

Analytical, Nutritional and Clinical Methods Section

Trans fatty acid content of a selection of foods in Argentina[☆]

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Abstract

Several studies have reported an association between consumption of trans fatty acids and risk of cardiovascular disease (CVD). These fatty acids enter the human diet most commonly as byproducts of hydrogenation of polyunsaturated fats. The amount of trans fats in foods exhibit great variation, due to differences in hydrogenation methods and intensity. In order to quantify the level of trans fats available in widely consumed commercial food items in Argentina, we measured total fat, saturated fat, and the trans fatty acid elaidic acid in 46 food items. As an example from most common items, total fat was 2.0–3.4% in sliced bread, 2.9–25% in cookies and crackers, 50–80% in margarines, 85% in butter, and 34–39% in snack products. In the same items, content of the trans fatty acid elaidic acid was: 2.35–27.7% in sliced bread, 2.85–28.95% in cookies and crackers, 18.15–31.84% in margarines, 4.63% in butter, and 0–10.58% in snacks. In order to compare the results on the fatty-acid composition by using different analysis methods, the same food items mentioned were analyzed in a column of lower polarity and shorter length, and we found trans fatty acids were masked by *cis* unsaturated fatty acids. A comparison with available data from similar products from other parts of the world indicates that Argentinian products in the categories studied have higher content of trans fatty acids. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Trans fatty acids; Dietary fat; Food composition; Argentina; Cardiovascular disease

1. Introduction

Non-communicable diseases represent the greatest health problem in industrialized countries and a rapidly growing problem in developing countries (Caballero & Rubinstein, 1997). It is estimated that by the year 2020, over 60% of deaths in the developing regions of the world will be linked to non-communicable chronic diseases (Lancet, 1997).

Argentina has the fourth highest rate of cardiovascular mortality in the Americas (PAHO, 1990). It has been shown that excess consumption of saturated fats

increases plasma total and LDL-cholesterol levels, thus increasing the risk for atherosclerotic vascular disease (WHO, 1990).

The high consumption of saturated fatty acids and cholesterol is mainly responsible for hypercholesterolemia, (Kromhout et al., 1995) which is in turn responsible for the increase of cardiovascular morbidity and mortality of ischemic origin. (Neaton & Wentworth, 1992). In order to reduce the saturated fat content of processed foods, the food industry in developed countries moved progressively from animal fat to vegetable fat sources. These vegetable oils have a high content of unsaturated fats, which are liquid at room temperature. In order to simulate the consistency of saturated fat, vegetable oils undergo a process of partial resaturation, called hydrogenation. Trans fatty acids are isomers generated by this process, to an extent that will depend, among other factors, on the intensity of the hydrogenation. The most common trans fatty acid resulting from hydrogenation is elaidic acid (18:1 n9 trans).

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[☆] In memory of Elisabeth Cavallero, M.D. (1958–1999). The authors of this study wish to pay tribute to our colleague and friend Elisabeth Cavallero, a remarkable researcher of international level who left us when she had just acquired her scientific maturity. For her energy and enthusiasm she will be a permanent stimulus for our scientific work.

Several clinical studies have shown that a high-trans fatty acid diet causes adverse changes in the plasma lipoprotein profile, with an increase in LDL and a decrease in HDL (Khosla & Hayes, 1996). Some epidemiological studies have also found a positive association between level of trans fatty acid intake and risk of cardiovascular disease (Ascherio & Willett 1997; Ascherio, Hennekens, Buring, Master, Stampfer & Willett, 1994; Hu et al., 1997). In part, due to these concerns, the Food and Agriculture Organization (FAO) and the World Health Organization (WHO) recommended in 1994 that fats for human consumption contain less than 4% of the total fat as trans, and urged the food industry to reduce the presence of trans fats in their product to these levels. (WHO, 1993)

The present study was undertaken to quantify the level of the most common trans fatty acid isomer, elaidic acid, in several commercial food products sold in Argentina.

2. Materials and methods

2.1. Selecting the type of food items to analyze

The food items to analyze were selected according to the results of a survey carried out by the National Institute of Statistics and Census, INDEC. This survey showed that over the last 10 years the consumption of industrial bread had increased twofold and that of cookies and crackers had increased fourfold (Instituto Nacional de Estadística y Censos, INDEC, 1992). These data prompted our group to examine the lipid composition of these and related products, which has not been reported before. Additional products were selected based on already recognized frequency of use (mayonnaise, oil, margarine and butter), or because they are used in the manufacture of other products. Representative best-selling brands from each product category were selected, based on supermarket chains inquiries. Samples of sliced white bread, margarine, butter, cookies, crackers, and snacks (potato crisps, cheetos, cheese flavored sticks) were included.

