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Fatty acid composition with special emphasis on unsaturated *trans* fatty acid content in margarines and shortenings marketed in Saudi Arabia

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Abstract

Margarines and shortenings from the Saudi Arabia market were analyzed for their fatty acid composition including *trans* fatty acids by gas chromatography. The saturated, monounsaturated and polyunsaturated fatty acids within the investigated margarines and shortenings were found to be in the range of 19.8-29.3%, 27.0-30.2%, 37.6-43.9% and 25.9-50.2%, 28.4-41.4%, 10.6-29.8% respectively. *Trans* fatty acids (TFAs) in margarines and shortenings were found to be in the range of 0.2-8.3% and 0.9-20.6% respectively. The mean of the sum of lauric acid ($C_{12:0}$), myristic acid ($C_{14:0}$) and palmitic acid ($C_{16:0}$) fatty acids in margarines and shortenings were found to be 19.15±6.57%.and 25.15±12.79% respectively. From the results of this study it has been found that the highest concentrations of TFAs were found in the sample containing least amount of linoleic acid and the lowest concentration of TFAs were found in the sample containing highest amount of linoleic acid. Higher *trans* contents were observed in shortenings compared to margarines. In various studies, TFAs in the high levels in the diet have repeatedly been shown to affect serum lipids/lipoproteins unfavorably and increased the risk of coronary heart diseases.

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Introduction

The fatty acid composition of triglycerides has a direct effect on the physical, chemical and biological properties of dietary fats (Alonso *et al.*, 2000).*Trans* fatty acids (TFAs) are the fatty acids containing one or more double bond in the *trans* configuration that are formed during hydrogenation process used in the food industry to harden oils (Griguol Chulich *et al.,* 2005) with more desirable physical properties, quality and texture. It occurs naturally in dairy and is predominant in vegetable oils and animals fats (Valenzuela and Morgado, 1999). Partially hydrogenated vegetable oils containing *trans* fatty acids have been used in food industry due to its low cost compared with other fats, ability to impart desirable characteristics to the food and availability to extend the shelf-life of products (Albuquerque *et al.*, 2011). The principal disadvantage of hydrogenation is that it removes large amount of the essential fatty acids which are present in the natural oils. Margarines, which is considered as more beneficial as compared to butter are preferentially consumed in processed foods, are among the main TFAs sources in the diet of Saudis and several western countries (Wolff, 1995; Ratnayake, *et al.,* 1998).

Since TFAs have higher melting points and better stability than their cis isomers, so formation of TFAs isomers was considered as a beneficial side reaction (Petrauskaite *et al.*, 1998). But several studies have shown that the consumption of some TFAs formed by the partial hydrogenation of oils and fats have negative health effects as it increases the risk of diseases (Alonso *et al.*, 2002).

Consumption of industrial TFAs at high levels have regularly been shown to affect serum lipids/lipoproteins adversely (Ovesen *et al.*, 1998). Higher intake of TFAs have been found to raise lipoproteins (Mensink *et al.*, 1992; Zock *et al.*, 1998; Valenzuela and Morgado, 1999; Stachowska *et al.*, 2001) and also serum low density lipoproteins (LDL) (Zock *et al.*, 1998; Valenzuela and Morgado, 1999; Stachowska *et al.*, 2001; Griguol Chulich *et al.*, 2005; Attia-Shikhri *et al.*, 2009) to a degree similar to saturated fats and it also decreases high density lipoproteins (HDL) (Zock *et al.*, 1998; Judd *et al.*, 1998; Valenzuela and Morgado, 1999; Stachowska *et al.*, 2001; Griguol Chulich *et al.*, 2005; Attia-Shikhri et al., 2009). Therefore trans isomers are considered more atherogenic than saturated fatty acids (Valenzuela and Morgado, 1999). Several epidemiological and clinical studies have reported adverse health effects resulted from the consumption of TFAs (Mensink *et al.*, 1992). In a study it has been found that replacement of dietary saturated fatty acids by *trans* fatty acids impaired flow- mediated vasodilation (FMD) of the branchial artery, which suggests increased risk of coronary heart disease (De Roos *et al.*, 2001). *Trans* fatty acids also affect early infant growth and development and increases the risk and incidence of diabetes (Ghafoorunissa, 2008). Since hydrogenated fats constitute a major portion of dietary fat and has been involved in various health related issues, it is important to have detailed information of fatty acids composition of margarines and shortenings. Although fatty acids composition of hydrogenated fats has been extensively analyzed in various parts of the world, the information on fatty acids composition is however limited on the margarines and shortenings used in Saudi Arabia. The objective of this work was to study the fatty acids composition with special emphasis on *trans* fatty acids in several margarines and shortenings marketed in Saudi Arabia.

