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Multivariate Characterization of the Fatty Acid Profile of Spanish Cookies and Bakery Products

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The fatty acid compositions of 39 brands of cookies and bakery products were analyzed with special attention to the their *trans*-fatty acid content. The average contents (percent w/w of the total fatty acids detected) for the different nutritional fractions were as follows: saturated fatty acids (SFA), 49.43% (SD = 17.04); monounsaturated fatty acids (MUFA), 32.87% (SD = 10.94); polyunsaturated fatty acids (PUFA), 12.48% (SD = 11.29); and *trans*-fatty acids (TFA), 5.20% (SD = 9.30), the mean fat content being 22.7% (SD = 7.4). C18:1t was the predominant *trans* isomer found in all of the analyzed samples (mean = 4.04, SD = 8.46). Factor analysis performed on the fatty acid profile allowed a meaningful classification of the samples according to the main source of fat employed in their elaboration. Four factors that explained 75% of the total variance were retained. According to the results obtained, the fat used to elaborate the samples was mainly composed of vegetable and animal fat, and only in a few samples were partially hydrogenated vegetable oils used as the main source of fat.

KEYWORDS: Capillary gas chromatography; fatty acid composition; *trans*-fatty acids; bakery; cookies

INTRODUCTION

Changes in the food habits of the Spanish population have been reported, with special concern on fat intake (*1*). It is known that changes in the fat profile of the usual diet can alter serum lipid levels and induce modification in cell membranes and finally affect cell function (*2*). Metabolic and epidemiological studies have pointed out the relationship between *trans*-fatty acid (TFA) intake and coronary heart disease (*3*−*5*). Its consumption has been linked with increased lipoprotein (a) and plasma low-density lipoprotein (LDL) cholesterol at levels similar to that induced by saturated fat (*6*). Also, a reduction of high-density lipoprotein (HDL) cholesterol levels has been described, as well as an increase in the ratio of LDL/HDL cholesterol compared to saturated fats (*7*, *8*). Special attention is drawn to the potential adverse effects of TFA consumption in the metabolism of essential fatty acids. TFA may impair elongation and desaturation of essential *n*−6 fatty acids and affects human growth and development (*9*).

Although reported data on *trans*-fatty acid intake vary widely from one country to another depending on food habits and food manufacturing techniques (*10*), the food elaborated with hydrogenated fats, such as margarines, cookies, and other industrially baked products, have been pointed out as the main source of TFA in the diet (*11*−*14*). Consumption of cookies and bakery has increased 6.8% in the past three years and represents 3.9% of the total food consumption in Spain (*15*). Due to the consumer concern over *trans*-fatty acid content in food, a trend to reduce its presence in shortenings and margarines has been detected in recent years in several European countries (*16*); however, this trend has not been observed in Spain (*17*). Therefore, processed foods such as bakery products and cookies may contain a complex mixture of fats from different sources: animal fat and vegetable oils or fats, besides hydrogenated or partially hydrogenated oils. This fact is not always clearly reflected on the food label (*18*). Recently the convenience of including TFA content on the standard food labels has been recognized by the U.S. Food and Drug Administration, which is proposing to include it in the amount and daily percent value (%DV) declared for saturated fatty acids (SFA) (*19*).

The objectives of this paper were to determine the fatty acid composition of products usually consumed by the Spanish population that may significantly contribute to TFA intake. Multivariate analysis, which has been successfully applied for characterization and discrimination of foods, such as coffee, on the basis of their acid profile (*20*), has been applied to characterize the main source of fat employed in manufacturing these products.

