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İ Ç İ N D E K İ L E R / C O N T E N T S

Determination of Fat Contents and Fatty Acid Compositions of Commercial Chocolates on the Turkish Market*

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The fat content and fatty acid composition of samples of five brands of most commonly consumed Turkish chocolates were determined by gas-liquid chromatography, using a polar 50 m capillary column CP Sil-88 and flame ionization detection. Generally, C16:0, C18:0 and C18:1 were the major fatty acids in all chocolate samples. The fat content varied from 26.10%-41.90%. Total saturated, monounsaturated, polyunsaturated and unsaturated fatty acids were determined in the range of 49.57%-64.04%, 32.21%-44.99%, 2.90%-4.94% and 35.15%-49.93%, respectively. *Trans* fatty acid contents in chocolate samples showed a variation between 0.44% and 0.98%. The low content of *trans* fatty acids showed that cocoa butter had been used in the chocolate manufacturing.

Keywords: chocolate, fat content, fatty acid composition, *trans* fatty acids

*This study is a part of Zeynep Selçuk's master thesis.

Türkiye'de Satışa Sunulan Çikolataların Yağ Oranları ve Yağ Asit Bileşimlerinin İncelenmesi*

Türkiye'de tüketiciler tarafından en çok tercih edilen ilk beş çikolata firmasının örneklerinin yağ oranları ve yağ asit bileşimleri kapiler gaz kromotografi tekniği (CP Sil 88, 50 m x 250 μm i.d.) kullanılarak belirlenmiştir. Genel olarak, tüm çikolata örneklerindeki en önemli yağ asitleri C16:0, C18:0 ve C18:1 olmuştur. Yağ oranı % 26.10 ile % 41.90 arasında değişiklik göstermiştir. Toplam doymuş, toplam tekli-doymamış, toplam çoklu-doymamış ve toplam doymamış yağ asitleri oranı sırası ile % 49,57-64,04, % 32,21-44,99, % 2,90-4.94 ve % 35,15-49,93 arasında belirlenmiştir. Çikolata örneklerindeki *trans* yağ asiti oranlarının % 0,44 ile % 0,98 arasında değiştiği görülmüştür. *Trans* yağ asitlerinin düşük çıkması çikolata üretiminde kakao yağı kullanıldığını ortaya koymuştur.

Anahtar Kelimeler: Çikolata, yağ oranı, yağ asit bileşimi, *trans* yağ asitleri

*Bu çalışma Zeynep Selçuk'un yüksek lisans tezinden türetilmiştir.

Introduction

There is sound scientific evidence that diet and nutrition are important factors in the promotion and maintenance of good health (Gracia et al., 2009). A diet is a lifestyle factor that plays a major role in the primary and secondary prevention of numerous chronic diseases, including certain cancers, cardiovascular disease, and diabetes (Heiss And Kelm, 2010). Indications on some functional cocoa constituents with beneficial effects on human health have been published (Rusconi and Conti, 2010). The consumption of cocoa products and chocolate contributes to human nutrition through the provision of lipids, sugars, minerals (potassium, magnesium, copper and iron) and antioxidants, principally polyphenols (Belscak et al., 2009). Recent studies indicated that the regular intake of chocolate, particularly dark chocolate, has beneficial effects on cardiovascular disease risk by lowering blood pressure, insulin resistance, serum triglycerides, vascular reactivity, endothelial dysfunction, oxidative stress, indicators of inflammation, and antiplatelet activity (Saftlas et al., 2010). Flavanols in cocoa are thought to be responsible for these effects (Buijsse et al., 2010). Tea and red wine have long been known for their high content of polyphenolic substances, but it is less known that cocoa beans, as well as cocoa derived products, also present a rich source of polyphenols, which exhibit an equal or even higher antioxidant capacity than some fruit or vegetables (Belscak et al., 2009). Cocoa has the highest flavanol content of all foods on a per-weight basis and is a significant contributor to the total dietary intake of flavonoids (Visioli et al., 2009), particularly in relation to chocolate with a high content of monomeric (epicatechin and catechin) and oligomeric (procyanidins) flavanols. Therefore, cocoa and chocolate may be considered functional foods, but the functional significance of each component has not been equally clarified (Rusconi and Conti, 2010).

