Trans Fatty Acid Contents in Chocolates and Chocolate Wafers in Turkey

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Abstract

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Fatty acid compositions and *trans* fatty acid contents of chocolate and chocolate wafers collected from Turkish markets were determined by GC. Total 62 samples, being all chocolates and chocolate wafers sold in Turkey, were investigated. 35 samples of chocolate which were categorised as milk chocolate, bitter chocolate, chocolate with nuts, chocolate with pistachio, chocolate with almond, other chocolates, and 27 samples of chocolate wafer which were categorised as pure chocolate wafers and chocolate wafers with nuts belonging to 12 different national chocolate brands, were analysed. Generally, C16:0 palmitic acid, C18:0 stearic acid, and C18:1 oleic acid were the major fatty acids in all samples. *Trans* fatty acid contents in chocolate wafers samples showed a variation between 0.00–7.92%. *Trans* fatty acids were determined as 0.00–6.23% in chocolate samples. In conclusion, it was shown that *trans* fatty acids contents in chocolates were relatively lower than those of other countries.

Keywords: *trans* fatty acid; fatty acid composition; chocolate; chocolate wafer; Turkey

Trans-isomeric fatty acids occur naturally in dairy and other animal fats by biological hydrogenation in the stomach of ruminants, but they originate mainly from the industrial process of catalytic hydrogenation of fats; 80–90% of dietary *trans* fatty acids are derived from this latter source, whereas 2–8% are provided by dairy products (Kromer 1976; Mounts 1979). Some fatty acids have one or more double bonds in the *trans* configuration: these are the so-called *trans* fatty acids (TFAs) (Fritsche & Steinhart 1998). TFAs are present in variable amounts in a wide range of foods, including most foods made with partially hydrogenated oils such as baked goods and fried foods, and some margarine products (Semma 2002). TFAs content varies considerably among foods, reflecting, the differences in the fats and oils used in the manufacturing or preparation processes (Innis *et al*. 1999).

TFAs increase plasma concentrations of low-density lipoprotein cholesterol and reduce the concentrations of high-density lipoprotein cholesterol (Ascherio &Willet 1997). TFAs have adverse effects on blood lipoproteins, and they are therefore expected to raise the risk of coronary heart disease (Katan 1998). As a result, the net effect of TFAs on the ratio of LDL to HDL cholesterol is approximately double that of the saturated fatty acids. These adverse effects of TFAs have been confirmed by other studies (Zock & Katan

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1992; Judd *et al.* 1994). In addition, there are concerns about recent suggestions that TFA may affect human fetal growth and infant development (Ayagari *et al.* 1996). Also, an association exists between the prevalence of asthma and allergies in children and the risk of diabetes in adults with the intake of *trans* isomers (Weiland *et al.* 1999; Stender & Dyerberg 2004).

Some research has been carried out into TFAs in various foods worldwide (Demmelmair *et al.* 1996; Wagner *et al.* 2000; Mojska *et al.* 2006) and in Turkey (Demirbas & Yilmaz 2000; Cetin *et al.* 2003; Daglioglu & Tasan 2003; Karabulut 2007; Basol & Tasan 2008). The most recent study concerning *trans* fatty acids in Turkey reports fatty acid compositions of Turkish shortenings (Basol & Tasan 2008). The authors stated that partially hydrogenated vegetable oils with a high TFA content were still the major raw materials used in the production of Turkish shortenings. However, there is limited research (Karabulut 2007) into fatty acids composition and *trans* fatty acids contents in chocolates. Therefore, the objective of this study is to determine the fatty acids compositions and *trans* fatty acids contents in all chocolate and chocolate wafers brands sold in Turkish markets.

MATERIALS AND METHODS

Sample collection. 35 samples of chocolates and 27 samples of chocolate wafers belonging to 12 different national chocolate brands were analysed in 2007. The chocolates were categorised as milk chocolates (n = 13), bitter chocolates (n = 4), chocolates with nuts (n = 8), chocolates with pistachio (n = 3), chocolates with almond (n = 2), other chocolates (n = 5), and chocolate wafers were categorised as pure chocolate wafers (n = 14) and chocolate wafers with nuts (n = 13). Total 62 samples were investigated, being all chocolate and chocolate wafers brands sold in Turkey.

Fatty acid analysis. The samples of chocolates and chocolate wafers were extracted using the Folch *et al.* (1957) method. The fatty acids in the total lipid were esterified into methyl esters by saponification with 0.5N methanolic NaOH and transesterified with 14% BF_3 (v/v) in methanol (Paquot 1979).