Materials and methods

Sample description and preparation of FAME for GC analysis

Four brands of margarines and six brands of shortenings were purchased from retail supermarket in Riyadh, Saudi Arabia. The choice of the brands was based on the highest consumption among those available in the market. All the chemicals used were of analytical grade. Pure standards of FA methyl esters (FAMEs) were purchased from Sigma Chemical Co. (St Louis, MO).The fat sources listed on the package were recorded in Table 1.

Table 1. Fat source of Margarine and shortening samples.

Product Code	**Fat Source**
Margarine	
PAR	Partially hydrogenated soybean oil+ liquid soybean oil + buttermilk
FLR	Sunflower oil, vegetable oils (canola oil, palm kernel oil, palm oil)
MAR	Pure corn-partially hydrogenated palm
MAU	Pure corn-partially hydrogenated palm
Shortenings	
CRS	Partially hydrogenated soybean and cotton seed oils with mono and diglycerides
MAZ	Pure corn oil, partially hydrogenated palm oil
DAL	Double refined and hydrogenated palm oil with mono and diglycerides
GDY	Partially hydrogenated soybean oil and palm with mono and diglycerides
MAC	Pure corn oil, partially hydrogenated palm oil
AMG	Partially hydrogenated soybean and cotton seed oils with mono and diglycerides

Table 2. Fatty acids composition (% total fatty acid) of the selected brands of margarine and shortenings available in Riyadh, Saudi Arabia.

	Margarines				**Shortening**					
	PAR	FLR	MAR	MAU	CRS	MAZ	DAL	GDY	MAC	AMG
Saturated fatty acids (SFA)										
Caprylic ($C_{8:0}$)	-	0.3	-	-	-	-	-	-	-	-
Capric ($C_{10:0}$)	-	0.3	-	-	-	-	-	-	-	-
Lauric ($C_{12:0}$)	-	4.4	0.2	0.2	-	0.2	0.2	0.1	0.2	-
Myristic ($C_{14:0}$)	0.1	1.7	0.5	0.4	0.2	0.6	1.0	0.3	0.6	0.1
Palmitic ($C_{16:0}$)	11.1	10.2	23.8	24.0	14.3	31.2	44.0	16.7	29.1	12.1
Margaric ($C_{17:0}$)	0.1	-	0.1	-	-	0.1	0.1	0.1	-	0.1
Stearic ($C_{18:0}$)	7.7	7.8	4.1	4.2	12.2	6.5	4.5	8.8	4.2	13.1
Arachidic ($C_{20:0}$)	0.8	0.5	0.4	0.5	0.4	0.5	0.4	0.5	0.4	0.5
Total SFA	19.8	25.2	29.1	29.3	27.1	39.1	50.2	26.5	34.5	25.9
Unsaturated fatty acids										
Palmitoleic ($C_{16:1}$)	0.1	0.1	0.1	0.1	-	0.1	0.1	0.1	0.1	0.1
Oleic ($C_{18:1}$)	26..6	29.2	29.8	29.5	31.0	28.1	37.4	33.5	29.6	41.1
Gadoleic ($C_{20:1}$)	0.3	0.5	0.3	0.3	0.2	0.2	0.1	0.2	0.2	0.2
Total MUFA	27.0	29.8	30.2	29.9	31.2	28.4	37.6	33.8	29.9	41.4
Linoleic ($C_{18:2}$)	39.3	40.6	37.0	37.1	26.6	29.4	10.9	22.1	13.8	10.3
Linolenic ($C_{18:3}$)	4.6	3.3	0.5	0.5	2.4	0.4	0.3	1.2	0.5	0.3
Total PUFA	43.9	43.9	37.5	37.6	29	29.8	11.2	23.3	14.3	10.6
Total TFA	8.3	0.2	4.2	3.2	12	2.6	0.9	14.8	3.3	20.6

Table 3. Trans fatty acids composition of the selected samples of margarines and shortenings available in Riyadh, Saudi Arabia.

	Margarines				Shortening					
	PAR	FLR	MAR	MAU	CRS	MAZ	DAL	GDY	MAC	AMG
Trans (18:1)	6.9	-	2.8	3.9	9.5	2.1	0.5	10.2	2.7	15.7
Trans (18:2)	0.9	0.1	0.4	0.3	2.5	0.5	0.4	4.4	0..6	4.8
Trans (18:3)	0.5	0.1	-	-	-	-	-	0.2	-	0.1
Total Trans	8.3	0.2	3.2	4.2	12.0	2.6	0.9	14.8	3.3	20.6

A sample of ten packages from each product/brand was homogenized and study samples were taken and converted to methyl esters according to AOCS, 1997. Methyl esters were recovered and separated by GLC using a SP 2560 (1100Mx0.25 mm id, 20mm film thickness). Pure hydrogen was used as carrier gas with a flow rate of 8ml/ min. Operating conditions were injection at 240°C and the flame ionization detector (FID) temperature was 250°C.Identification of trans and cis fatty acids were based on reference standards.