Cocoa and cocoa based products like chocolate are widely consumed in many countries and cultures. For example, in the Dutch population chocolate contributes up to 20% of the total flavonoids intake in adults, and in children the percentage is even higher. In the cultures of some countries such as Switzerland, Austria and Germany, the annual per capita chocolate consumption rate ranged from 8 to 10 kg. In the United States of America, France and the Netherlands, this ratio is between 4 and 5 kg (Rusconi and Conti, 2010). As for the situation in Turkey, chocolate is consumed much at weddings, religious holidays, etc. as a gift rather than through daily consumption. Chocolate consumption in Turkey is very low compared to developed countries; this rate per year ranged from approximately 1.5 to 2 kg. Initially this rate seems very low compared to consumption in developed countries, but the chocolate sector is developing and growing day by day in Turkey. As a matter of fact, of the 6 million tonnes of chocolate consumed in the world in 2009, Turkey's share was 140,000 tonnes. According to data in 2009, while 2% of the chocolate industry grew in developed countries, this rate in Turkey was up to 10-15%, making it the fastest growing country. Whilst there were chocolate exports of $ 155 million in 2005, in the last five years this rate increased from 10% to 63% and was $ 355 million in 2008. In 2010, the chocolate industry will grow 15% in Turkey. The 315,000-320,000 thousand tonnes of exports are estimated to pass the $ 400 million mark (Anon., 2010).

The quality of fats plays a very important role in food processing technology. The quality and dietary character of edible oils has been a topic of concern among food scientists, nutritionists and consumers (Kandhro et al., 2008). The main ingredients of chocolate are cocoa butter, cocoa solids, and sugar, together with milk solids in the case of milk chocolate. During processing, these ingredients are mixed to form a dispersion of cocoa solids (particles) and sugar crystals in a continuous fat phase, consisting of fat crystals and liquid fat (Keijbets et al., 2009). Among these ingredients, cocoa butter has a fairly significant role. Cocoa butter comprises a mixture of saturated and unsaturated fats: C16:0 (palmitic acid), C18:0 (stearic acid), and C18:1 (oleic acid), account for more than 95% of the fatty acids in cocoa butter (Galdomez et al., 2009).

It is well-known that a high fat intake, particularly the consumption of saturated fatty acids (SFA) and *trans* fatty acids (TFA), is associated with a greater risk of various health disorders such as cardiovascular disease, endothelial dysfunction, coronary heart disease, the resistance to insulin, increased levels of blood lipids, inflammation, and the increase in LDL/HDL ratio (Asgary et al., 2009; Beninca et al., 2009; Richter et al., 2009; Mayneris-Perxachs et al., 2010). However, some researchers reported that the health effects of dietary C18:0, a possible replacement for TFAs, particularly in foods that require solid fats (eg, spreads, margarines, baking shortenings, and baked goods). In addition, dietary saturated fatty acids, including C18:0, provide energy, are structural components of cell membranes, and contribute desirable texture and palatability to foods in the diet (Hunter et al., 2010). Similarly, C18:0 is an unusual SFA, in that it does not elevate blood cholesterol levels to the same extent as other fatty acids. Possible explanations for this disparity may include chain length, inefficient absorption, metabolism kinetics, and hepatic desaturation of stearic into oleic acid. As a result, the inclusion of a moderate amount of chocolate containing C18:0 into the diet is not predicted to have adverse effects on the lipid and lipoprotein profile of individuals, as long as the total fat and caloric intake is held constant (Steinberg et al., 2003).

Despite the fact that some previous studies (Daglioglu et al., 2002; Karabulut 2007; Cakmak et al., 2010) have revealed the presence of fatty acid compositions in Turkish chocolate bars, chocolate wafers, and chocolates, until now, according to the Turkish Food Codex Communiqué on Chocolate and Chocolate Products (2003/23) (Anon., 2003) classification and according to consumer prefers, no study has been found on the quality of fatty acid compositions and especially fat contents in chocolates consumed in Turkey. The increasing pressure to decrease the calories intake from fat, to remove *trans* fat from foods, and to supply products with much lower saturated fat contents has become the global concern of many international food organizations such as the Food and Agriculture Organization (FAO), the World Health Organization (WHO) and Food and Drug Administration (FDA), etc. (Kandhro et al., 2008). Therefore, the present study aimed to investigate the amount of fat and the fatty acid composition of the most consumed

bitter, milk, white and nut chocolate brands available in the Turkish market.