Fatty acid methyl esters (FAMEs) were analysed on a HP (Hewlett Packard, Palo Alto, USA) Agilent 6890N model gas chromatograph (GC), equipped with a flame ionisation detector (FID) and fitted with a HP-88 capillary column (100 m, 0.25 mm i.d. and 0.2 µm). The injector and detector temperatures were 240°C and 250°C, respectively. The oven was programmed at 160°C initial temperature and 2 min initial time. Thereafter, the temperature increased by 4°C/min to 185°C, then it increased by 1°C/min to 200°C and was held for 46.75 min at 200°C. Total run time was 70 minutes. Carrier gas was helium (1 ml/min).

The identification of fatty acids was carried out by comparing the sample FAME peak relative retention times with those obtained for Alltech (Carolean Industrial Drive, Satate Collage, USA) standards. The results were expressed as FID response area relative percentages. Each reported result is the average value of three GC analyses. The results are presented as means ± SD.

RESULTS

Fatty acids compositions and trans fatty acids contents in 35 chocolates (milk chocolates, bitter chocolates, chocolates with nuts, chocolates with pistachio, chocolates with almond, and other chocolates) and 27 chocolate wafers (pure chocolate wafers, and chocolate wafers with nuts) belonging to 12 different national brands obtained from markets in Turkey are presented in Tables 1 and 2. The TFA content varied depending on the chocolate and chocolate wafers groups (Table 3).

Twenty five fatty acids in chocolates and chocolate wafers lipids were identified and evaluated. Palmitic acid, stearic acid, and oleic acid were high in chocolates and chocolate wafers. Oleic acid was the major fatty acids in all samples except for bitter chocolate (Tables 1 and 2).

Palmitic and stearic acids were the major saturated fatty acids (SFA) in chocolates and chocolate wafers. Palmitic and stearic acids ranged between 20.39–25.80% and 25.77–34.66% in chocolate groups, and 24.89–26.52% and 17.86–21.10% in chocolate wafers groups, respectively (Tables 1 and 2). High amounts of lauric acid were determined in pure chocolate wafers (5.11%) and chocolate wafers with nuts (4.08%).

Oleic acid was the major monounsaturated fatty acid (MUFA) in all samples. Oleic acid was found to amount to 43.33%, 38.49%, 36.88%, 36.39%, 33.80%, 33.16%, 32.99%, and 32.31%, in chocolates

Table 1. Fatty acid composition and *trans* fatty acid content of analysed chocolates (mean ± SD)