Results and discussion

The details of the fat source listed on the packaging materials are provided in Table-1. All margarines and shortenings were mixture of a number of oils in their native state with fat and oils hydrogenated or partially hydrogenated. Table-2 depicts the fatty acids composition of the margarine and shortenings. A large section of people consumes margarines and shortenings because of their eating habits and lifestyles although nutritional characteristics and health consequences of these products have been repeatedly mentioned in various studies.

Caprylic acid ($C_{8:0}$) and capric acid ($C_{10:0}$) was absent in all studied margarines and shortenings except the margarine coded as FLR. Lauric acid ($C_{12:0}$) was not detected in one sample of margarine (PAR) and two samples of shortenings (CRS and AMG). The sum of lauric acid ($C_{12:0}$), myristic acid ($C_{14:0}$) and palmitic acid ($C_{16:0}$) fatty acids in margarines were11.2,16.3, 24.5 and 24.6 % in PAR, FLR, MAR and MAU respectively with a mean of 19.15±6.57%. Alonso *et al.*, 2000 studied the fatty acid composition of 12 Spanish margarines, and 7 margarines had lesser (sum of C $_{12:0}$, $C_{14:0}$ and $C_{16:0}$) value than the result obtained in this study. Ovesen *et al.*, 1998 obtained a slight higher value (sum of $C_{12:0}$, $C_{14:0}$ and $C_{16:0}$) in margarines (marketed in Denmark) having 20-40% of linoleic acid than the value obtained in this study. The sum of lauric acid ($C_{12:0}$), myristic acid ($C_{14:0}$) and palmitic acid ($C_{16:0}$) fatty acids in shortenings were14.5, 32.0, 45.2, 17.1, 29.9 and 12.2% in CRS, MAZ, DAL, GDY, MAC, AMG respectively with a mean of 25.15±12.79%. Ovesen *et al.*, 1998 obtained a slight higher value (sum of $C_{12:0}$, $C_{14:0}$ and $C_{16:0}$) in shortenings (marketed in Denmark) than the value obtained in this study. Palmitic acid ($C_{16:0}$) was found to be at the highest level (dominant fatty acid), both in margarines and shortenings. Margaric acid ($C_{17:0}$) was not identified in two samples of margarines (FLR and MAU) and in two samples of shortenings (CRS and MAC). The saturated fatty acids within the investigated margarines were found to be in the range of 19.8-29.3%. The amount of saturated fatty acids within the investigated shortenings (33.8±9.57%) were more than, in margarines (25.85±4.53%). In thirty brands of margarines marketed in Greek, the range of saturated fatty acid reported was 11.26-51.75% (Kroustallaki *et al.*, 2011). Similarly, in another study, in 15 margarine samples (marketed in Greek), this range was 24.1-53.3% (Triantafillou *et al.*, 2003). Total saturated fatty acids obtained by Ovesen *et al.*, 1998 in their study was higher than the result obtained in this study. The sum of saturated lauric acid ($C_{12:0}$), myristic acid ($C_{14:0}$) and palmitic acid ($C_{16:0}$) fatty acids, suspected to have higher atherogenic potential than stearic acid. The short chain fatty acids, especially the short chain fatty acids lauric, myristic acids and palmitic, elevate blood cholesterol (Mensink and Katan, 1992). Total MUFA in margarines were 27.0, 29.8, 30.2 and 29.9% in samples PAR, FLR, MAR and MAU respectively. Similarly total MUFA in shortenings were 31.2, 28.4, 37.6, 33.8, 29.9 and 41.4% in CRS, MAZ, DAL, GDY, MAC and AMG respectively. Average MUFA content reported in this study was 29.22±1.43% and 33.72±4.97% in margarines and shortenings respectively. Oleic acid ($C_{18:1}$) was the major MUFA identified in all samples of margarines and shortenings. A small amount of palmitoleic acid ($C_{16:1}$) was also detected in different samples of margarines and shortenings except in CRS (shortening). Average of total PUFA was higher in margarines (40.73±3.67%) than in shortenings (19.7±8.78%). Total PUFA in margarines were reported as 43.9, 43.9, 37.5 and 37.6% in PAR, FLA, MAR and MAU respectively. Total PUFA in shortenings were 29, 29.8, 11.2, 23.3, 14.3 and 10.6% in CRS, MAZ, DAL, GDY, MAC and AMG respectively. Total PUFA content was higher in the samples having greater content of linoleic acid. Essential fatty acids are of key importance for biological and nutritional value of dietary fats.

