Fatty Acid Composition of Caprine Milk: Major, Branched-Chain, and *Trans* Fatty Acids

ABSTRACT

The fatty acid composition of caprine milk fat was studied using capillary gas chromatography. Milk was obtained from five goat herds belonging to different breeders in the Murcia region (Spain) and collected monthly (from November to May). The results showed significant differences among herds mainly in long-chain fatty acids ($C_{16:0}$, $C_{18:0}$, and $C_{18:2}$). There were five branched-chain fatty acids (iso- and anteiso-C_{15:0}, *iso*- and *anteiso*-C_{17:0}, and *iso*-C_{16:0}) with >0.1% of the total fatty acid methyl esters and another 31 (the most monomethylated) with <0.1%, including 4-ethyloctanoate, which is implicated in goat-like flavors. To study the content of trans unsaturated fatty acids, the fatty acid methyl esters were previously fractionated by AgNO₃-thin layer chromatography. The mean contents of *trans*-C_{16:1} and *trans*-C_{18:1} were 0.16 and 2.12%, respectively. The distribution profile of *trans*-C₁₈₋₁ was also studied.

(**Key words**: caprine milk fat, gas chromatography, fatty acid composition)

Abbreviation key: BCFA = branched-chain fatty acids, **FAME** = fatty acid methyl esters, **GC** = gas chromatography, **MSD** = mass spectrometer detector, **TG** = triglycerides.

INTRODUCTION

Although the world production of caprine milk is relatively minor compared with that of bovine milk $(1.8\% \text{ vs. } 86.9\% \text{ of the total milk production, respec$ $tively})$ (10), consumption (largely in the form of cheese) is considerable in some Mediterranean coun-

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tries. Thus, in Greece and Spain, the consumption of caprine cheese is 23 and 7%, respectively, of the consumption of cheese made from bovine milk (21, 22). Moreover, in Spain, almost 50% of cheese production

uses a mixture of bovine, ovine, and caprine milks. In recent years, the intake of *trans* fatty acids has been associated with the risk of coronary heart disease (12). The main source of *trans* unsaturated fatty acids consumed daily by humans is partially hydrogenated vegetable fats and oils, although these compounds also occur naturally in caprine milk, as in milk of ewes and cows. Data on trans unsaturated fatty acid contents of caprine milk (1) and cheese (22) are very scarce. In gas chromatography (GC) analysis, even using high polarity, 100-m capillary columns (with cyanopropyl siloxane as the stationary phase), some trans unsaturated fatty acids coelute with saturated fatty acids (*trans*-C_{16:1} with *iso*- and anteiso- $C_{17:0}$) or with their cis isomers (trans- $C_{18:1}$ with cis-C_{18:1}). Therefore, to obtain reliable data, the methyl esters should be first fractionated by TLC or HPLC.

Other minor components of milk fat are branchedchain fatty acids (**BCFA**). The main point of quantifying these components is that volatile BCFA lend characteristic flavors to many dairy foods (23). The amount of these compounds contained in a cheese is largely dependent on the composition of the milk fat substrate (8). In caprine milk, Massart-Leen et al. (14) identified and quantified numerous BCFA (all having more than 11 carbon atoms), and, subsequently, Ha and Lindsay (7) identified over 20 volatile BCFA in caprine cheese. By combining GC with a 100-m capillary column and mass spectrometry, these components can be separated and identified without any previous separation technique.

The aim of this work was to determine the range of variation in composition of the major, branched-chain, and *trans* fatty acids of caprine milk fat from five herds belonging to different breeders and collected during 7 consecutive mo.

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MATERIALS AND METHODS

Samples

Thirty-five samples of raw caprine milk were collected monthly (from November to May) from five herds belonging to five different breeders in the Murcia region (Spain) and were used to analyze the content of major fatty acids. Refrigerated raw milk samples (250 ml) were tempered at 20°C for 20 min, then filtered, and centrifuged (Beckman J2 MC; Beckman Instruments, Fullerton, CA) at $6000 \times g$ for 30 min at 20°C prior to fat separation. The tubes containing the centrifuged milk were placed on ice until the milk fat had solidified, at which point fat was removed and treated with anhydrous sodium sulfate. The mixture was extracted four times with diethyl ether, and the total organic fraction was filtered, exposed to a stream of N2, and dried by evaporation under low pressure at 4°C. The extracted fat residue was stored frozen at -20°C until analysis. To study the content and the composition of trans monoenoic and minor odd- and branched-chain fatty acids, all of the caprine milk fat samples from each herd were combined in equal volumes to obtain five mixture samples.

Preparation of Fatty Acid Methyl Esters

For preparation of fatty acid methyl esters (**FAME**) of milk fat, 0.1 g of fat was dissolved in 1 ml of hexane, and 0.05 ml of 2N potassium hydroxide in methanol was added as described by Christopherson and Glass (4).

