TF	2001	0.88 $\pm$ 0.01 j	0.94 $\pm$ 0.01 m
Lamela	TF	2001	0.89 $\pm$ 0.02 k	0.96 $\pm$ 0.02 n
Longal	TF	2001	0.65 $\pm$ 0.00 h	0.70 $\pm$ 0.00 k
Rebordã	TF	2001	0.56 $\pm$ 0.00 d	0.61 $\pm$ 0.00 h
Redonda	TF	2001	0.65 $\pm$ 0.00 h	0.70 $\pm$ 0.00 k
Trigueira	TF	2001	0.61 $\pm$ 0.00 g	0.63 $\pm$ 0.00 h
Zeive	TF	2001	0.56 $\pm$ 0.00 d	0.59 $\pm$ 0.00 e
Cancela	SL	2001	0.60 $\pm$ 0.00 fg	0.66 $\pm$ 0.00 j
Carreiró	SL	2001	0.51 $\pm$ 0.00 c	0.56 $\pm$ 0.00 d
Demanda	SL	2001	0.58 $\pm$ 0.00 f	0.62 $\pm$ 0.00 g
Ferreirinha	SL	2001	0.59 $\pm$ 0.00 ef	0.63 $\pm$ 0.00 gh
Longal	SL	2001	0.38 $\pm$ 0.00 a	0.42 $\pm$ 0.00 b
Martainha	SL	2001	0.58 $\pm$ 0.00 e	0.62 $\pm$ 0.00 g
Riscada	SL	2001	0.52 $\pm$ 0.01 c	0.56 $\pm$ 0.01 d
Cota	P	2001	0.37 $\pm$ 0.00 a	0.41 $\pm$ 0.00 a
Judia	P	2001	0.59 $\pm$ 0.02 ef	0.65 $\pm$ 0.00 I
Lada	P	2001	0.40 $\pm$ 0.00 b	0.44 $\pm$ 0.00 c
Aveleira	TF	2002	0.85 $\pm$ 0.01 h	0.91 $\pm$ 0.01 h
Boaventura	TF	2002	0.85 $\pm$ 0.01 h	0.91 $\pm$ 0.01 h
Lamela	TF	2002	0.90 $\pm$ 0.02 i	0.97 $\pm$ 0.02 i
Longal	TF	2002	0.62 $\pm$ 0.01 def	0.68 $\pm$ 0.01 ef
Rebordã	TF	2002	0.57 $\pm$ 0.01 c	0.63 $\pm$ 0.01 cd
Redonda	TF	2002	0.63 $\pm$ 0.01 ef	0.70 $\pm$ 0.02 fg
Trigueira	TF	2002	0.94 $\pm$ 0.02 i	1.00 $\pm$ 0.02 i
Zeive	TF	2002	0.64 $\pm$ 0.01 f	0.68 $\pm$ 0.01 ef
Cancela	SL	2002	0.60 $\pm$ 0.00 cde	0.66 $\pm$ 0.00 de
Carreiró	SL	2002	0.51 $\pm$ 0.00 b	0.56 $\pm$ 0.00 b
Demanda	SL	2002	0.58 $\pm$ 0.00 c	0.62 $\pm$ 0.00 c
Ferreirinha	SL	2002	0.59 $\pm$ 0.00 cd	0.63 $\pm$ 0.00 cd
Longal	SL	2002	0.51 $\pm$ 0.01 b	0.56 $\pm$ 0.00 b
Martainha	SL	2002	0.68 $\pm$ 0.01 g	0.72 $\pm$ 0.01 g
Riscada	SL	2002	0.52 $\pm$ 0.01 b	0.56 $\pm$ 0.01 b
Cota	P	2002	0.40 $\pm$ 0.00 a	0.45 $\pm$ 0.00 a
Judia	P	2002	0.59 $\pm$ 0.01 cd	0.65 $\pm$ 0.01 cde
Lada	P	2002	0.38 $\pm$ 0.01 a	0.44 $\pm$ 0.01 a

MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids.

Different letters within one column denote statistically significant differences ($P < 0.05$) by ANOVA and Fisher's LSD test.

Values are average of three individual samples each analysed in triplicate $\pm$ standard deviation.

In addition, a statistical regression study was performed between the parameters of oleic, linoleic and linolenic acid levels. The statistical results for correlation coefficient (R^2)

and significant level (P) are shown in Fig. 3. There is a positive correlation between linoleic and linolenic FAs content ($R^2 = 0.54$, $P < 0.0001$). On other hand, the

Fig. 3. Portuguese chestnuts: relationship between linolenic and linoleic, oleic and linoleic and between oleic and linolenic, all samples, both years.

correlation between oleic and linoleic ($R^2 = 0.74$, $P < 0.0001$) and between oleic and linolenic ($R^2 = 0.67$, $P < 0.0001$) are negative, these correlations are in agreement with previous observations by Garcia et al. (1992, 1994) for pistachio and hazelnut, and by Abdallah et al. (1998) for almond.