MATERIALS AND METHODS

Samples. A total of 39 samples of cookies and bakery products were sampled among commercially available products, during 1999−2000. These samples were classified into 13 different categories as follows:

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Table 1. Mean Fatty Acid Contents (Percent w/w of the Total Fatty Acids) and Standard Deviation for the Eight Categories of Cookies Analyzed

fatty acid	chocolate coated, $n = 4$	chocolate filled, $n = 4$	butter cookies, $n = 3$	Marie wheat, $n = 5$	wholemeal cracker, $n = 3$	cracker, $n = 2$	tea pastry, $n = 2$	chocolate bars with cracker, $n = 2$
$C_{8:0}$	0.44 ± 0.76	1.05 ± 1.02	0.7 ± 0.00	1.49 ± 1.85	0.03 ± 0.02	1.09 ± 1.51	0.76 ± 0.99	1.31 ± 1.74
$C_{10:0}$	0.47 ± 0.66	0.97 ± 0.87	1.46 ± 0.02	1.27 ± 1.47	0.03 ± 0.01	0.98 ± 1.22	0.61 ± 0.73	1.39 ± 1.70
$C_{12:0}$	5.33 ± 10.11	8.13 ± 8.27	2.99 ± 0.00	10.92 ± 13.74	0.2 ± 0.14	8.4 ± 11.53	5.71 ± 5.56	18.85 ± 26.34
$C_{14:0}$	2.69 ± 3.36	4.23 ± 4.13	6.74 ± 0.03	4.5 ± 3.99	0.36 ± 0.18	3.43 ± 3.12	2.67 ± 1.75	5.87 ± 7.52
$C_{16:0}$	26.49 ± 3.38	23.95 ± 5.67	40.3 ± 0.22	22.56 ± 8.45	12.58 ± 0.23	26.88 ± 14.28	29.82 ± 6.31	41.86 ± 43.31
$C_{18:0}$	21.76 ± 7.92	14.13 ± 10.35	12.01 ± 0.03	9.63 ± 6.73	8.49 ± 0.46	7.63 ± 3.87	10.01 ± 6.37	13.16 ± 4.89
$C_{20:0}$	0.51 ± 0.18	0.43 ± 0.15	0.21 ± 0.01	0.3 ± 0.08	0.45 ± 0.05	0.36 ± 0.18	0.14 ± 0.14	0.31 ± 0.04
$C_{22:0}$	0.19 ± 0.12	0.11 ± 0.1	0.11 ± 0.00	0.08 ± 0.06	0.62 ± 0.06	0.11 ± 0.02	0.09 ± 0.01	0.06 ± 0.04
SFA[a]	57.88 ± 10.94	53.00 ± 12.61	64.52 ± 0.25	50.75 ± 16.54	22.45 ± 0.11	48.88 ± 6.77	49.80 ± 8.80	82.81 ± 1.12
$C_{16:1}$	0.42 ± 0.35	0.53 ± 0.6	0.63 ± 0.05	0.95 ± 1.15	0.08 ± 0.03	0.15 ± 0.13	1.25 ± 1.44	0.13 ± 0.01
$C_{18:1}$	32.14 ± 8.8	34.51 ± 8.63	27.74 ± 0.34	36.63 ± 11.13	27.76 ± 1.86	35.02 ± 10.03	39.07 ± 8.65	11.23 ± 1.73
MUFA[a]	32.56 ± 8.90	35.04 ± 9.15	28.37 ± 0.29	37.58 ± 12.15	27.84 ± 1.84	35.17 ± 10.16	40.32 ± 10.09	11.37 ± 1.74
$C_{18:2}$	8.57 ± 2.63	9.14 ± 1.99	5.32 ± 0.18	10.05 ± 3.54	13.53 ± 0.45	8.45 ± 5.37	8.52 ± 0.93	2.09 ± 1.36
$C_{18:3}$	0.62 ± 0.58	0.3 ± 0.11	0.38 ± 0.00	0.52 ± 0.33	0.32 ± 0.09	0.29 ± 0.28	0.35 ± 0.22	0.15 ± 0.06
PUFA[a]	9.18 ± 3.18	9.44 ± 2.10	5.70 ± 0.19	10.56 ± 3.84	13.86 ± 0.37	8.74 ± 5.65	8.87 ± 0.71	2.23 ± 1.42
$C_{16:1t}$	tr[b]	tr	0.26 ± 0.34	0.05 ± 0.12	tr	0.01 ± 0.02	0.14 ± 0.12	tr
$C_{18:1t}$	0.09 ± 0.08	1.93 ± 2.94	0.57 ± 0.42	0.69 ± 1.23	31.64 ± 2.06	6.29 ± 8.54	0.22 ± 0.03	0.25 ± 0.07
$C_{18:2t}$	0.22 ± 0.13	0.55 ± 0.51	0.35 ± 0.01	0.32 ± 0.16	3.79 ± 0.61	0.87 ± 0.54	0.44 ± 0.36	0.25 ± 0.07
$C_{18:3t}$	0.07 ± 0.12	0.04 ± 0.09	0.24 ± 0.31	0.03 ± 0.04	0.12 ± 0.00	0.04 ± 0.05	0.21 ± 0.3	0.03 ± 0.04
TFA[a]	0.38 ± 0.26	2.53 ± 3.48	1.41 ± 0.23	1.10 ± 1.34	35.54 ± 1.48	7.21 ± 9.05	1.01 ± 0.57	3.59 ± 1.44
fat (%)	17.92 ± 7.46	31.82 ± 9.90	27.09 ± 1.99	14.83 ± 3.80	19.70 ± 0.45	22.23 ± 12.52	23.29 ± 3.24	16.46 ± 5.59

