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Trans fatty acid content of Brazilian biscuits

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Abstract

The fatty acid composition and *trans* fatty acid (TFA) contents of samples of five brands Brazilian cream cracker biscuits were determined by gas–liquid chromatography, using a polar 100 m capillary column CP Sil-88 and flame ionization detection. The identification of fatty acids done by equivalent chain length for *trans* fatty acids. Total TFA ranged from 12.2% to 31.2% of total fatty acid and the mean was 20.1%. *Trans* 18:1 isomers were the major group of TFA present in all the analyzed brands, representing 83.2% of total *trans* isomers. The mono-*trans* 18:2 isomer content ranged from 1.6% to 4.2% of total fatty acids, this being the most prevalent group of *trans* polyunsaturated acid. The di-*trans* 18:2 isomer (9*t*, 12*t*) was found at very low levels (0.10–0.15% of total fatty acids). *Trans* 18:3 isomer content ranged from 0.11% to 0.75% of total fatty acids representing 24.4–75.0% of total α -linolenic acid. The results indicate that Brazilian cream cracker biscuits contain considerable proportions of *trans* fatty acids, both monounsaturated and polyunsaturated.

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1. Introduction

Coronary heart disease (CHD) remains the leading cause of death and disability in many countries around the world, including Brazil. According to Maranhão (1999), ca. 34% of all deaths in Brazil are caused by CHD. There are several multiple risk factors that act both independently and jointly. Among dietary factors, the type of fat intake and total amount of fat in the diet play important roles in determining risk of CHD (Hu, Manson, & Willett, 2001).

It has been known for many years that a high intake of saturated fat contributes to the development of CHD. More recently, *trans* fatty acids (TFA) have also been

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associated with adverse effects. Several studies have shown that a high intake of TFA raises low density lipoprotein (LDL) cholesterol and lowers high density lipoprotein (HDL) cholesterol, affecting the LDL/HDL cholesterol ratio in a way that is unfavourable compared with all other fatty acids. Moreover, *trans* fats have been reported to raise lipoprotein (a) and plasma triglyceride levels, that are independently associated with the increased risk of CHD (Aro, Jauhiainen, Partanen, Salminen, & Mutanen, 1997; Ascherio, Katan, Zock, Stampfer, & Willett, 1999; Mensink & Katan, 1990; Mensink, Zock, Katan, & Hornstra, 1992; Nestel et al., 1992).

Trans isomers are mainly present in fats produced by partial hydrogenation of vegetable or marine oils, but they also occur in minor quantities in dairy and other animal fats by biological hydrogenation of the unsaturated fatty acids in the rumen. In addition, *trans* isomers

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may be formed in small amounts in vegetable oils during deodorization and food-frying operations (Ackman & Tag, 1998).

In Brazil, commercial hydrogenation of vegetable oils dates from the late 1950s, for producing shortening and hard margarine. With the development of partial and selective hydrogenation techniques, processed vegetable fats rapidly displaced animal fats in the diet of the Brazilian people. These fats have been widely used in the production of several foods, such as margarine, chocolate spreads, biscuits, potato chips, and bakery products. In past decade, biscuit per capita consumption has increased twofold in our country, becoming an important source of dietary fat (Coelho, Gramacho, Bressan-Filho, Contini, & Venturili, 2000; Pelaez-Alvarez, Szmrecsányi, & Tango, 1991).

Thus, the aim of this study was to determine the *trans* fatty acid composition of the cream cracker, a type of biscuit frequently consumed in Brazil.

2. Materials and methods

2.1. Sampling

Twelve units of each brand of cream cracker biscuits were purchased from local supermarkets and bakeries between December 2002 and June 2003. Each brand was coded with a letter (A, B, C, D, and E). Lot numbers were checked to ensure that each unit belonged to a different lot. Samples were selected to include the major manufacturers of the biscuits in Brazil. The analyses were carried out in triplicate.

2.2. Methods

Total lipids were extracted using a procedure published by Folch, Lees, & Stanley (1957) and fatty acid methyl esters were prepared by methylation of fatty acids, as described by the method of Hartman & Lago (1973).

Fatty acid methyl esters (FAME) were analyzed by using a Shimadzu model 14A gas chromatograph, equipped with a flame ionization detector (FID) and a fused silica capillary column CP Sil-88 (100 m \times 0.25 mm id., 0.20 m film thickness, Chrompack). Hydrogen carrier flow was 1.3 ml/min at a split ratio of 1/100. Injector and detector temperatures were both 250 °C. The column was operated isothermally at 170 °C. Retention times and peak area percentages were determined by the CG-300 computing integrator (CG Instruments). The identification of fatty acids was based on authentic reference standards (Sigma) and on literature reports of equivalent chainlength (ECL) for *trans* fatty acids (Pawlowicz & Drozdowski, 1998; Ratnayake & Pelletier, 1992; Wolff, 1992). The values of ECL were determined for all analyzed acids using a method described by Ackman (1972), with methyl esters of 16:0, 18:0, and 20:0 acids as reference compounds. The final results are expressed as relative percentages.