Materials and Methods

Sample collection and preparation

Various international and national chocolate companies are present in Turkey. The choices of the brands was based on the top five best-selling brands (A, B and E national; C and D international) and the most preferred by consumers, and was selected for analysis and then were randomly purchased from local markets in Istanbul, Turkey. Chocolates produced by these companies are sold in all regions of Turkey. Four different types of chocolates from these companies were classified, such as bitter, milk, nut and white chocolate, according to the Turkish Food Codex Communiqué on Chocolate and Chocolate Products (Anon., 2003). Each chocolate type was coded with a number (1, 2, 3, 4, 5, etc.). One package of 200 g of each brand was taken between January and April 2010. A composite sample was prepared from three samples of bitter chocolates of the same brand, and the same preparation was also done for the milk, nut and white chocolates. A total of 15 samples were analysed for each chocolate type. The solid chocolates were chopped into small pieces and kept at -18 ºC until the analysis. All reagents, chemicals and solvents used were from E. Merck (Darmstadt, Germany). Both *trans* and *cis* fatty acids methyl esters (FAMEs) standards (99% purity) were purchased from Nu-Check-Prep, Inc (Elysian, MN).

Extraction of the lipids

Total lipid extraction from the chocolate samples was carried out by petroleum ether extraction under the operating conditions specified in the AOAC (1990) method 963.15 and the total fat contents of the samples are expressed as a percentage by mass of the product as received. Extractions and analysis were performed in triplicate for each sample. The fat obtained from the chocolate samples was transferred into 10 mL glass vials. The decanted samples were all frozen at -18 ºC until further analysis.

Preparation of fatty acid methyl esters

FAMEs were prepared according to the American Oil Chemists' Society official method Ce 2-66 (AOCS, 1992). The FAMEs were obtained from the lipid fractions after alkaline hydrolysis, followed by methylation in methanol with a 12.5% boron trifluoride catalyst. The final concentration of the FAMEs was approximately 7 mg/mL in heptane.

Capillary gas-liquid chromatography (GLC) analyses of FAMEs

The analyses of the FAMEs by capillary GLC were carried out on a Hewlett-Packard 6890 chromatograph (Hewlett-Packard, Wilmington, DE) equipped with a Chrompack Autosampler-M911 (Chrompack, Middelburg, the Netherlands) for split-type injection, and a flame-ionization detector. A fused-silica capillary column (Chrompack) was used for the FAME analysis; CP-Sil 88, 50 m x 0.25 mm i.d., 0.20-µm film thickness. The GLC operating conditions were: a temperature program of 130 ºC for 5 min increasing at a rate of 5 ºC/min to 210 ºC; injector and flame-ionization detector temperatures, 250 ºC; carrier gas, helium at a flow rate of 1 mL/min; split ratio 1:100; and volume of injected sample, 1µL. The peaks were identified by comparing the retention times and area percentages with those of authentic standards of FAMEs obtained from Nu-Chek-Prep Inc. and on the basis of literature data (Lipp et al., 2001).

Statistical analysis

The samples were analyzed, one by one, in triplicate. The results were compared with an analysis of variance using the SPSS software program (SPSS Inc., Chicago, IL). Results for each sample were analyzed separately and are presented by individual sample. Differences among the means were compared using Duncan's multiple range tests.

Results and Discussion

Fifteen fatty acids in chocolate lipids were identified and evaluated (Tables 1 and 2). The highest amount of fat in bitter chocolate samples was found to be 41.90% and the lowest 32.57%. In addition, the amounts of fat in the other chocolate samples such as milk, nut and white ranged from 26.10% to 37.94, from 27.35% to 39.01%, and from 29.29% to 38.69%, respectively (Table 1). The means of fat content in the bitter, milk, nut and white chocolate samples were 35.98%, 33.10%, 34.61% and 25.14%, respectively. These levels were comparable to New Zealand chocolates which were reported to have from 28.0% to 36.5% (Saunders et al., 2008).