The analysis of variance for cv. Longal, the only traditional cv. growing in the PDO Soutos da Lapa and PDO Terra Fria (Table 8) showed significant differences for PDO, year and the interaction PDO $\times$ year for MUFA and PUFA values, and for the ratios of MUFA:PUFA and oleic:linoleic. The SFA values were significantly dependent for location, where Terra Fria had the lowest value (14.67 g/100 g) in 2002 (Table 4) and the highest value for USFA (85.09 g/100 g) (Table 5) in the same year.

The values of MUFA had ranges between 23.08 g/100 g in PDO Soutos da Lapa and 32.96 g/100 g in PDO Terra Fria. As expected, the lowest average value for PUFA was found in samples from PDO Terra Fria (50.76 g/100 g) and the highest in samples from Soutos da Lapa (60.04 g/100 g). The ratio of MUFA to PUFA was higher in Terra Fria than in Soutos da Lapa and the ratio of oleic to linoleic showed the same behavior (Table 7). The crude fat content was significantly dependent on location and the interaction PDO $\times$ year, varying between 1.97 g/100 g in Soutos da Lapa in 2001 and 2.53 g/100 g in PDO Terra Fria in the same year.

For this cultivar, the PDO with consistently better nutritional qualities for chestnuts, like low SFA and high USFA, is PDO Terra Fria. The cv. Longal is not traditionally grown in PDO Padrela. The soil genesis is very different among these two regions: Soutos da Lapa has granite soils whereas Terra Fria has schist soils (Table 1). Also Gomes et al. (1997) detected a relationship between protein content of chestnuts and the type of soil in which they had grown. Chestnuts from places in which schists are predominant showed much higher protein content than those from granite-based soils.

4. Conclusions

The cultivars studied confirm that chestnuts contain low crude fat, low SFA and high unsaturated fatty acid. The major FAs are linoleic, oleic and palmitic and these accounted for more than 85% of the total FA content.

Table 8

Analysis of variance for cv. Longal to SFA, USFA, MUFAs, PUFAs, ratios of MUFA to PUFA and C18:1 to C18:2 and crude fat content

	SFAs	USFA	MUFAs	PUFAs	MUFA/PUFA	C18:1/C18:2	Crude fat (%)
PDO	*	***	***	***	***	***	***
Year	Ns	***	***	***	***	***	Ns
PDO $\times$ year	Ns	Ns	***	***	***	***	**

MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; SFA = saturated fatty acids; USFA = unsaturated fatty acids.

From these results, two cultivars were identified that could be promoted to consumers based on their increased potential health effects: cv. Lada with consistently high linoleic acid content, and cv. Aveleira that has a higher proportion of USFA. With respect to the individual PDO regions, PDO Padrela consistently produced chestnuts with these health-beneficial properties such as the highest content of linoleic acid. The variation detected among cultivars and PDO areas may reflect differences in genotype and is clearly influenced by environmental and growing conditions.

The Longal chestnuts, the only traditional cv. growing on two different PDO areas, had higher nutritional qualities in PDO Terra Fria, which further confirms that the ecological conditions where chestnut orchards are established influence the subsequent qualities of the chestnuts.

Acknowledgement

The authors wish to thank Dr. Richard Bennett (UTAD) for critically reviewing the versions of the manuscript.