[a] SFA = $C_8 + C_{10} + C_{12} + C_{14} + C_{16} + C_{18} + C_{20} + C_{22}$; MUFA = $C_{16:1} + C_{18:1}$; PUFA = $C_{18:2c9c12} + C_{18:3}$; TFA = $C_{16:1t} + C_{18:1t} + C_{18:2t} + C_{18:3t}$. [b] Trace.

croissants (4), sponge cakes (2), chocolate-filled Swiss rolls (2), Swiss roll (1), chocolate cakes (5), chocolate-coated cookies (4), chocolate-filled cookies (4), butter cookies (2), Maria wheat cookies (5), wholemeal crackers (3), crackers (2), tea pastries (2), and chocolate bars with cracker (2). Samples were provided by a distributor and corresponded to different brand names. The selection of samples was based on confidential information from the distributor on the most frequently sold products and brand names. Each sample was analyzed in duplicate.

Fat Extraction. Fat was extracted following a modification of the procedure proposed by Folch et al. (*21*). Two grams of the homogenized sample was extracted twice with 30 mL of a mixture of methylene chloride/methanol (2:1 v/v) and shaken for 20 min in an Erlenmeyer flask with a magnetic stirrer. After filtering, the liquid phases were mixed and washed with a 0.58% (w/v) sodium chloride solution. After phase separation, the methylene chloride layer was filtered and dried with sodium sulfate and the solvent was evaporated in a rotary vacuum pump. The residue was transferred to a 10 mL glass vial and maintained at −18 °C until analyses were carried out.

Fatty Acids Analyses. Fatty acid methyl esters (FAME) were prepared from the extracted fat following Slover and Lanza's method (*22*). Approximately 200 mg of fat was saponified with 4 mL of sodium methoxide in methanol (0.5 N) for 10 min in a boiling water bath. The solution was then cooled at room temperature, and 5 mL of a boron trifluoride/methanol complex (14%) was added. Before transesterification, 200 μL of heptadecanoic acid (5 mg/mL) (Merck, Darmstadt, Germany) was added as internal standard. The solution was then heated for 10 min in a boiling water bath (100 °C). After cooling, 2 mL of hexane was added, and the mixture was heated for another 2 min and cooled at room temperature; 2.5 mL of saturated sodium chloride was then added. The mixture was shaken vigorously and, after phase separation, the organic layer was suctioned with a Pasteur pipet and transferred to a 2 mL vial containing 1 mm of anhydrous sodium sulfate. This vial was sealed and stored at −18 °C until the chromatographic analyses. FAME were analyzed by gas−liquid chromatography (GLC). One microliter of methylated sample was injected into the gas chromatograph (GC), a Hewlett-Packard 6890 equipped with a 60 m SP 2340 capillary column (0.25 mm i.d. and 0.20 μm film thickness) (Palo Alto, CA). The oven was programmed as follows: 160 °C for 13 min, raised to 190 °C at 1.5 °C/min. The final oven temperature was maintained for 5 min. The injector and detector temperatures were 225 and 250 °C, respectively. Hydrogen was used as carrier gas (1.3 mL/min, constant flow). FAME were identified by comparing their retention times using appropriate pure standards (Merck).