Trans fatty acids composition of margarines and shortenings are shown in Table 3. As shown in Table 3 only one margarine (FLR) and one shortening (DAL) had total *trans* percent less than 1, otherwise all other samples had high total *trans* content, among which trans content of AMG (shortening) was highest. From the results of this study it has been found that the highest concentrations of TFAs were found in the sample containing least amount of linoleic acid and the lowest concentration of TFAs were found in the sample containing highest amount of linoleic acid. Trans 18:1 was the dominant *trans* fatty acid found in this study. In observational studies utilizing biomarkers of TFA consumption, both 18:1 and 18:2 isomers appear to contribute to risk of CHD; conversely most studies did not detect any effect of 16:1 TFA (Uauy *et al.*, 2009). Higher *trans* contents observed in shortenings (which may be due to presence of greater percentage of hydrogenated oil) compared to margarines are similar to observations described previously (Bayard and Wolff; 1995 Henninger and Ulberth, 1996; Ovesen *et al.,* 1998). TFAs in the high levels in the diet have repeatedly been shown to affect serum lipids/lipoproteins unfavorably (Bhangar and Anwar, 2004). Epidemiological studies suggest a relationship between TFAs and increased risk of coronary heart diseases (Ovesen *et al.*, 1998; Bhangar and Anwar, 2004). Ovesen *et al.*, 1996 in his study has mentioned that significant amount of *trans* 18:1 were found only in hard margarines and shortenings whereas semisoft and soft margarines contained substantially less *trans* 18:1. Total TFAs reported by Kroustallaki *et al.*, 2011 in their study on 31 brands of margarines from Greek market was lesser than the values obtained in this study. TFAs reported by them was in the range of 0.16%-0.97% with no sample found to have TFAs in a concentration more than 1% of total fat content. Similarly the *trans* fatty acid content of margarines reported by Triantafillo *et al.*, 2003 varied from 0.1 to 19%.*Trans* fatty acids reported in German margarines and shortenings were 0.17-25.90% and 0.04-32.51% respectively (Molkentin and Precht, 1995). A comparison of the mean of total TFA values for the investigated margarines (3.975%) was lower than the Greece (10%) (Kafatos *et al.*, 1994) and higher than the France (3.80%) (Bayard and Wolff 1995). In a study on the fatty acid composition of Spanish shortenings the amount of *trans*-18:1 isomers showed a wide range of variation, that is; from 0.4 to 69%, although more than 63% of shortenings contained <3% of trans-18:1. The mean *trans*-18:2 isomers content was 0.58%, ranging from 0 to 3.4%. Small amounts of *trans*-18:3 isomers (<0.3%) were also observed (Alonso *et al.*, 2002).

A low intake of TFA is required to reduce the health risks because a high intake of industrially processed *trans* fatty acid is associated with increased risk of coronary heart disease probably through effect on lipoprotein. Dietary intake of such fatty acids increases the level of LDL cholesterol and at the same time reduces the level of HDL cholesterol in plasma. However, an interventional trial allowed for a comparision of TFAs from different sources suggests that the intake of natural TFAs have no or neutral effects on plasma lipids and other cardiovascular risk factors. But the researchers suggested further investigations as the mechanism underlying the isomer specific effects were not well understood (Attia Skhiri *et al.*, 2009). A high intake of *trans* fatty acids in children may be disadvantageous because of the unwanted effects on lipoprotein metabolism and a possible impairment of arachidonic acid synthesis (Demmelmair *et al.*, 1996). Reviews of epidemiologic studies have suggested that adverse effects of TFA from hydrogenated sources were related with some inflammatory markers (Gebauer *et al.*, 2007). During refining of vegetable oils, deodorization step contributes to formation of TFA, so it is therefore recommended that the severity of deodorization conditions should be reduced to limit TFA content in refined oils <1% of total fatty acids (Ghafoorunissa, 2008).

Conclusion

The process of partial hydrogenation hardens vegetable oils that are commonly used for deep frying in fast food chains, restaurants and bakery. Because there are no nutritional benefits of *trans* fatty acids and its consumption has been associated with negative health effects, therefore its use should be reduced practically in food. Consumer education about negative health effects of TFAs by proper labeling and providing food based guidelines to reduce TFA consumption in the entire population need to be actively pursued.

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