Fatty acids	Milk chocolate (n = 13)	Bitter chocolate (n = 4)	Chocolate with nuts (n = 8)	Chocolate with pistachio (n = 3)	Chocolate with almond (n = 2)	Other chocolate (n = 5)	All chocolates (n = 35)
C 8:0	0.13 ± 0.19	0.02 ± 0.01	0.01 ± 0.04		–	–	0.026 ± 0.13
C 10:0	0.50 ± 0.50	0.17 ± 0.09	0.30 ± 0.22	0.64 ± 0.08	0.93 ± 0.02	0.13 ± 0.12	0.447 ± 0.45
C 11:0	0.06 ± 0.08	0.01 ± 0.01	0.05 ± 0.04	0.05 ± 0.05	0.14 ± 0.01	0.01 ± 0.02	0.056 ± 0.07
C 12:0	0.56 ± 0.29	0.37 ± 0.06	0.36 ± 0.17	0.36 ± 0.14	1.10 ± 0.03	1.63 ± 1.32	0.729 ± 0.66
C 13:0	0.02 ± 0.02	–	0.02 ± 0.01	0.02 ± 0.02	0.03 ± 0.01	–	0.015 ± 0.02
C 14:0	1.71 ± 0.34	0.48 ± 0.06	1.24 ± 0.32	1.17 ± 0.20	1.66 ± 0.04	1.67 ± 0.53	1.320 ± 0.52
C 15:0	0.13 ± 0.10	0.06 ± 0.01	0.02 ± 0.03	0.04 ± 0.01	0.01 ± 0.01	0.10 ± 0.06	0.059 ± 0.08
C 16:0	25.70 ± 1.78	25.11 ± 0.17	20.39 ± 2.20	22.91 ± 0.92	23.65 ± 0.21	25.80 ± 2.73	23.925 ± 3.04
C 17:0	0.29 ± 0.04	0.22 ± 0.01	0.21 ± 0.04	0.22 ± 0.03	0.23 ± 0.01	0.24 ± 0.07	0.235 ± 0.05
C 18:0	31.10 ± 2.24	34.66 ± 0.34	25.11 ± 4.89	25.77 ± 6.89	25.95 ± 1.63	26.73 ± 1.58	28.220 ± 5.00
C 20:0	0.03 ± 0.08	–	0.06 ± 0.11	–	–	0.21 ± 0.26	0.048 ± 0.12
Σ SFA	60.23 ± 3.01	61.10 ± 0.27	47.77 ± 6.50	51.18 ± 7.37	53.70 ± 1.75	56.52 ± 3.70	55.080 ± 7.12
C 14:1 n5	0.21 ± 0.05	0.04 ± 0.01	0.15 ± 0.05	0.14 ± 0.01	0.18 ± 0.01	0.10 ± 0.06	0.135 ± 0.07
C 15:1 n5	0.06 ± 0.02	0.01 ± 0.01	0.05 ± 0.03	0.06 ± 0.01	0.05 ± 0.01	0.01 ± 0.01	0.040 ± 0.03
C 16:1 n7	0.62 ± 0.09	0.30 ± 0.04	0.42 ± 0.15	0.51 ± 0.09	0.49 ± 0.01	0.39 ± 0.09	0.455 ± 0.15
C 17:1 n8	0.08 ± 0.02	0.03 ± 0.01	0.08 ± 0.01	0.07 ± 0.01	0.07 ± 0.01	0.05 ± 0.01	0.062 ± 0.02
C 18:1 n9	33.16 ± 2.51	33.80 ± 0.60	43.33 ± 5.61	38.49 ± 5.46	36.39 ± 1.28	32.31 ± 2.82	36.246 ± 5.74
C 20:1 n9	0.17 ± 0.05	0.04 ± 0.03	0.08 ± 0.09	0.24 ± 0.05	0.13 ± 0.01	0.08 ± 0.04	0.124 ± 0.08
Σ MUFA	34.30 ± 2.44	34.22 ± 0.52	44.11 ± 5.49	39.51 ± 5.61	37.31 ± 1.32	32.94 ± 2.92	37.062 ± 5.70
C 18:2 n6	3.89 ± 0.87	3.33 ± 0.21	5.82 ± 1.44	7.45 ± 2.04	6.95 ± 0.48	9.00 ± 4.76	6.074 ± 2.68
C 18:3 n6	0.69 ± 0.33	0.76 ± 0.29	0.72 ± 0.22	0.76 ± 0.35	0.77 ± 0.08	0.12 ± 0.25	0.638 ± 0.34
C 18:3 n3	0.45 ± 0.29	0.43 ± 0.21	0.21 ± 0.05	0.43 ± 0.02	0.21 ± 0.01	0.92 ± 0.40	0.445 ± 0.32
C 20:4 n6	0.02 ± 0.06	–	–	0.06 ± 0.01	–	0.13 ± 0.11	0.034 ± 0.07
Σ PUFA	5.05 ± 0.80	4.52 ± 0.29	6.75 ± 1.28	8.70 ± 1.71	7.93 ± 0.40	10.17 ± 4.87	7.191 ± 2.64
C 16:1 *t*9	0.03 ± 0.04	–	0.06 ± 0.03	0.07 ± 0.01	0.05 ± 0.01	0.02 ± 0.02	0.037 ± 0.04
C 18:1 *t*9	0.11 ± 0.17	–	0.72 ± 1.65	0.22 ± 0.01	0.61 ± 0.01	0.06 ± 0.13	0.287 ± 0.85
C 18:2 *t*9*t*12	0.04 ± 0.03	–	0.19 ± 0.42	0.06 ± 0.01	0.06 ± 0.01	0.05 ± 0.04	0.066 ± 0.21
C 18:2 *t*9*c*12	–	–	0.05 ± 0.06	0.02 ± 0.01	0.04 ± 0.01	–	0.017 ± 0.04
Σ TFA	0.18 ± 0.20	–	1.02 ± 2.12	0.37 ± 0.01	0.76 ± 0.01	0.13 ± 0.12	0.407 ± 1.09
Other	0.24 ± 0.14	0.16 ± 0.04	0.35 ± 0.17	0.24 ± 0.05	0.30 ± 0.03	0.24 ± 0.18	0.260 ± 0.17

SFA – saturated fatty acid; MUFA – monounsaturated fatty acid; PUFA – polyunsaturated fatty acid; TFA – *trans* fatty acid; Other – unknown peak

Table 2. Fatty acid composition and *trans* fatty acid content of analysed chocolate wafers (mean ± SD)