All solvents used were of reagent grade from Panreac (Sant Cugat del Valles, Barcelona, Spain), and all reactives were of analytical grade. Results are expressed as percent (w/w) of the total fatty acids detected.

Statistical Analysis. Analysis of variance (ANOVA) was carried out for the SFA, monounsaturated fatty acids (MUFA), polyunsaturated fatty acids (PUFA), and TFA between the two groups of samples. Factor analysis method was used as multivariate analysis method. Principal component analysis (PCA) with varimax rotation was performed on the mean data matrix to classify samples according to their fatty acid profile, without making prior assumptions on food type. The varimax rotation of the normalized factor loadings (raw factor loadings divided by the square roots of the respective communalities) is aimed at maximizing the variances of the squared normalized factor loadings across variables for each factor, which is equivalent to maximizing the variances in the columns of the matrix of the squared normalized factor loadings. Statistical analyses were performed using a Statistica software package (*23*).

RESULTS AND DISCUSSION

Mean total fat contents and mean *cis*- and *trans*-fatty acid isomer contents for the 13 categories of Spanish cookies and bakery products analyzed are shown in **Tables 1** and **2**. Total fat contents ranged from 14.8 to 31.8% in cookies and from 15.8 to 31.2% in bakery products, the mean value being slightly higher for bakery products than for cookies (**Table 3**). Similar fat contents, oscillating between 9.4 and 31% in cookies and between 5.0 and 35.0% in cakes and bakery products, have been reported previously in a selection of European products (*18*).

Table 2. Mean Fatty Acid Contents (Percent w/w of the Total Fatty Acids) and Standard Deviation for the Five Categories of Bakery Products Analyzed

fatty acid	croissant, $n = 4$	sponge cakes, $n = 2$	Swiss roll/chocolate, $n = 2$	Swiss roll, $n = 1$	chocolate cake, $n = 5$
$C_{8:0}$	0.06 ± 0.03	0.12 ± 0.12	1.3 ± 1.82	1.21	0.4 ± 0.42
$C_{10:0}$	0.08 ± 0.05	0.05 ± 0.01	1.14 ± 1.56	0.98	0.4 ± 0.42
$C_{12:0}$	0.58 ± 0.21	0.14 ± 0.01	14.12 ± 19.73	8.01	0.55 ± 0.01
$C_{14:0}$	1.01 ± 0.14	1.19 ± 1.19	15.85 ± 10.7	2.92	2.33 ± 0.18
$C_{16:0}$	32.77 ± 0.67	11.32 ± 0.08	32.2 ± 2.40	7.97	29.52 ± 0.80
$C_{18:0}$	9.44 ± 0.56	4.54 ± 0.00	9.41 ± 6.96	7.54	7.59 ± 0.19
$C_{20:0}$	0.43 ± 0.14	1.42 ± 0.01	0.13 ± 0.04	0.46	1.09 ± 0.11
$C_{22:0}$	0.16 ± 0.04	0.41 ± 0.02	0.16 ± 0.05	0.34	0.37 ± 0.03
SFA[a]	44.52 ± 0.75	19.19 ± 0.98	74.31 ± 16.96	29.42	42.24 ± 0.21
$C_{16:1}$	0.58 ± 0.57	0.31 ± 0.01	0.18 ± 0.02	0.26	0.77 ± 0.05
$C_{18:1}$	38.6 ± 1.74	22.74 ± 0.32	10.77 ± 5.28	42.16	42.07 ± 0.75
MUFA[a]	39.18 ± 1.79	23.04 ± 0.32	10.95 ± 5.26	42.43	42.84 ± 0.74
$C_{18:2}$	8.69 ± 1.34	49.35 ± 0.37	13.17 ± 12.8	23.70	9.79 ± 0.64
$C_{18:3}$	1.67 ± 1.64	6.32 ± 0.21	0.13 ± 0.04	4.30	1.02 ± 0.08
PUFA[a]	10.36 ± 2.97	55.66 ± 0.66	13.30 ± 12.84	28.01	10.82 ± 0.67
$C_{16:1t}$	tr[b]	tr	tr	tr	0.071 ± 0.09
$C_{18:1t}$	5.16 ± 0.71	nd[c]	1.3 ± 0.21	0.08	1.31 ± 0.02
$C_{18:2t}$	0.46 ± 0.06	0.66 ± 0.04	0.13 ± 0.08	0.07	0.59 ± 0.05
$C_{18:3t}$	0.32 ± 0.08	1.44 ± 0.04	0.01 ± 0.01	nd	1.49 ± 1.38
TFA[a]	5.94 ± 0.69	2.10 ± 0.08	1.44 ± 1.14	0.15	4.10 ± 1.50
fat (%)	24.56 ± 3.04	31.17 ± 3.21	15.80 ± 0.89	20.82	26.63 ± 3.97