2.2. Sample preparation

One unit of each brand of each product category was purchased every month for 6 months. Lot numbers were checked to insure that each unit belonged to a different lot. A pooled sample was prepared by combining portions of each unit. An aliquot of this pooled sample was taken, divided in two, and each analyzed separately. All solvents and reagents used were of analytical grade. Standards of fatty acid methyl esters of 99% purity were purchased from NuCheck Prep, Inc (Minnesota, USA). Total lipids were extracted with chloroform: methanol

(2:1 v/v) using the Folch technique (Folch et al., 1957). Total fat was determined by extracting lipids with a Folch mixture (chloroform:methanol 2:1 v/v) and then making a partition with the 20% v/v water of the resulting extract, which was dried in a N₂ current until constant weight. Fat content was expressed as % w/w of the wet product. The result obtained was compared with the values declared in the container of the products and an acceptable correspondence with a difference of $\pm 10\%$ between the two was obtained.

In a second step, cholesterol was separated by saponification with OHK/methanol. The remnant of the saponification process was acidified with HCl and free fatty acids were extracted three times with petroleum ether and, after vaporizing until dry, they were transformed into methyl esters by using BF₃/methanol at 10% at 80°C for 30 min. A nitrogen atmosphere was kept as long as possible during the whole procedure. Methyl esters were extracted with hexane and were analyzed by using a Shimadzu 9 1A chromatographer modified so as to be used as capillary columns. The fatty acid composition of foods was obtained with a 50 m long and 0.25 mm inside diameter capillary column (CPSil 88, Chrompack, The Netherlands), through a comparison of the retention relative times with commercial standards. Conditions of work were: injection temperature: 250°C, detector temperature (FID): 250°C, initial temperature: 185°C, initial time: 3 min; final temperature: 230°C, rate: 3°C/min, with helium as the carrying gas, with a pressure of 19 psi and a split ratio of 70/1. Both regular fatty acids and elaidic fatty acid (18:1 n₉ trans) were analyzed, since the rest of the trans isomers were found in concentrations below 0.3%. The GLC peak areas for methyl esters were not corrected for losses of procedure and response to the detector of flame ionization, and they were considered as directly proportional to the percentages in weight.

The same samples were analyzed with another capillary column 30 m long, 0.25 mm inside diameter and 0.1 μ m film thickness (Supelco Omegawax 250, Supelco, Bellefonte, USA). Different working conditions were tried but geometrical isomers of unsaturated fatty acids were not resolved with any of them, though position isomers were actually separated. This was achieved in the same conditions used before when working with the 50 m column, as described above. Some of the results of these comparison results are shown in Table 3.

Data for total fat content are expressed as % of wet weight. Data for trans fatty acids are expressed as percentage of total fat.

3. Results

Trans fatty acids were found in all the samples of cookies and crackers, margarine, butter, and sliced

bread, though not in the processed vegetable oils, mayonnaises or potato crisps. This is consistent with the use of vegetable oils in the manufacture of the product, as stated on the label.

The content of specific fatty acids in each of the products evaluated is presented in Tables 1 and 2. The contents of total fat are expressed as percentage of wet weight and the levels (percentage p/p of total content of fatty acid) of elaidic acid in the seven groups of foods were as follows: sliced bread 2.8 and 8.20%; cookies/crackers 12.30 and 10.50%, margarine 71.0 and 27.50%; butter 85 and 4.63%; mayonnaise 32 and 0%; potato crisps 35 and 0%; cheetos and cheese-flavored sticks 38 and 6%; vegetable oils 100 and 0%.

When the results obtained in the two columns were compared, we found that oils, mayonnaises and potato crisps showed the same composition in both. As expressed before, they showed to have no trans fatty acids in their composition. The rest of the food items

studied showed discrepancies between the results in both columns, specially as regards the contents of oleic acid (18:1 n9 *cis*). This fatty acid was found in a much smaller proportion since in the column with less resolution capacity both isomers appeared together.

4. Discussion

Natural fats and oils that are present in the human diet contain only small amounts of trans fatty acids. Animal sources of fats such as milk, dairy products, and beef usually contain between 2 and 4.5% of their total fat as trans fatty acids, which are generated during rumination and subsequently absorbed and stored in animal tissues. Natural oils of vegetable origin are essentially devoid of trans fats, but, as discussed above, these fatty acids are generated during processing by hydrogenation.

Table 1

Mean percentage values of fat (in parentheses) and mean percent values of fatty acid composition of margarines, oils (sunflower, corn and mixed) and mayonnaises and percent values of fat and percent values of fatty acid composition of butter and canola oil

Fatty acid	Margarine (82%) <i>n</i> = 2	Margarine (50%) <i>n</i> = 1	Butter (85%) <i>n</i> = 1	Sunflower oil (100%) <i>n</i> = 2	Corn oil (100%) <i>n</i> = 2	Mixed seed oil (100%) <i>n</i> = 3	Rapeseed oil (Canola) (100%) <i>n</i> = 1	Mayonnaise (32%) <i>n</i> = 3
10:0	ND ^a	ND	2.30	ND	ND	ND	ND	ND
12:0	ND	ND	2.95	ND	ND	ND	ND	ND
14:0	ND	ND	11.12	ND	ND	ND	ND	ND
14:1n7	ND	ND	ND	ND	ND	ND	ND	ND
16:0	12.74	14.41	30.88	7.14	13.09	7.00	4.75	6.92
16:1n7	ND	ND	2.53	ND	ND	ND	ND	ND
18:0	11.07	10.29	14.59	3.75	2.36	3.74	1.75	3.36
18:1n 9t	25.38	31.84	4.63	ND	ND	ND	ND	ND
18:1n 9c	24.79	23.90	29.51	25.00	33.81	26.20	57.31	26.92
18:1n 7	1.94	3.58	ND	ND	ND	ND	ND	ND
18:2n6	24.13	16.00	1.54	64.12	49.87	59.13	25.01	62.57
18:3n3	ND	ND	ND	ND	0.54	2.95	11.22	ND

^a ND = not detected (< 0.3% content).