Fatty acids	Pure chocolate wafers (n = 14)	Nut chocolate wafers (n = 13)	All chocolate wafers (n = 27)
C 8:0	0.20 ± 0.33	0.21 ± 0.51	0.201 ± 0.37
C 10:0	0.51 ± 0.53	0.42 ± 0.68	0.466 ± 0.57
C 11:0	0.03 ± 0.03	0.01 ± 0.01	0.018 ± 0.02
C 12:0	5.11 ± 9.90	4.08 ± 8.53	4.595 ± 8.77
C 13:0	0.02 ± 0.02	0.01 ± 0.01	0.015 ± 0.02
C 14:0	2.97 ± 3.86	2.14 ± 2.70	2.553 ± 3.26
C 15:0	0.06 ± 0.08	0.06 ± 0.04	0.060 ± 0.07
C 16:0	26.52 ± 5.24	24.89 ± 5.42	25.705 ± 5.16
C 17:0	0.18 ± 0.06	0.15 ± 0.04	0.167 ± 0.06
C 18:0	21.10 ± 5.65	17.86 ± 4.82	19.479 ± 5.55
C 20:0	0.04 ± 0.08	0.10 ± 0.15	0.068 ± 0.12
Σ SFA	56.74 ± 9.85	49.93 ± 13.32	53.327 ± 11.50
C 14:1 n5	0.12 ± 0.08	0.06 ± 0.03	0.094 ± 0.07
C 15:1 n5	0.04 ± 0.03	0.02 ± 0.03	0.029 ± 0.04
C 16:1 n7	0.37 ± 0.17	0.26 ± 0.12	0.316 ± 0.16
C 17:1 n8	0.05 ± 0.03	0.04 ± 0.02	0.045 ± 0.02
C 18:1 n9	32.99 ± 8.51	36.88 ± 10.85	34.932 ± 9.63
C 20:1 n9	0.11 ± 0.06	0.13 ± 0.15	0.121 ± 0.12
Σ MUFA	33.68 ± 8.61	37.39 ± 11.00	35.537 ± 9.74
C 18:2 n6	7.69 ± 2.31	10.28 ± 4.78	8.989 ± 3.68
C 18:3 n6	0.55 ± 0.22	0.29 ± 0.27	0.417 ± 0.29
C 18:3 n3	0.37 ± 0.07	0.55 ± 0.38	0.463 ± 0.30
C 20:4 n6	0.01 ± 0.01	0.02 ± 0.03	0.011 ± 0.03
Σ PUFA	8.62 ± 2.29	11.14 ± 4.97	9.880 ± 3.76
C 16:1 *t*9	0.03 ± 0.03	0.03 ± 0.07	0.027 ± 0.06
C 18:1 *t*9	0.56 ± 0.87	1.19 ± 2.06	0.874 ± 1.55
C 18:2 *t*9*t*12	0.03 ± 0.03	0.13 ± 0.26	0.080 ± 0.19
C 18:2 *t*9*c*12	0.03 ± 0.04	0.04 ± 0.10	0.038 ± 0.07
Σ TFA	0.65 ± 0.90	1.39 ± 2.38	1.019 ± 1.77
Other	0.32 ± 0.16	0.16 ± 0.15	0.237 ± 0.17

SFA – saturated fatty acid; MUFA – monounsaturated fatty acid; PUFA – polyunsaturated fatty acid; TFA – *trans* fatty acid; Other – unknown peak

with nuts, chocolates with pistachio, chocolate wafers with nuts, chocolates with almond, bitter chocolates, milk chocolates, pure chocolate wafers, and other chocolates groups, respectively. The percentage of oleic acid was found to be 36.246% and 34.932% in chocolates and chocolate wafers groups, respectively (Tables 1 and 2).

Linoleic acid was the primary polyunsaturated fatty acid (PUFA) in all samples. This fatty acid was found to represent 10.28%, 9.00%, 7.69%, 7.45%, 6.95%, 5.82%, 3.89%, and 3.33%, in chocolate wafers with nuts, other chocolates, pure chocolate wafers, chocolates with pistachio, chocolates with almond, chocolates with nuts, milk chocolates,

Table 3. Minimum and maximum range of fatty acid composition and *trans* fatty acid content of chocolate and chocolate wafers