Fractioning by AgNO₃-TLC of FAME

The FAME were fractionated according to the number and geometry of double bounds by TLC following the Precht and Molkentin (17) procedure slightly modified. The TLC glass plates (20×20 cm) with silica gel (0.25 mm) (Merck, Darmstadt, Germany) were incubated with 20% aqueous solution of AgNO₃ (Panreac, Barcelona, Spain) for 16 h, were partially air dried, and were activated at 120°C for 30 min. A 100- μ l solution of FAME (80 mg/ml) was applied to the activated TLC glass plate in a narrow band. The plate was developed twice in a saturated chamber in hexane and diethyl ether (9:1, vol/vol) with 15-cm migration. At the end of chromatographic runs, the plates were air dried and spraved with a 0.20% ethanol solution of 2',7'-dichlorofluorescein, and the bands were visualized under UV light. The bands corresponding to the saturated and *trans* monoenoic fatty acid methyl esters, which were previously identified by a mixture of stearic fatty acid methyl ester ($C_{18:0}$) and elaidic fatty acid methyl ester (*trans*-9- $C_{18:1}$) (Sigma Chemical, St. Louis, MO) running in AgNO₃-TLC, were scraped into a flask. The FAME were extracted with 80 ml of diethyl ether in four extractions, and the solvent was evaporated in a rotovapor and stream of nitrogen. The residue was dissolved in 200 μ l of heptane and used for GC analysis.

GC Analysis of Total and trans Fatty Acids

Some of the FAME solution (0.2 μ l) was injected into an Autosystem model GC (Perkin-Elmer Co., Beaconsfield, United Kingdom) equipped with a flame ionization detector. Analysis were performed with a CP Sil 88 column (100 m \times 0.25 mm i.d.) containing 100% cyanopropyl siloxane as the stationary phase with a film thickness 0.20 μ m (Chrompack, Middelburg, The Netherlands). The initial temperature of 70°C was maintained for 3 min, then raised to 175°C at a rate of 9°C/min, maintained for 27.5 min, and then increased to 210°C at a rate of 1.3°C/min for 10 min. The split ratio was 1:50, and hydrogen was the carrier gas with a head pressure of 1.2 kg/m². The injector and detector temperatures were 250°C. For quantitative determinations of FAME of AgNO3-TLC fractions, the column and chromatographic conditions were the same as total FAME. Duplicate analyses were performed for each sample.

For quantitative studies of total FAME, an anhydrous milk fat with a certified fatty acid composition (reference material CRM-164), obtained from Commission of the European Communities, Brussels, Belgium, was used to determine the response factors. To calculate the total content of *trans*-C_{18:1} isomers, the ratio of C_{18:0} to total *trans*-C_{18:1} was determined in the saturated plus *trans* monoenoic AgNO₃-TLC fraction and was related to the C_{18:0} content of total FAME. To calculate the total content of *trans*-C_{16:1} isomers, the ratio of total *trans*-C_{16:1} to *trans*-10-C_{18:1} plus *trans*-11-C_{18:1} was determined in the *trans* monoenoic AgNO₃-TLC fraction and related to the ratio of C_{18:0} to *trans*-10-C_{18:1} plus *trans*-11-C_{18:1} in the saturated plus *trans* monoenoic AgNO₃-TLC fraction and to the C_{18:0} content of total FAME.

GC Mass Spectrometry

Analyses were performed on a Hewlett-Packard 5890 gas chromatograph coupled with a 5972 mass

spectrometer detector (MSD) (Hewlett-Packard, Palo Alto, CA). Manual tuning of the MSD with perfluorotributylamine was used to adjust relative abundance for m/z 69, m/z 219, and m/z 502. The MSD was run in the scan mode (m/z range 33 to 250 with a threshold of 100 and sampling of 3 scans/s). Ultrapure helium was passed through moisture and oxygen traps and was used as carrier gas. Column and GC operating conditions were the same as the total FAME interface line to MSD at 280°C. The electron energy and multiplier voltage of the quadropole were 70 eV and 1670 V. Data were recorded and analyzed with the HP G1034C MS Chemstation software (XX). The fatty acids were identified by comparing their mass-spectral data to the mass-spectral data base in the library Wiley 138, incorporated into the MS Chemstation software installed in a Vectra 486/33 VL personal computer (Hewlett-Packard, Palo Alto, CA).

Statistical Analysis

The results were analyzed according to the BMDP (2) software package. Sources of variation in variance analysis were herd and period of year.

RESULTS AND DISCUSSION

For the given chromatographic conditions, 64 fatty acids (54 saturated, 7 monounsaturated, and 3 polyunsaturated) were separated and quantified. The total content of *trans* fatty acids and the proportion of $C_{18:1}$ *trans* isomers was also quantified. Thirty-six fatty acids were detected in very small amounts, together representing 0.77% of the total fatty acids. Therefore, the influences of time of year in which the samples were collected (monthly from November to May) and herd were only studied for the fatty acids that accounted individually for >0.1% of the total fatty acids.