2.3. Statistical analysis

The samples were analyzed, one by one, in triplicate, and the results were expressed as mean values \pm standard deviation (SD). The results were compared using analysis of variance (ANOVA) with 5% significance level using Statistica 5.0 software (StatSoft, USA, 1995). The average values were compared by Tukey's test.

3. Results and discussion

Fatty acid compositions of cream cracker are presented in Table 1. The amount of total TFA in the samples ranged from 12.2% to 31.2% of total fatty acids and the mean value was 20.1%. Total *trans* content was significantly higher (P < 0.05) in brands A and E. The TFA comprised isomers of 16:1, 18:1, 18:2, and 18:3 acids and *trans* 18:1 isomers were the major group of TFA present in all the brands analyzed, ranging from 8.8% to 28.3%. The mean value represented 83.2% of the total *trans* isomers. For the *trans* 16:1 isomer, only small amounts (0.25% of total fatty acids) were detected in some lots of brand C.

Trans polyunsaturated acids included geometrical and positional isomers of linoleic acid for all the brands analyzed and geometrical isomers of α -linolenic acid only for brands B, D, and E. The mono-*trans* 18:2 isomer content ranged from 1.6% to 4.2% of total fatty acids being the most prevalent group of *trans* polyunsaturated acid. The di-*trans* 18:2 isomer (9*t*, 12*t*) was found at very low levels (0.10–0.15% of total fatty acids). *Trans* 18:3 isomer content ranged from 0.11% to 0.75% of total fatty acids, representing 24.4–75.0% of total α -linolenic acid.

These results show that the amount of *trans* monounsaturated and polyunsaturated varied considerably among the analyzed brands because of the differences in hydrogenation process conditions, such as temperature, pressure, type and amount of catalyst and agitation rate affect the resulting TFA content of the starting oil. Moreover, food producers may use single hydrogenated fats or many possible combinations of hydrogenated and non-hydrogenated fats and oils to achieve a final product with the desirable characteristics (Innis, Green, & Halsey, 1999; Karabulut, Kayahan, & Yaprak, 2003).

For similar biscuits, the *trans* content found was 12.7% for US (Enig, Pallansch, Sampugna, & Keeney, 1984), 11.1% for Argentina (Tavella et al., 2000), 9.1% for Greece (van Erp-baart et al., 1998) and 2.0% for New Zealand (Lake, Thomson, Devane, & Scholes,