Table 2

Mean percent values of fat (in parentheses) and mean percent values of fatty acid composition of crackers, cookies, sliced bread, potato crisps and sticks, and percent values of fat and percent values of fatty acid composition of cheetos

Fatty acid	Crackers (12.1%) <i>n</i> = 15	Cookies (15.0%) <i>n</i> = 3	Sliced bread (2.7%) <i>n</i> = 7	Potato crisps (35.33%) <i>n</i> = 3	Sticks (cheese flavored) (38.0%) <i>n</i> = 2	Cheetos (38.0%) <i>n</i> = 1
10:0	ND	ND	ND	ND	ND	ND
12:0	ND	ND	ND	ND	ND	ND
14:0	2.40	1.81	1.50	ND	0.82	2.72
14:1n7	ND	ND	ND	ND	ND	2.31
16:0	23.38	23.51	18.81	7.05	10.79	25.26
16:1n7	2.25	2.29	1.37	ND	1.24	1.83
18:0	18.85	16.88	11.84	3.22	9.86	22.02
18:1n 9t	11.07	7.62	9.73	0.30	3.74	10.58
18:1n 9c	30.78	33.32	26.8	23.96	25.53	14.48
18:1n 7	0.53	ND	0.36	ND	ND	ND
18:2n 6	14.44	14.21	28.37	65.52	48.37	20.75
18:3n 3	0.48	ND	1.25	ND	ND	ND

Table 3
Comparison of fatty acid composition of some food items analyzed with two different columns

Fatty acid	Sunflower Oil		Cracker		Sodium Free Cracker		Sliced Bread No.1		Margarine No.1	
	No. 1 ^a	No. 2 ^b	No. 1	No. 2	No. 1	No. 2	No. 1	No. 2	No. 1	No. 2
14:0	ND	ND	2.78	3.31	0.7	ND	0.51	0.38	ND	ND
16:0	6.61	6.63	24.11	26.51	19.11	19.11	16.11	17.11	12.61	11.85
16:1n 7	ND	ND	2.41	2.44	ND	ND	ND	ND	ND	ND
18:0	3.81	3.84	19.11	18.19	12.61	12.35	11.19	12.09	12.14	11.32
18:1n 9t	ND	ND	5.11	ND	16.11	ND	27.71	ND	32.62	ND
18:1n 9c	25.91	24.58	38.19	40.12	23.59	35.41	21.39	42.51	23.22	51.11
18:1n 7	ND	ND	ND	ND	1.19	3.11	2.68	4.32	3.91	6.51
18:2n 6	63.72	65.11	8.81	8.46	27.52	30.31	19.79	23.81	15.72	19.22
18:3n 3	ND	ND	ND	1.11	ND	ND	1.11	ND	ND	ND

^a Capillary column no. 1 CP Sil 88.

^b Capillary column no. 2 OmegaWax 250.

In industrialized countries like the United States, where diet-related chronic diseases are a leading cause of death, there has been a strong effort to reduce the population's consumption of saturated fat. In response to this, the food industry has largely replaced animal fat with hydrogenated vegetable oils. Products manufactured with hydrogenated vegetable oils may claim a low saturated fat and low or no cholesterol content, attracting customers trying to reduce their fat intake. It is important to point out that there has been a great increase in the consumption of industrial bread, cookies and crackers in our country in the last decade (400% increased consumption of cookies and crackers, 200% increased consumption of industrial bread) (INDEC, 1992).

The possible public health relevance of dietary trans fats was emphasized by a study reporting that the level of trans fatty acid intake was directly associated with increased risk of cardiovascular disease (Hu et al., 1997).

Compiling a comprehensive and reliable database on food composition is one of the most challenging tasks for any country or region. The mass production of foods and the extremely large variety of food products available to the public makes the quantification of their nutrient composition a daunting task. Regulations on mandatory product labeling vary from country to country, but rarely include details of fatty acid composition. In many countries, there is no requirement to disclose product composition other than additive or artificial sweetener use. In those cases where there is information about fatty acids, trans fatty acids are usually not included. In summary, this report found a remarkably higher level of trans fatty acids in seven groups of food products manufactured in Argentina, relative to similar products from other parts of the world. Given the high prevalence of cardiovascular diseases in Argentina, the possible increase in risk for those diseases caused by a high trans fatty acid consumption should be carefully considered.

Finally, we would like to emphasize in the presence of the results obtained that, beside regulating on the labeling of food it would be necessary to standardize the type of instrument used — more specifically the type of column used — since on its resolution capacity depends the veracity of the information provided to the consumer.

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