[a] SFA = $C_8 + C_{10} + C_{12} + C_{14} + C_{16} + C_{18} + C_{20} + C_{22}$; MUFA = $C_{16:1} + C_{18:1}$; PUFA = $C_{18:2} + C_{18:3}$; TFA = $C_{16:1t} + C_{18:1t} + C_{18:2t} + C_{18:3t}$. [b] Trace. [c] Not detected.

Table 3. Mean and Standard Deviation for the Nutritional Fractions, Total Fat Content, and Nutritional Indices for the Cookies and Bakery Products Analyzed[a]

		nutritional fractions				nutritional indexes	
	fat	SFA	MUFA	PUFA	TFA	[SFA]/[PUFA]	[SFA + TFA]/[MUFA + PUFA]
bakery products, $n = 14$	24.7 (5.39)	43.27(16.60)a	34.38 (12.12)a	18.68 (16.78)a	3.68 (2.16)a	0.67 (0.97)a	1.45 (1.07)a
cookies, $n = 25$	21.5 (8.13)	52.88 (17.20)b	32.03 (10.39)a	9.03 (3.80)b	6.06 (11.50)a	0.22 (0.18)b	0.78 (0.42)b
mean	22.7 (7.4)	49.43 (17.04)	32.87(10.94)	12.48 (11.29)	5.20 (9.30)	0.38 (0.62)	1.02 (0.78)

[a] Means within a column not sharing the same letter are significantly different ($p < 0.05$).

Fatty acids were classified into four nutritional fractions: SFA; MUFA; PUFA; and TFA. The mean values for the nutritional fractions in cookies and bakery products are shown in **Table 3**. In relation to the fatty acid profile, SFA (mean = 49.43%, SD = 17.40) predominated in the majority of the products. The main SFA found were $C_{16:0}$ (mean = 26.77, SD = 11.88) followed by $C_{18:0}$ (mean = 10.92, SD = 6.40), $C_{12:0}$ (mean = 5.78, SD = 9.73), $C_{14:0}$ (mean = 3.80, SD = 4.48), $C_{10:0}$ (mean = 0.72, SD = 0.89), and $C_{8:0}$ (mean = 0.72, SD = 1.01). Other SFA occurred in smaller amounts (<1%). MUFA accounted for 32.87% (SD = 10.94%), and oleic acid was the main MUFA found (mean = 32.33%). Low contents of $C_{16:0}$ were also found in all samples (<1.25%). PUFA (mean = 12.48%, SD = 11.29) varied widely among food products, from very low contents in chocolate bars with crackers (2.23%) to very high contents in sponge cakes (55.66%), as a clear indicator of the wide variety of fats and oils that can be employed in the manufacturing of these products.

The proportion of TFA ranged from 0.38 to 35.54% in cookies (mean = 6.06, SD = 11.50) and from 0.15 to 15.94% (mean = 5.20, SD = 9.30) in bakery products. Previously published results for TFA in bakery products (mean = 6.5%, SD = 4.2) (*11*) and cookies (TFA mean = 12.63%) (*18*) in Spain are slightly higher than those found in our samples. This would indicate that the tendency to reduce the TFA content in foods, already seen in European margarines (*16*), is also applicable to cookies and bakery products. The great variability in TFA content within a food category should also be noted, especially for the two brands of crackers analyzed (0.81 and 13.60% TFA contents, respectively). This variability is due to the different recipes uses by the producers, which can be based on a low *trans*-fat or high *trans*-fat.