Fatty acids	Milk chocolate (n = 13)	Bitter chocolate (n = 4)	Chocolate with nuts (n = 8)	Chocolate with pistachio (n = 3)	Chocolate with almond (n = 2)	Other chocolate (n = 5)	Pure chocolate wafers (n = 14)	Chocolate wafers with nuts (n = 13)
C 8:0	0.00–0.62	0.00–0.07	0.00–0.10	–	–	–	0.00–1.17	0.00–1.84
C 10:0	0.10–2.10	0.03–0.33	0.04–0.74	0.00–1.04	0.96–0.99	0.00–0.31	0.00–2.07	0.00–2.35
C 11:0	0.00–0.30	0.00–0.06	0.00–0.11	0.00–0.12	0.08–0.09	0.00–0.06	0.00–0.09	0.00–0.03
C 12:0	0.31–1.42	0.11–1.01	0.17–0.60	0.18–0.45	0.32–0.36	0.40–3.66	0.33–38.76	0.22–31.90
C 13:0	0.00–0.06	0.00–0.01	0.00–0.03	0.00–0.04	0.03	0.00–0.01	0.00–0.05	0.00–0.05
C 14:0	0.94–2.15	0.22–1.07	0.68–1.75	1.01–1.44	1.25–1.31	0.98–2.27	0.91–15.95	0.63–10.90
C 15:0	0.00–0.22	0.04–0.09	0.00–0.09	0.01–0.11	0.01	0.00–0.17	0.00–0.21	0.00–0.11
C 16:0	21.87–29.98	24.37–25.53	16.97–24.87	20.79–25.32	21.41–21.71	21.99–28.35	14.10–35.02	15.38–36.28
C 17:0	0.22–0.35	0.22–0.23	0.16–0.25	0.16–0.28	0.25–0.26	0.16–0.34	0.04–0.29	0.05–0.24
C 18:0	26.28–33.49	30.51–36.51	17.27–32.81	16.97–37.06	27.88–30.18	24.51–29.38	14.56–29.41	12.95–28.18
C 20:0	0.00–0.30	–	0.00–0.25	–	–	0.00–0.54	0.00–0.28	0.00–0.42
Σ SFA	51.32–62.81	57.75–62.54	38.46–57.96	43.99–62.14	52.33–54.78	50.04–59.73	45.09–83.68	34.75–87.18
C 14:1 n5	0.11–0.27	0.01–0.09	0.08–0.22	0.10–0.17	0.16–0.17	0.02–0.18	0.01–0.27	0.01–0.13
C 15:1 n5	0.04–0.11	0.00–0.04	0.01–0.12	0.05–0.09	0.05	0.00–0.03	0.00–0.10	0.00–0.12
C 16:1 n7	0.43–0.74	0.22–0.39	0.12–0.62	0.44–0.63	0.55–0.57	0.29–0.48	0.04–0.68	0.00–0.45
C 17:1 n8	0.04–0.13	0.00–0.04	0.06–0.10	0.05–0.09	0.08–0.09	0.03–0.06	0.00–0.09	0.00–0.06
C 18:1 n9	31.29–40.03	32.27–37.02	34.21–53.34	31.58–42.49	37.04–38.85	27.61–34.48	8.40–44.01	7.57–52.50
C 20:1 n9	0.09–0.25	0.00–0.12	0.00–0.19	0.20–0.30	0.25–0.27	0.02–0.13	0.00–0.20	0.00–0.61
Σ MUFA	32.16–41.10	32.70–37.70	35.25–53.87	32.50–43.74	38.12–40.00	28.06–35.18	8.45–44.58	7.62–53.01
C 18:2 n6	3.06–6.03	2.95–3.67	3.96–8.75	2.99–12.31	5.03–5.71	4.17–14.05	3.48–12.34	4.03–18.96
C 18:3 n6	0.01–0.99	0.34–0.96	0.45–1.17	0.31–1.34	0.87–0.99	0.00–0.56	0.01–0.86	0.02–0.81
C 18:3 n3	0.24–1.19	0.30–0.77	0.14–0.29	0.22–0.82	0.24–0.25	0.24–1.25	0.21–1.18	0.16–1.53
C 20:4 n6	0.00–0.20	–	–	0.00–0.17	–	0.00–0.23	0.00–0.09	0.00–0.12
Σ PUFA	4.25–7.04	4.09–4.98	5.35–9.51	4.55–13.60	6.26–6.83	4.96–15.24	4.34–13.61	4.32–20.52
C 16:1 *t*9	0.00–0.10	–	0.00–0.09	0.00–0.11	0.10	0.00–0.06	0.00–0.07	0.00–0.27
C 18:1 *t*9	0.00–0.40	–	0.00–4.77	0.00–0.34	0.33	0.00–0.29	0.00–2.66	0.00–6.64
C 18:2 *t*9*t*12	0.00–0.09	0.00–0.01	0.00–1.23	0.02–0.13	0.02	0.00–0.10	0.00–0.09	0.00–0.94
C 18:2 *t*9*c*12	0.00–004	–	0.00–0.19	0.00–0.03	0.02–0.03	–	0.00–0.10	0.00–0.33
Σ TFA	0.00–0.53	0.00–0.01	0.00–6.23	0.13–0.48	0.48	0.00–0.35	0.00–2.90	0.00–7.92
Other	0.06–0.47	0.00–0.48	0.05–0.57	0.00–0.38	0.35–0.36	0.00–0.47	0.09–0.66	0.01–0.51

SFA – saturated fatty acid; MUFA – monounsaturated fatty acid; PUFA – polyunsaturated fatty acid; TFA – *trans* fatty acid; Other – unknown peak and bitter chocolates groups, respectively. The percentage of linoleic acid found was 6.074% and 8.989% in chocolate and chocolate wafers groups, respectively (Tables 1 and 2).