Table 1 Fatty acid composition of Brazilian cracker biscuits^a

Fatty acid	Brands				
	A	В	С	D	Е
14:0	$0.10 \pm 0.02a$	$0.10 \pm 0.02a$	$1.47 \pm 1.08b$	$0.11 \pm 0.03a$	$0.12 \pm 0.03a$
14:1c	ND	ND	0.88 ± 0.73	ND	ND
16:0	$11.5 \pm 0.46a$	$10.9 \pm 1.04a$	$20.2 \pm 5.53b$	$11.2 \pm 0.76a$	$11.8 \pm 0.38a$
16:1c	ND	ND	2.09 ± 1.65	ND	ND
16:1t	ND	ND	0.25 ± 0.20	ND	ND
18:0	$11.5 \pm 1.30a$	$4.94 \pm 0.20b$	$17.2 \pm 9,50c$	$5.53 \pm 0.49b$	9.99 ± 1.26a
18:1c	$34.5 \pm 1.18a,b$	36.6 ± 1.03 a,b	$33.6 \pm 4.78b$	33.9 ± 1.33 a,b	$36.8 \pm 4.30a$
18:1t	$28.3 \pm 2.70a$	$8.80 \pm 0.82b$	$10.4 \pm 8.82b$	$9.75 \pm 0.80b$	$25.2 \pm 5.63a$
18:2cc	$9.34 \pm 1.50a$	$31.2 \pm 1.01b$	$10.3 \pm 3.51a$	$32.60 \pm 1.04b$	$11.05 \pm 2.68a$
18:2tt	$0.15 \pm 0.08a$	$0.10 \pm 0.05a$	ND	$0.10 \pm 0.04a$	$0.11 \pm 0.06a$
18:2 c/t and t/c	$2.72 \pm 0.33a$	$4.20 \pm 0.49b$	$1.64 \pm 1.50c$	3.43 ± 0.46 a,b	$2.77 \pm 0.56a$
18:3ccc	$0.40 \pm 0.10a$	$1.01 \pm 0.10b$	$0.53 \pm 0.12a$	$1.00 \pm 0.11b$	$0.45 \pm 0.07a$
18:3t	ND	$0.53 \pm 0.18a$	ND	$0.75 \pm 0.10b$	$0.11 \pm 0.09c$
20:0	$0.32 \pm 0.03a$	$0.39 \pm 0.06b$	$0.25 \pm 0.07c$	$0.36 \pm 0.06a$	0.36 ± 0.07 a,b
20:1c	$0.19 \pm 0.02a$	$0.15 \pm 0.02b$	$0.21 \pm 0.04a$	$0.22 \pm 0.02a$	$0.22 \pm 0.05a$
22:0	$0.38 \pm 0.04a$	$0.39 \pm 0.09a$	$0.13 \pm 0.10b$	$0.37 \pm 0.15a$	$0.39 \pm 0.06a$
24:0	$0.14 \pm 0,03a$	$0.15 \pm 0.05a$	ND	$0.13 \pm 0.08a$	$0.14 \pm 0.02a$
SFA ^b	$23.9 \pm 1.57a$	$16.9 \pm 0.77 b$	$39.2 \pm 15.8c$	17.7 ± 3.44a,b	22.8 ± 1.32a,b
PUFA ^c	$9.74 \pm 1.59a$	$32.2 \pm 1.10b$	$10.8 \pm 3.44a$	$33.6 \pm 1.13b$	$11.5 \pm 2.74a$
TFA ^d	$31.2 \pm 2.64a$	$13.63 \pm 3.44b$	$12.2 \pm 10.3b$	$14.0 \pm 1.25b$	$28.2 \pm 5.61a$
PUFA/SFA ^e	$0.41 \pm 0.07a$	$1.91 \pm 0.08b$	$0.38 \pm 0.29a$	$1.90 \pm 0.11a$	$0.51 \pm 0.14b$
Trans/cis	$0.70 \pm 0.08a$	$0.20 \pm 0.02b$	$0.26 \pm 0.18b$	$0.21 \pm 0.02b$	$0.58 \pm 0.19a$

^a Results expressed as percentage of total fatty acid methyl esters. Values are means \pm SD for six samples of triplicate analyses. Averages followed by different letters in the same line are significantly different (P < 0.05) by Tukey's test. Fatty acids less than 0.1%: 12:0, 15:0, 17:0, 17:1, 20:2, 22:1 and not identified fatty acids.

^b SFA = saturated fatty acids.

^c PUFA = polyunsaturated fatty acids.

^d TFA = *trans* fatty acids.

1996). These values are similar to those found in the present study. However, Grecian and New Zealander biscuits, contained a high proportion of saturated fatty acids. Furthermore, higher levels were found in Canada and Suedew, 40.3% and 29.1%, respectively (Innis et al., 1999; van Erp-baart et al., 1998).

For *cis* monounsaturated 18:1, differences (P < 0.05) were observed between brands C and E, with values varying from 33.6% to 36.8%, respectively. The oleic acid (18:1n9) was the main fatty acid of this group (data are not shown); however, unusual isomers were also found. There is little information on the biological activity of these positional isomers, but a preliminary study suggested that unusual isomers can influence the growth of infants (Ayagari, Peepies, & Carlson, 1996).

Total saturated fatty acids (SFA) were significantly higher in brand C (P < 0.05), followed by brands A, E and finally by brands B and D. Brand C was an exception due to the use of lard in the production of some of the units analyzed. Among the SFA, palmitic acid (16:0) presented the highest value ranging from 10.9% to 20.2%, followed by stearic (18:0), that varied from 4.9% to 17.2%.

The content of polyunsaturated fatty acids (PUFA) ranged from 9.7% to 32.2%, being significantly higher

in brands B and D (P < 0.05). Because essential fatty acids (EFA) are included in this group, the PUFA content is very important for the biological and nutritional value of these biscuits. A disturbing feature was the combination of high levels of TFA and lower levels of essential fatty acids in brands A and E.

It should be stressed that the Department of Health (UK) (HMSO, 1994) recommends a minimal PUFA/SFA ratio value of 0.45, and this was observed only in brands B, D, and E, with values of 1.91, 1.90, and 0.51, respectively.

4. Conclusion

The results in the present study indicate that Brazilian cream cracker biscuits contain considerable proportions of *trans* fatty acids, both monounsaturated and polyunsaturated.

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