$C_{18:1t}$ was the predominant *trans*-isomer found (mean = 4.04, SD = 8.46), ranging from 0.09 to 31.64% in cookies and between traces and 5.16% in bakery products. $C_{18:2t}$, which is known to adversely affect the metabolism of children (*9*), was detected in the cookies and cake samples analyzed at <1% [except for the wholemeal cracker, for which higher levels were detected (3.79%)]. Improvements in the hydrogenation process have reduced the amount of this isomer in margarines and hydrogenated fats. $C_{16:1t}$ and $C_{18:3t}$ were detected in the samples at low levels (<0.5%). The presence of $C_{18:3t}$ is related to physically refined or deodorized oils (*25*).

ANOVA was performed to find significant differences between bakery products and cookies with regard to the fatty acid profile (SFA, MUFA, PUFA, and TFA). No significant differences ($p < 0.05$) were found between groups for the MUFA and TFA contents; however, significant differences were

Table 4. Eigen Values and Varimax Rotated Factor Loadings (Eigenvectors) for the Fatty Acids Analyzed (Boldface Loadings Are $p > 0.700$)

	factor 1	factor 2	factor 3	factor 4
variance explained (%)	31.6	18.4	15.5	10.1
eigenvalue	5.062075	2.943819	2.475657	1.618826
eigenvectors				
$C_{8:0}$	**−0.901819**	0.146656	0.133105	0.029183
$C_{10:0}$	**−0.889342**	0.191613	0.218301	0.029954
$C_{12:0}$	**−0.887803**	0.127166	0.109333	0.156520
$C_{14:0}$	**−0.700787**	0.202280	0.099971	0.060543
$C_{16:0}$	0.089374	0.306873	0.519658	−0.109170
$C_{16:1t}$	0.208308	0.108531	0.275370	**−0.858533**
$C_{16:1}$	0.598677	0.362374	0.159212	0.005600
$C_{18:0}$	0.297715	0.246735	0.218709	0.448562
$C_{18:1t}$	0.062351	**−0.964462**	0.052243	0.137903
$C_{18:1}$	**0.743339**	0.158522	0.113531	−0.182799
$C_{18:2t}$	0.085911	**−0.975487**	−0.020670	0.005775
$C_{18:2}$	0.117742	−0.058182	**−0.944622**	−0.126209
$C_{18:3t}$	0.119232	0.023820	−0.226871	**−0.846372**
$C_{20:0}$	0.301963	−0.020701	−0.532568	−0.641660
$C_{18:3}$	0.136108	0.120204	**−0.908356**	−0.191046
$C_{22:0}$	0.271611	**−0.751675**	−0.325930	−0.380249

found for the SFA and PUFA contents (**Table 3**). This could be related to the fact that, in comparison to cookies, bakery products are food products that may be elaborated with both hard and soft fats, as indicated by the different proportions of SFA and PUFA found in the these products. For example, sponge cakes were low in SFA and high in PUFA, which could indicate that part of the fat they contain comes from a vegetable oil, as confirmed by the TFA detected, $C_{18:2t}$ and $C_{18:3t}$, which are known to be formed during the refining process of vegetable oils. On the contrary, croissants were high in SFA and TFA and low in PUFA, indicating a composition probably based on a mixture of saturated vegetable fat and partially hydrogenated oils.

The nutritional value of these products in relation to their fat composition could be deduced using the traditional indices, which consider SFA and PUFA [PUFA/SFA], or those used in recent years, which have included also TFA and MUFA, [PUFA + MUFA]/[SFA + TFA] (**Table 3**). Significant differences between cookies and bakery products were found for these indices. According to the calculated values (0.67 and 1.45 for bakery products and 0.22 and 0.78 for cookies), it seems that bakery products would be preferable to cookies, attending to their possible health effects.