References

- Abdallah, A., Ahumada, M.H., Gradziel, T.M., 1998. Oil content and fatty acid composition of almond kernels from different genotypes and California production regions. *Journal of American Society and Horticulture* 123, 1029–1033.
- Aguilera, M.P., Beltrán, G., Ortega, D., Fernández, A., Jiménez, A., Uceda, M., 2005. Characterisation of virgin olive oil of Italian olive cultivars: 'Frantoio' and 'Leccino', grown in Andalusia. *Food Chemistry* 89, 387–391.
- AOAC, 2000. Official Methods of Analysis of AOAC International, 17th Edition. Association of Official Analytical Chemists, USA.
- Benatti, P., Peluso, G., Nicolai, R., Calvani, M., 2004. Polyunsaturated fatty acids: biochemical, nutritional and epigenetic properties. *Journal of the American College of Nutrition* 23 (4), 281–302.
- Desmaison, A., Adrian, J., 1986. La place de la châtaigne en alimentation. *Medicine et Nutrition* 22 (3), 174–180.
- European Union Commission, 1991. Regulation EEC 2568/91 on the characteristics of olive oil and olive pomace and their analytical methods. Official Journal of European Communities L428.
- FAO, 1998. World reference base for soil resources. FAO, Rome.
- Ferreira-Cardoso, J.V., 2002. Caracterização e valorização do fruto de *Castanea sativa* Mill. Composição química, valor nutritivo e efeito da sua utilização na engorda de porcos Duroc. Ph.D. Thesis, UTAD, Portugal.
- Ferreira-Cardoso, J.V., Sequeira, C.A., Rodrigues, L., Gomes, E.F., 1999. Lipid composition of *Castanea sativa* Mill. fruits of some native portuguese cultivars. *Acta Horticulturae* 494, 133–138.
- Garcia, J.M., Agar, I.T., Ve Streif, J., 1992. Fat content and fatty acid composition in individual seeds of pistachio varieties grown in Turkey. *Gartenbauwissenschaft* 57, 130–133.
- Garcia, J.M., Agar, I.T., Ve Streif, J., 1994. Lipid characteristics of kernels from different hazelnut varieties. *Turkish Journal of Agriculture and Forestry* 18, 199–202.
- Gomes, A.L., Abreu, C.G., Castro, L.T., 1997. COLUTAD um clone de castanheiro com resistência à doença da tinta. NATO/SFS Programme III PO-Chestnut. UTAD, Vila Real.
- GPPAA, 2003. Anuário vegetal. Gabinete de planeamento e política agro-alimentar. Ministério da Agricultura, Pescas e Florestas, 304pp.
- Groff, J.L., Gropper, S.S., Hunt, S.M., 1996. Lipids. *Advanced Nutrition and Human Metabolism* 00, 113–146.
- Hardman, E.W., 2002. Omega-3 fatty acids to augment cancer therapy. *Journal of Nutrition* 132, 3508S–3512S.
- Hartman, L., Lago, R.C., 1973. Rapid preparation of fatty acid methyl esters from lipids. *Laboratory Practices* 22 (6), 475–476.
- Kitta, K., Ebihara, M., Iizuka, T., Yoshikawa, R., Isshiki, K., Kawamoto, S., 2005. Variations in lipid content and fatty acid composition of major non-glutinous rice cultivars in Japan. *Journal of Food Composition and Analysis* 18, 269–278.
- Künsch, U., Scharer, H., Conedera, M., Sassella, A., Jermini, M., Jelmini, G., 1999. Quality assessment of chestnut fruits. *Acta Horticulturae* 494, 119–127.
- McCarthy, M.A., Meredith, F.I., 1988. Nutrient data on chestnuts consumed in the United States. *Economy Botany* 42, 29–36.
- Melgarejo, P., Artes, F., 2000. Total lipid content and fatty acid composition of oil seed from lesser known sweet pomegranate clones. *Journal of the Science of Food and Agriculture* 80, 1452–1454.
- Mensink, R.P., Katan, M.B., 1989. Effect of a diet enriched with monounsaturated or polyunsaturated fatty acids on levels of low-density and high-density lipoprotein cholesterol in healthy women and men. *New England Journal of Medicine* 321, 436–441.
- Parcerisa, J., Boatella, J., Codony, R., Farrán, A., Garcia, J., Lopez, A., Rafecas, M., Romero, A., 1993. Influence of variety and geographical origin on the lipid fraction of hazelnuts (*Corylus avellana* L.) from Spain: I. Fatty acid composition. *Food Chemistry* 48, 411–414.
- Parcerisa, J., Rafecas, M., Castellote, A.I., Codony, R., Farrán, A., Garcia, J., Lopez, A., Romero, A., Boatella, J., 1994. Influence of variety and geographical origin on the lipid fraction of hazelnuts (*Corylus avellana* L.) from Spain: II. Triglyceride composition. *Food Chemistry* 50, 245–249.
- Pazianas, M., Butcher, G.P., Subhani, J.M., Finch, P.J., Ang, L., Collins, C., Heaney, R.P., Zaidi, M., Maxwell, J.D., 2005. Calcium absorption and bone mineral density in celiacs after long term treatment with gluten-free diet and adequate calcium intake. *Osteoporosis International* 16, 56–63.
- Senter, S.D., Payne, J.A., Miller, G., Anagnosakis, S.L., 1994. Comparison of total lipids, fatty acids sugars and non-volatile organic acids in nuts from four *Castanea* species. *Journal of the Science of Food and Agriculture* 65, 223–227.
- Üstün, N., Tosun, Y., Serdar, U., 1999. Technological properties of chestnut varieties grown in Erfelek district of Sinop city. *Acta Horticulturae* 494, 107–110.