SFAs contents were found to be higher than those of MUFAs and PUFAs in all samples and were determined to be 61.10%, 60.23%, 56.74%, 56.52%, 53.70%, 51.18%, 49.93%, and 47.77%, in bitter chocolates, milk chocolates, pure chocolate wafers, other chocolates, chocolates with almond, chocolates with pistachio, chocolate wafers with nuts, and chocolates with nuts, respectively. The percentage of SFA was found to be 55.08%, and 53.327% in chocolate and chocolate wafers groups, respectively (Tables 1 and 2).

In our study, the percentages of C 16:1 *t*9, C 18:1 *t*9, C 18:2 *t*9*t*12, and C 18:2 *t*9*c*12 ranged from 0.00% to 0.07%, 0.00% to 0.72%, 0.00% to 0.19%, and 0.00% to 0.05%, in chocolates groups, respectively (Table 1). The percentages of C 16:1 *t*9, C 18:1 *t*9, C 18:2 *t*9*t*12, and C 18:2 *t*9*c*12 ranged from 0.03% to 0.03%, 0.56% to 1.19%, 0.03% to 0.13%, and 0.03% to 0.04%, in chocolate wafers groups, respectively (Table 2). Total *trans* fatty acids contents were found to be higher in chocolate wafers with nuts than in other samples (1.39%). C 18:1 *t*9 elaidic acid, was found to be the most abundant *trans* fatty acid in all samples. TFAs were found in all samples of milk chocolates, pure chocolate wafers, other chocolates, chocolates with pistachio, chocolates with almond, chocolate wafers with nuts and chocolates with nuts but none of the TFAs was determined in bitter chocolates. Minimum and maximum ranges of TFAs were determined as 0.00–6.23% in the chocolate samples and 0.00–7.92% in the wafer chocolate samples (Table 3). The percentages of TFAs were determined as 0.407% and 1.019% in chocolates and chocolate wafers groups, respectively (Tables 1 and 2).

DISCUSSION

Tarkowski and Kowalczyk (2007) investigated the fatty acids contents in milk chocolates marketed in Poland. They analysed fourteen chocolate samples. It was observed that palmitic, stearic, oleic, and linoleic acids were the predominant fatty acids. Similarly, in our study, oleic, stearic, palmitic, and linoleic acids were the major fatty acids with proportions of 36.246%, 28.22%, 23.925%, and 6.074%, in chocolate groups, and 34.932%, 19.479%, 25.705%, and 8.989% in chocolate wafers groups, respectively. Tarkowski and Kowalczyk (2007) also observed that the chocolates supplemented with nuts had higher oleic and linoleic acids contents. Similarly, in our study, oleic acid was also found to be contained in the highest proportion in chocolates with nuts (43.33%).

Demmelmair *et al*. (1996) investigated TFAs contents in 42 different brands of spreads and cold meats including chocolate spreads consumed by German children (4–7 years old). They determined *trans* fatty acid content in chocolate spreads in the range of 0.7–11.1%. In our study, TFAs content was relatively lower than in their findings. We observed that TFAs content was 0.0–6.23% in chocolates and 0.00–7.92% in chocolate wafers.

Wagner *et al*. (2000) investigated the contents of TFAs in margarines, plant oils, fried products, and chocolate spreads in Austria. They observed that the levels of TFAs were 0.6–8.9% (mean 4.9%) in chocolate spreads. They suggested the use of partially hydrogenated fats in the manufacture of these spreads. In our study, we determined that TFAs contents were 0.00–6.23% (mean 0.407%) in chocolates and 0.00–7.92% (mean 1.019%) in chocolate wafers. Thus, TFAs content in chocolates was in Turkey relatively lower than in Austria. Wagner *et al*. (2000) observed that SFA values for these spreads ranged between 11.9% and 29.4%, with a mean value of 22.4%. Their observed MUFA values ranged between 29.0% and 43.2% with the mean value 36.5%, and PUFA values ranged between 31.30% and 41.90% with the mean value 36.2%. In our study, SFA content was observed as 38.46%–62.81% in chocolates and 34.75–87.18% in chocolate wafers. MUFA content was determined as 28.06%–53.87% in chocolates and 7.62–53.01% in chocolate wafers. PUFA content was observed, to be 4.09%–15.24% in chocolates and 4.32–20.52% in chocolate wafers (Table 3). Indicating that, SFA levels were higher in Turkey than in Austria but PUFA levels were lower than in Austria.