Table 1. Fat Content and Saturated Fatty Acid Composition of Chocolates Most Common Consumed in Turkey (%)

Samples	Brands	Fat	C8:0	C10:0	C12:0	C14:0	C16:0	C17:0	C18:0	C20:0
Bitter	A	36.25±0.14c	0.02±0.00b	0.06±0.00dc	0.11±0.02d	0.33±0.01d	26.67±0.08b	0.23±0.02c	35.10±0.10b	0.99±0.01b
	B	32.78±0.15d	0.02±0.00b	0.04±0.01d	0.08±0.01e	0.26±0.01e	26.03±0.06d	0.23±0.02c	35.92±0.10a	1.03±0.02a
	C	36.41±0.14b	0.17±0.01a	0.30±0.03b	0.83±0.05a	1.35±0.02b	26.16±0.07c	0.28±0.03b	33.94±0.12c	1.01±0.03a
	D	41.90±0.14a	0.20±0.03a	0.48±0.03a	0.59±0.04b	1.72±0.02a	27.90±0.08a	0.26±0.01b	31.37±0.14d	0.82±0.01d
	E	32.57±0.13e	0.04±0.01b	0.08±0.01c	0.17±0.02c	0.43±0.01c	22.96±0.06e	0.33±0.01a	29.22±0.12e	0.84±0.03c
	p value	**	**	**	**	**	**	**	**	**
	Mean	35.98±0.90	0.09±0.02C	0.19±0.04B	0.36±0.08B	0.82±0.16C	25.94±0.43B	0.27±0.01AB	33.11±0.66A	0.94±0.02A
Milk	A	30.67±0.14d	0.23±0.03ab	1.47±0.04a	0.63±0.02b	2.19±0.03a	26.54±0.09c	0.33±0.02a	31.56±0.11c	0.97±0.01a
	B	33.99±0.13c	0.20±0.02c	0.42±0.02d	0.57±0.01d	1.78±0.02d	26.43±0.08d	0.30±0.03b	33.04±0.10a	0.94±0.01b
	C	37.94±0.12a	0.22±0.02b	0.49±0.03c	0.61±0.02c	2.10±0.03c	26.70±0.08b	0.23±0.02c	31.64±0.10b	0.92±0.02c
	D	26.10±0.10e	0.24±0.03a	0.54±0.04b	0.66±0.03a	2.13±0.03b	26.90±0.07a	0.30±0.01b	30.30±0.13e	0.80±0.01e
	E	36.69±0.11b	0.13±0.02d	0.30±0.03e	0.45±0.03e	1.30±0.02e	22.40±0.09e	0.30±0.01b	31.07±0.14d	0.88±0.01d
	p value	**	**	**	**	**	**	**	**	**
	Mean	33.10±1.14	0.20±0.01B	0.64±0.11A	0.58±0.01B	1.90±0.08B	25.97±0.45B	0.29±0.00A	31.52±0.23A	0.90±0.01A
Nut	A	36.36±0.15c	0.20±0.02a	0.47±0.05a	0.56±0.02c	1.89±0.03a	23.46±0.08b	0.28±0.01a	26.73±0.12d	0.81±0.02b
	B	33.89±0.10d	0.14±0.03c	0.32±0.03c	0.42±0.03d	1.43±0.03d	22.87±0.08c	0.26±0.01b	28.88±0.10a	0.88±0.02a
	C	36.46±0.11b	0.17±0.02b	0.34±0.04b	0.65±0.02b	1.54±0.04c	22.83±0.06d	0.27±0.02ab	27.22±0.14c	0.82±0.02b
	D	27.35±0.10e	0.20±0.03a	0.47±0.05a	0.56±0.02c	1.84±0.04b	24.20±0.07a	0.21±0.01c	27.96±0.11b	0.79±0.01c
	E	39.01±0.13a	0.13±0.02c	0.24±0.02d	0.82±0.04a	1.08±0.03e	20.91±0.09e	0.21±0.01c	25.41±0.12e	0.77±0.01d
	p value	**	**	**	**	**	**	**	**	**
	Mean	34.61±1.06	0.17±0.00B	0.37±0.02B	0.48±0.03B	1.56±0.07B	22.85±0.29C	0.25±0.00B	27.24±0.31B	0.81±0.01B
White	A	35.21±0.10d	0.35±0.02a	0.80±0.04a	1.13±0.03c	3.50±0.03a	41.03±0.08a	0.27±0.02b	15.50±0.13e	0.47±0.01c
	B	36.70±0.12b	0.30±0.02b	0.55±0.05c	1.40±0.04b	2.37±0.02c	25.76±0.08d	0.22±0.02d	30.56±0.15b	0.80±0.02b
	C	35.81±0.14c	0.29±0.03b	0.65±0.05b	0.80±0.02d	2.69±0.02b	28.48±0.06b	0.33±0.03a	29.78±0.11c	0.80±0.02b
	D	29.29±0.11e	0.16±0.03c	0.38±0.06e	0.46±0.02e	1.57±0.02e	27.87±0.07c	0.21±0.03d	31.60±0.15a	0.85±0.02a
	E	38.69±0.12a	0.35±0.02a	0.45±0.05d	3.49±0.03a	2.10±0.03d	23.00±0.07e	0.23±0.03cd	28.83±0.13d	0.85±0.01a
	p value	**	**	**	**	**	**	**	**	**
	Mean	25.14±0.84	0.29±0.01A	0.57±0.03A	1.46±0.28A	2.45±0.17A	29.23±1.65A	0.25±0.01B	27.25±1.58B	0.75±0.03B
p value of the means		*ns*	**	**	**	**	**	**	**	**