In an attempt to classify the samples according to the main source of fat employed in their manufacture, multivariate analyses were carried out. Factor analysis was performed on the 39 individual samples and the 16 variables measured (individual fatty acid). The main criteria used to decide how many factors to retain was the amount of variance explained and the interpretability of the results. Four factors with eigenvalues greater than unity were retained, explaining 75% of the total variance. Factors were subjected to varimax rotation to obtain a clear pattern of loadings, that is, factors that are somehow clearly marked by high loadings for some variables and low loadings for others. Results are shown in **Table 4**.

Factor 1 (F1) explained 32% of the variation and was marked by high negative loadings for $C_{8:0}$−$C_{14:}$ and a high positive loading for $C_{18:1}$, which could be interpreted as the addition of either vegetable fat/dairy fat or animal fat to elaborate the products. Vegetable fat, mainly from coconut oil or palm kernel oil, is rich in C_{12}−C_{14} fatty acids. $C_{8:0}$−C_{10} are related to dairy fats, such as butter, where they represent 4−5%. $C_{18:1}$ is a widely distributed fatty acid, which can be found in high amounts in animal fats and also in olive oil, but due to its high price it is not used as the source of fat to manufacture these products. Factor 2 (F2), which still explained 17% of the variation, was marked by high negative loadings for $C_{18:1t}$ and $C_{18:2t}$, which could be related to the presence of partially hydrogenated oil, and specifically to sunflower oil, in which $C_{22:0}$ is a representative fatty acid. Factor 3 (F3), which accounted for 14% of the total variance, was marked by high loadings on variables clearly related to vegetable oils, such as $C_{18:2}$ and $C_{18:3}$. Minor TFA ($C_{18:3t}$ and $C_{16:1t}$) were related to factor 4 (F4).

To facilitate the interpretation of these results, the obtained scores were plotted by selecting the first three factors as axes (**Figures 1** and **2**). As can be seen in **Figure 1**, samples were

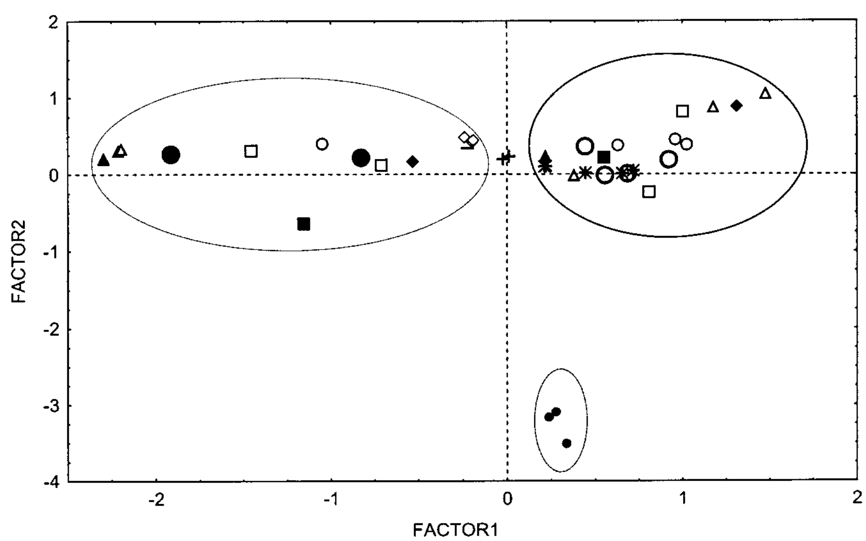

Figure 1. Plot values of the first two factors (factor 1 vs factor 2) according to the type of food product. Group 1 (- – -): samples based on hydrogenated vegetable oils. Group 2 (—): samples based on animal fat. Group 3 (- - -): samples based on vegetable fat. Key: (○) chocolate-coated cookies; (□) chocolate-filled cookies; (◇) butter cookies; (△) Marie wheat cookies; (●) wholemeal cracker; (■) cracker; (◆) tea pastry; (▲) chocolate bars with cracker; (○) croissant; (+) sponge cakes; (●) Swiss roll filled with chocolate; (–) Swiss roll; (∗) chocolate cakes.