Karabulut (2007) investigated fatty acids compositions and TFAs contents in 134 frequently consumed foods including 19 samples of chocolates in Turkey. The author categorised 7 groups of chocolates. He stated that chocolate samples contained TFAs less than 0.17 g/100 g fatty acids, with the exceptional national product of chocolate bars and hazelnut cocoa cream (2.03 and 3.68 g/100 g fatty acids, respectively). He observed that the dark and bitter chocolate samples were found as *trans* free products in their categories. In our study, we analysed more chocolate samples for fatty acids composition and TFAs content than did Karabulut (2007). Similarly, we found that bitter chocolates were *trans* free products. Karabulut (2007) found that chocolate bar and coconut bar covered with chocolate contained the highest lauric acid (24.87–13.60%) and myristic acid (9.38–9.27%) contents. Similarly, in our study, pure chocolate wafers and chocolate wafers with nuts contained higher levels of lauric (5.11–4.08%) and myristic (2.97–2.14%) acids than other groups.

Mojska *et al.* (2006) determined TFAs in foods in Poland including 12 chocolate products. TFAs were detected to make 7.86% in the chocolate products. We observed that the minimum–maximum range of TFAs content was 0.00–6.23% for chocolates and 0.00–7.92% for chocolate wafers. Innis *et al.* (1999) determined TFAs content in over 200 foods including 9 chocolate bars in Canada. They observed that TFAs content varied between 0.1% and 35.9% with average values 9.16% in chocolate bars. They stated that the use of average values for theTFAs content in a food category was of a limited value.

Fu *et al.* (2008) investigated TFAs in national-brand western-style products including 10 chocolate samples in China. The authors determined the average contents of TFAs as 0.80%, 0.57%, and 1.87% of total fatty acids in chocolate with nuts, dark chocolate, and milk chocolate, respectively. In our study, we determined the average TFAs contents as 1.02%, 0.00%, and 0.18% of total fatty acids in chocolate with nuts, bitter chocolate, and milk chocolate, respectively.

The body of short-term randomised feeding trials suggest that cocoa and chocolate may exert beneficial effects on cardiovascular risk via lowering the blood pressure, anti-inflammation, anti-platelet function, higher HDL, and decreased LDL oxidation (Ding *et al.* 2006). Similarly, Mursu *et al.* (2004) stated that cocoa polyphenols may increase the concentration of HDL cholesterol. The consumption of cocoa with dark chocolate increased the serum concentration of HDL cholesterol by 4% (Wan *et al.* 2001). Cocoa butter, a fat derived from cocoa plants and predominantly found in dark chocolate (Kris-Etherton *et al.* 1993) contains on average 33% of oleic acid, 25% of palmitic acid, and 33% of stearic acid (USDA National Nutrient Database http://www.nal.usda.gov/fnic/foodcomp/search/). Similarly, in our study, in bitter chocolates, palmitic, oleic, and stearic acids contents were determined as 25.11%, 33.80%, and 34.66%, respectively. Stearic acid was suggested to be a non-atherogenic type of dietary saturated fat (Ding *et al.* 2006) and the most recent trial also shows the effects of stearic acid on lipids to be even similar to those of oleic and linoleic acids (Thijssen & Mensink 2005).

The results of this research have demonstrated that fatty acids composition and TFAs contents of chocolate vary. As a result of the extensive coverage of the present research which included all chocolate and chocolate wafers brands in Turkey, it was shown that *trans* fatty acid levels in chocolates and chocolate wafers on Turkish markets were relatively lower than those of other countries.

References

Ascherio A., Willet W.C. (1997): Health effects of *trans* fatty acids. American Journal of Clinical Nutrition, **66**: 1006–1010.

Ayagari A., Peepies J.M., Carlson S.E. (1996): Relationship of isomeric-fatty acids in human cord blood to N3 and N6 status. Pediatric Research, **39**: 1808–1808 (Abstract).

Basol B., Tasan M. (2008): Fatty acid compositions of Turkish shortenings with emphasis on trans fatty acids. Journal of Food Lipids, **15**: 240–250.

Cetin M., Yildirim A., Sahin A.M. (2003): Determination of fatty acids and some undesirable fatty acid isomers in selected Turkish margarines. European Journal of Lipid Science and Technology, **105**: 683–687.

Daglioglu O., Tasan M. (2003): Fatty acid composition of traditional fermented and unfermented Turkish corn bread with the emphasis on *trans* fatty acids. European Food Research and Technology, **217**: 125–127.

Demirbas A., Yilmaz N. (2000): *Trans* fatty acid content of edible margarines in Turkey. Deutsche Lebensmittel-Rundschau, **96**: 136–138.

Demmelmair H., Festl B., Wolfram G., Koletzko B. (1996): *Trans* fatty acid contents in spreads and cold cuts usually consumed by children. Zeitschrift für Ernährungswissenschaft, **35**: 235–240.