[a, b, c, d, e]Each value is an average of three determinations, and value of each sample in the same column with different letters show statistically significant differences. Each value is expressed as weigh (wt)% of total fatty acid methyl esters; **Significant at $P < 0.01$; ns: not significant

Table 1 reveals the average saturated fatty acids (SFAs) composition expressed in the percentage of total fatty acids. The main SFAs in the analysed samples were C16:0 and C18:0, while a small quantity of caprylic (C8:0), capric (C10:0), lauric (C12:0), myristic (C14.0), margaric (C17:0) and arachidic acids (C20:0) were also determined in all chocolate samples. C16:0 and C18:0 were the major SFAs in chocolates. The highest amount of C16:0 was present in A (41.03%) and the lowest in E (23.00%) in white chocolates. The means of C16:0 content in the bitter, milk, nut and white chocolates were 25.94%, 25.97%, 22.85% and 29.23%, respectively ($P < 0.01$). C18:0 was found to amount to 33.11%, 31.52%, 27.24%, and 27.25%, in bitter, milk, nut, and white chocolates, respectively ($P < 0.01$). C8:0, C10:0, C12:0, C17:0 and C20:0 were present in small amounts of less than 1.0%. The mean values of the C14:0 content in the bitter, milk, nut and white chocolate samples were 0.82%, 1.90%, 1.56% and 2.45%, respectively ($P < 0.01$). In our study, SFA content was observed as 61.71% in bitter, 61.84% in milk, 53.85% in nut, and 62.25% in white chocolates ($P < 0.01$). These results show that cocoa butter was used to make the chocolates.

C18:1 was the major monounsaturated fatty acid (MUFA) in all chocolate samples and it was found at a range of 31.76%-40.37% in bitter chocolates. The maximum amount of C18:1 was determined in sample E (44.60%) and the minimum amount in sample D (38.61%) with a mean value of 40.81% in nut chocolates. Furthermore, C18:1 was found to amount to 31.55%-37.89% and 31.56%-35.26% in milk and white chocolates, respectively. The mean values of the palmitoleic (C16:1) and heptadecanoic (C17:1) acids were the other members of MUFA determined at less than 1% in all chocolate samples.