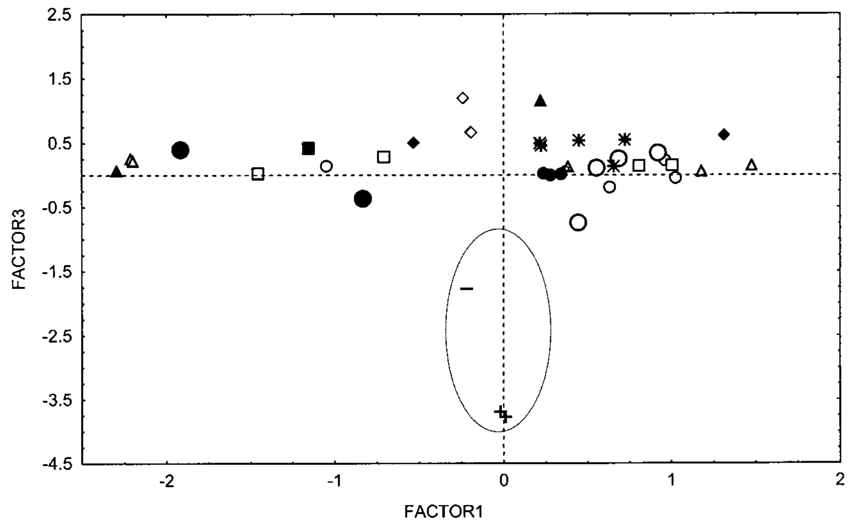

Figure 2. Plot values of factor 1 vs factor3 according to the type of food product. Group 4 (···): samples based on vegetable oil. Key: (○) chocolate-coated cookies; (□) chocolate-filled cookies; (◇) butter cookies; (△) Marie wheat cookies; (●) wholemeal cracker; (■) cracker; (◆) tea pastry; (▲) chocolate bars with cracker; (○) croissant; (+) sponge cakes; (●) Swiss roll filled with chocolate; (–) Swiss roll; (∗) chocolate cakes.

grouped according to the type of fat employed, being independent of the category of food product; besides, no clear clustering of cookies or bakery products could be observed. The samples with positive F1 values are those in which animal fat is mainly present, and negative values are obtained for samples in which vegetable or dairy fat is used as the main source of fat. For F2, positive values were obtained for samples with low contents of TFA and negative values for samples containing important amounts of partially hydrogenated fats (high in TFA). Taking into account that the fat incorporated can be a mixture of different fats, including vegetable oil and hydrogenated or partially hydrogenated oils, as well as vegetable or animal fat, samples have been grouped according to their location in the plot in relation to the main source of fat incorporated: group 1, samples probably based on hydrogenated vegetable oils, with high contents of TFA and low in SAT (this group includes the three samples of wholemeal cracker analyzed); group 2, samples that incorporate mainly fat from an animal origin (this group includes most of the samples of cookies and bakery products analyzed); group 3, samples probably based on vegetable and/or dairy fat.

When F1 is plotted versus F3 (**Figure 2**), three samples of bakery products seem to distinguish themselves from the rest of the group along the negative side of F3: the two samples of sponge cake and the Swiss roll sample. These are products in which vegetable oil has been probably used as a source of fat, and so, higher amounts of PUFA were found (group 4). It should be noted that different brands of the same category of product are located at different sides of the diagram, which confirms that, for some cookies and bakery products (croissant, chocolate bars with cereal, cracker, and tea pastry), each producer elaborates the same category of product using a different source of fat (*18*, *24*).

According to the results obtained, we could state that hydrogenated fats are not used as the main source of fat for manufacturing cookies and bakery products in Spain. However, the *trans*-fatty acid content may vary within the same category of products depending on the brand. Even though the levels of TFA detected in this study are slightly lower than others previously published for bakery products and cookies (*11*, *18*), we have still found high levels of TFA in some brands. If we consider that cookies and bakery products are one of the main sources of TFA in the diet (*10*, *14*) and that consumption of these types of products is increasing (*15*), producers should make an effort to replace partially hydrogenated oils with healthier fats (unhydrogenated oils). Due to the adverse effects of TFA on health, it is advisable to include the *trans*-fatty acid contents on the nutritional labeling.

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