Ding E.L., Hutfless S.M., Ding X., Girotra S. (2006): Chocolate and prevention of cardiovascular disease: A systematic review. Nutrition & Metabolism, **3**: 1–12.

Folch J., Lees M., Sloane Stanley G.H. (1957): A simple method for the isolation and purification of total lipides from animal tissues. Journal of Biological Chemistry, **226**: 497–509.

Fritsche J., Steinhart H. (1998): Analysis, occurence, and physiological properties of *trans* fatty acids (TFA) with particular emphasis on conjugated linoleic acid isomers (CLA) – a review. Fett/Lipid, **100**: 190–210.

Fu H., Yang L., Yuan H., Rao P., Lo Y.M. (2008): Assessment of *trans* fatty acids content in popular western-style products in China. Journal of Food Science, **73**: 383–391.

Innis S.M., Green T.J., Halsey T.K. (1999): Variabilty in the *trans* fatty acid content of foods within a food category: implications for estimation of dietary *trans* fatty acid intakes. Journal of the American College of Nutrition, **18**: 255–260.

Judd J.T., Clevidence B.A., Muesing R.A., Wittes J., Sunkin M.E., Podczasy J.J. (1994): Dietary *trans* fatty acids: effects of plasma lipids and lipiproteins on healthy men and women. American Journal of Clinical Nutrition, **59**: 861–868.

Karabulut I. (2007): Fatty acid composition of frequently consumed foods in Turkey with special emphasis on *trans* fatty acids. International Journal of Food Sciences and Nutrition, **58**: 619–628.

Katan M.B. (1998): Health effects of *trans* fatty acids, European Journal of Clinical Investigation, **28**: 257–258.

Kris-Etherton P.M., Mustad V., Derr J.A. (1993): Effects of dietary stearic acid on plasma lipids and thrombosis. Nutrition Today, **28**(3): 30–38.

Kromer GW. (1976): Trends and patterns in soybean oil use for food and industrial products. In: Hill L.D. (ed.): World Soybean Conference. Danville. Interstate Print and Publ., Champaign: 789–804.

Mojska H., Gielecinska I., Balas J., Pawlicka M., Szponar L. (2006): *Trans* fatty acids in foods in Poland: monitoring study. Zywienie Czlowieka i Metabolizm, **33**(2): 107–122.

Mounts T.L. (1979): Hydrogenation. In: Pryde E.H. (ed.): Fatty Acids. Champaign, American Oil Chemists Society: 391–402.

Mursu J., Voutilainen S., Nurmi T., Rissanen T.H., Virtanen J.K., Kaikkonen J., Nyyssönen K., Salonen J.T. (2004): Dark chocolate consumption increases HDL cholesterol concentration and chocolate fatty acids may inhibit lipid peroxidation in healthy humans. Free Radical Biology & Medicine, **37**: 1351–1359.

Paquot C. (1979): Standards Methods for Analysis of Oils, Fats and Derivatives. IUPAC, 6th Ed. Oxford, Pergamon Press.

Semma M. (2002): *Trans* fatty acids: Properties, benefits and risks. Journal of Health Science, **48**(1): 7–13.

Stender S., Dyerberg J. (2004): Influence of *trans* fatty acids on health. Annals of Nutrition and Metabolism, **48**: 61–66.

Tarkowski A., Kowalczyk M. (2007): Fat and fatty acids chosen in chocolates content. Roczniki Panstwowego Zakladu Higieny, **58**: 351–356. (in Polish)

Thijssen M.A., Mensink R.P. (2005): Small differences in the effects of stearic acid, oleic acid, and linoleic acid on the serum lipoprotein profile of humans. American Journal of Clinical Nutrition, **82**(3): 510–516.

Wan Y., Vinson J.A., Etherton T.D., Proch J., Lazarus S.A., Kris-Etherton P.M. (2001): Effects of cocoa powder and dark chocolate on LDL oxidative susceptibility and prostaglandin concentrations in humans. American Journal of Clinical Nutrition, **74**: 596–602.

Wagner K.H., Auer E., Elmadfa I. (2000): Content of *trans* fatty acids in margarines, plant oils, fried products and chocolate spreads in Austria. European Food Research and Technology, **210**: 237–241.

Weiland S.K., Von Mutius E., Husing A., Asher M.I. (1999): Intake of *trans* fatty acids and prevalence o childhood asthma and allergies in Europe: ISAAC Steering Committee. Lancet, **353**: 2040–2041.

Zock P.L., Katan M.B. (1992): Hydrogenation alternatives: effects of *trans* fatty acids and stearic acid versus linoleic acid on serum lipids and lipoproteins in humans. Journal of Lipid Research, **33**: 399–410.

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