Linoleic acid (C18:2) was the primary polyunsaturated fatty acid (PUFA) in all samples. The mean values of the C18:2 content in the bitter, milk, nut and white chocolate samples were 3.18%, 3.18%, 3.99% and 3.42%, respectively. The C18:2 concentration was equivalently lower in the bitter and milk chocolates than in the white chocolates and significantly highest in the nut chocolates ($P < 0.01$). PUFAs are very important for biological and nutritional values, because essential fatty acids (EFA) are included in this group (Kandhro et al., 2008). Linolenic acid (C18:3) was the other member of PUFAs, determined at less than 1% in all chocolate samples.

Vegetable oils are rich in unsaturated fatty acids in the *cis* configuration. The unsaturated constituents can be isomerized to the *trans* form during oilseed extraction, (Yilmam And Tasan, 2008) during vegetable oil refining (Tasan And Demirci, 2005) and conversion during partial hydrogenation of plant oils (Musavi et al., 2008). When oil is heated during processing, *cis* double bonds occurring naturally in plant oils can change to *trans* double bonds. *Trans* fatty acids (TFA) are formed at temperatures of approximately 190 ºC, which is common during these processes, in addition to their natural generation during the biological hydrogenation of unsaturated fatty acids in the rumen (Wagner *et al*. 2000). Compared with *cis* unsaturated fatty acids, the structure, physical properties, chemical stability and the physiological effects (atherogenic effects) of TFAs resemble those of the SFAs (Kandhro et al., 2008). The amount of TFAs in the milk chocolates ranged from 0.65%-0.93% with a mean value of 0.84% (Table 3). A higher value of TFA was determined in sample E (0.98%) and a lower level in sample B (0.44%) in bitter chocolates. In addition, TFAs were found to amount to 0.50%-0.85% and 0.51%-0.94% in nut and white chocolates, respectively. According to these values, TFA content in chocolates was in Turkey relatively lower than chocolate samples in some countries such as Austria, Poland and New Zealand (Saunders et al., 2008; Wagner et al., 2000; Mojska et al., 2006). In our study, the lowest levels of TFA were observed for chocolates. Similarly, the results in this study are in agreement with the findings of other studies that have reported low levels of TFA values in chocolate samples (Karabulut, 2007; Cakmak et al., 2010; Leth et al., 2006; Fu et al., 2008).

The ratio of unsaturated/saturated and polyunsaturated/saturated fatty acids ranged from 0.549%-1.007%, and from 0.045%-0.099%, respectively (Table 3). A polyunsaturated/saturated fatty acids ratio of 1 is suggested in popular dietary guidelines (Karabulut, 2007). The polyunsaturated/saturated ratio of all analysed chocolate samples was lower than the suggested values.

Chocolates are popular and frequently consumed by the younger population, especially children, and large intakes are common. The presence of the TFAs at low amounts in the chocolate samples

Table 2. Monounsaturated, Polyunsaturated and *Trans* Fatty Acid Composition of Chocolates Most Common Consumed in Turkey (%)

Samples	Brands	C16:1	C17:1	C18:1*c*	C18:1*t*	C18:2*c*	C18:2*t*	C18:3
Bitter	A	0.27±0.02d	0.06±0.01ab	32.03±0.12c	0.78±0.05a	3.00±0.07c	0.16±0.02b	0.19±0.03b
	B	0.27±0.03d	0.02±0.00c	32.50±0.14b	0.26±0.03d	2.97±0.08c	0.18±0.01a	0.19±0.03b
	C	0.40±0.02b	0.05±0.00b	31.76±0.11e	0.57±0.04c	2.81±0.08d	0.17±0.02ab	0.20±0.02b
	D	0.48±0.03a	0.05±0.01b	31.97±0.13d	0.66±0.05b	3.09±0.07b	0.17±0.02ab	0.24±0.03a
	E	0.30±0.01c	0.07±0.02a	40.37±0.15a	0.80±0.05a	4.05±0.07a	0.18±0.01a	0.16±0.01c
	p value	**	**	**	**	**	**	**
	Mean	0.34±0.02C	0.05±0.00B	33.71±0.89B	0.61±0.05A	3.18±0.11C	0.17±0.00	0.20±0.00B
Milk	A	0.82±0.04a	0.08±0.02a	31.55±0.12e	0.74±0.04a	2.64±0.08e	0.19±0.02a	0.26±0.03b
	B	0.48±0.03c	0.06±0.01b	31.95±0.11d	0.49±0.04d	2.97±0.08d	0.16±0.02c	0.21±0.03c
	C	0.53±0.03b	0.05±0.00b	32.24±0.13c	0.75±0.06a	3.09±0.07c	0.18±0.01ab	0.25±0.03b
	D	0.52±0.04b	0.08±0.01a	33.01±0.14b	0.70±0.06b	3.37±0.07b	0.17±0.03bc	0.28±0.02a
	E	0.33±0.04d	0.08±0.02a	37.89±0.12a	0.68±0.05c	3.82±0.09a	0.16±0.02c	0.21±0.02c
	p value	**	**	**	**	**	*	**
	Mean	0.54±0.04A	0.07±0.00A	33.33±0.62B	0.67±0.02A	3.18±0.10C	0.17±0.00	0.24±0.00A
Nut	A	0.55±0.03a	0.07±0.02a	40.12±0.14c	0.66±0.05a	3.77±0.08d	0.19±0.01a	0.24±0.02a
	B	0.42±0.04c	0.06±0.01ab	39.65±0.12d	0.48±0.03c	3.84±0.08c	0.16±0.02b	0.19±0.02b
	C	0.43±0.04c	0.06±0.01ab	41.08±0.15b	0.34±0.03d	3.88±0.07b	0.19±0.01a	0.18±0.02b
	D	0.46±0.03b	0.05±0.00b	38.61±0.13e	0.52±0.05b	3.70±0.07e	0.19±0.01a	0.24±0.03a
	E	0.33±0.03d	0.06±0.00ab	44.60±0.14a	0.32±0.04e	4.76±0.07a	0.18±0.02a	0.18±0.01b
	p value	**	*	**	**	**	*	**
	Mean	0.44±0.01B	0.06±0.00AB	40.81±0.54A	0.46±0.03B	3.99±0.10A	0.18±0.00	0.21±0.00B
White	A	0.27±0.03e	0.06±0.01ab	32.03±0.11d	0.78±0.05a	3.00±0.09d	0.16±0.03b	0.19±0.02ab
	B	0.42±0.03c	0.05±0.00b	33.06±0.10b	0.48±0.03d	3.68±0.08b	0.15±0.02c	0.20±0.02c
	C	0.62±0.04a	0.07±0.03a	31.56±0.12e	0.40±0.03c	3.15±0.07c	0.16±0.01b	0.22±0.02b
	D	0.46±0.04b	0.07±0.03a	32.42±0.12c	0.57±0.04b	2.95±0.06e	0.19±0.02a	0.24±0.03a
	E	0.37±0.03d	0.05±0.02b	35.26±0.13a	0.33±0.03e	4.33±0.06a	0.18±0.02a	0.18±0.03d
	p value	**	*	**	**	**	**	**
	Mean	0.43±0.02AB	0.06±0.00AB	32.87±0.35B	0.51±0.02B	3.42±0.12B	0.17±0.00	0.21±0.00B
p value of the means		**	**	**	**	**	*ns*	**

[a, b, c, d, e] Each value is an average of three determinations, and value of each sample in the same column with different letters show statistically significant differences. Each value is expressed as weight (wt)% of total fatty acid methyl esters. *Significant at $P < 0.05$; **Significant at $P < 0.01$; ns: not significant

Table 3. Groups from Fatty Acid Composition for Chocolates Most Common Consumed in Turkey (%)

Samples	Brands	SFA	TFA	MUFA	PUFA	UFA	PUFA/SFA	UFA/SFA
Bitter	A	63.51±0.20c	0.94±0.05b	32.36±0.12d	3.19±0.06c	35.55±0.13d	0.050	0.559
	B	63.61±0.16b	0.44±0.04e	32.79±0.12b	3.16±0.09d	35.95±0.14b	0.049	0.565
	C	64.04±0.18a	0.74±0.05d	32.21±0.14e	3.01±0.08e	35.22±0.14e	0.047	0.549
	D	63.34±0.20d	0.83±0.05c	32.50±0.14c	3.33±0.08b	35.83±0.12c	0.052	0.565
	E	54.07±0.16e	0.98±0.04a	40.74±0.15a	4.21±0.06a	44.95±0.15a	0.077	0.831
	p value	**	**	**	**	**		
	Mean	61.71±1.02A	0.79±0.05A	34.12±0.93A	3.38±0.11C	37.50±0.99B	0.054	0.607
Milk	A	63.92±0.18a	0.93±0.05a	32.25±0.12e	2.90±0.06e	35.15±0.11e	0.045	0.549
	B	63.68±0.18b	0.65±0.05d	32.49±0.11d	3.18±0.07d	35.67±0.11d	0.049	0.560
	C	62.91±0.16c	0.93±0.05a	32.82±0.11c	3.34±0.07c	36.16±0.12c	0.053	0.574
	D	61.87±0.20d	0.87±0.04b	33.61±0.12b	3.65±0.08b	37.26±0.13b	0.058	0.602
	E	56.83±0.18e	0.84±0.03c	38.30±0.13a	4.03±0.09a	42.33±0.14a	0.070	0.744
	p value	**	**	**	**	**		
	Mean	61.84±0.69A	0.84±0.02A	35.89±0.60B	3.42±0.10C	37.31±0.68B	0.055	0.603
Nut	A	54.40±0.16c	0.85±0.04a	40.74±0.15c	4.01±0.08d	44.75±0.14c	0.073	0.822
	B	55.20±0.18b	0.64±0.03c	40.13±0.15d	4.03±0.08c	44.16±0.15d	0.073	0.800
	C	53.84±0.18d	0.53±0.03d	41.57±0.14b	4.06±0.09b	45.63±0.15b	0.075	0.847
	D	56.23±0.20a	0.71±0.04b	39.12±0.14e	3.94±0.07e	43.06±0.14e	0.070	0.765
	E	49.57±0.19e	0.50±0.04e	44.99±0.15a	4.94±0.08a	49.93±0.16a	0.099	1.007
	p value	**	**	**	**	**		
	Mean	53.85±0.61B	0.65±0.03B	41.31±0.53A	4.20±0.10A	45.15±0.63A	0.077	0.838
White	A	63.05±0.19c	0.94±0.05a	32.36±0.12d	3.19±0.08d	35.55±0.10e	0.050	0.563
	B	61.96±0.18d	0.63±0.04c	33.53±0.11b	3.88±0.07b	37.41±0.11b	0.062	0.603
	C	63.82±0.16a	0.56±0.04d	32.25±0.12e	3.37±0.08c	35.62±0.11d	0.052	0.558
	D	63.10±0.16b	0.76±0.06b	32.95±0.11c	3.19±0.07d	36.14±0.12c	0.050	0.572
	E	59.30±0.19e	0.51±0.05e	35.68±0.11a	4.51±0.08a	40.19±0.13a	0.076	0.677
	p value	**	**	**	**	**		
	Mean	62.25±0.42A	0.68±0.02B	33.35±0.32B	3.63±0.12B	36.98±0.43B	0.058	0.594
p value of the means		**	**	**	**	**		

[a, b, c, d, e]Each value is an average of three determinations, and value of each sample in the same column with different letters show statistically significant differences. Each value is expressed as weight (wt)% of total fatty acid methyl esters. **Significant at $P < 0.01$

SFA = Saturated fatty acids; TFA = *Trans* fatty acids; MUFA = Monounsaturated fatty acids; PUFA = Polyunsaturated fatty acids; UFA = Unsaturated fatty acids

show that hydrogenated fats or hydrogenated oils were not used; cocoa butter was used in the manufacturing process. Therefore, the choice of the type of fat is important with the zero *trans* or low *trans* content in the production of foods.

Conclusions

In conclusion, we have presented the fatty acid composition and fat content of the most commonly consumed Turkish chocolates. Our findings indicated low contents of TFAs in chocolate samples.

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