Major Fatty Acids in Caprine Milk

The composition of fatty acids in caprine milk fat did not differ (P > 0.05) with respect to the time of year of sampling. This result is in agreement with the findings of Fontecha et al. (5) for triglyceride (**TG**) composition using the same samples. Others (3, 16) have reported changes, mainly in the proportions of long-chain fatty acids, related to stage of lactation and feeding. Although kidding occurs in autumn in the Murcia region, the possible differences in milk yield, diet, and stage of lactation from one herd to another were not controlled.

The five most important fatty acids in quantitative terms (C_{16:0}, C_{18:1}, C_{10:0}, C_{14:0}, and C_{18:0}) accounted for >75% of total fatty acids (Table 1). Individually, the percentages of major fatty acids were within the range of variation of those reported by other authors. Results showed the characteristic fatty acid pattern of caprine milk; values to caprylic (2.7%) and capric (9.9%) acids were higher than were those of bovine milk. The C_{12:0}:C_{10:0} ratio, proposed by Ramos and Juárez (18) to detect the authenticity of caprine milk, obtained in the present work (0.50 ± 0.04) was similar to the values reported by Iverson and Sheppard (11) in caprine milk (0.46 ± 0.04) and by Wolf (22) in caprine cheese (0.56 ± 0.05) and very different from the values reported for bovine milk by the same authors (1.16 and 1.14, respectively).

With regard to the influence of the herd (see Table 1), the largest differences (a range of about 10% between extreme values) in short-chain fatty acids were found for acids $C_{6:0}$, $C_{8:0}$, and $C_{10:0}$ (P < 0.05). The content of these acids was higher for herd 1 than for herd 5, and the rest were intermediate. The C_4 content for herds 1 and 5 was lower (P < 0.05) than for the rest. All of these differences were very small in absolute quantitative terms. The largest differences in long-chain fatty acids were found in the percentages of $C_{18:0}$, $C_{18:2}$, and $C_{18:2}$ conj (P < 0.01) and above all in $C_{16:0}$ content (P < 0.001); differences of 4 units existed between extremes. High variability of milk fatty acid composition among different animals was also observed by Sauvant and Morant-Fehr (19) and Massart-Leen et al. (14).

A study of the TG composition of the same samples (5) similarly revealed a significant herd effect, which was especially important quantitatively in the longchain TG (C_{48} to C_{54}); the lowest values were found for herd 1. Although in the present work there were not significant differences in the total $C_{18:1}$ contents of milk from different herds, a high correlation (r = 0.78) of the amounts of total $C_{18:1}$ to C_{54} was found. This correlation was similar to that reported for bovine milk fat by Precht and Molkentin (17) despite the differences in the C_{54} contents of caprine and bovine milks (5).

Branched-Chain and Odd-Numbered Chain Saturated Fatty Acids

Under the given analytical conditions, 36 minor acids were quantified (Table 2). For the oddnumbered chain acids shown in Table 2, which were identified by mass spectrometry, there are no previous references in the literature to caprine milk, although the percentages of $C_{5:0}$, $C_{7:0}$, and $C_{19:0}$ were similar to those reported by Wolf (22) for caprine cheese.

With regard to BCFA, 36 (mostly monomethylates) were quantified. Of these, the most important in quantitative terms (> 0.1%) were the *iso-* and *anteiso-*C_{15:0}, *iso-* and *anteiso-*C_{17:0}, and *iso-*C_{16:0} (see Table 1), which confirms the findings of Gönc et al. (6), Massart-Leen et al. (14), and Wolf (22) for caprine milk and cheese, respectively. Only the *iso-*C_{16:0} was affected by the herd (P < 0.001). Herds 1 and 5 presented the highest values. These five BCFA are also predominant in bovine milk (14, 20).

Previous studies (14) on BCFA in caprine milk fat revealed only the long-chain fatty acids with carbon numbers >11, but, more recently, Ha and Lindsay (8) determined the fraction of volatile fatty acids and quantified other 5 BCFA with carbon numbers <10. Those authors emphasized the wide range of monomethyl-branched components, other than isoand anteiso-fatty acids, mainly with methyl substitution on C_4 and C_6 , which are present in caprine milk fat but are virtually absent from bovine milk. In the present work, 31 minor BCFA (in proportions that ranged from 0.004 to 0.765 mg/g of total fatty acids, Table 2) were identified by mass spectrometry or by comparison with data from the literature (7, 14) (see Table 2) and were quantified. Of these BCFA, 25 were identified as monomethyl BCFA, 2 were dimethyl BCFA, and 4 were ethyl BCFA. Noteworthy in the ethyl BCFA was the presence of 4ethyloctanoate (227 μ g/g). This acid and 4methyloctanoate (391 $\mu g/g$) lend characteristic (goaty and muttony) flavors to dairy products, but other BCFA observed in this work (3 methylbutanoate, 4-methylpentanoate, and 8methylnonanoate) and also identified in bovine milk

Fatty	Herd 1	Herd 2	Herd 3	Herd 4	Herd 5		
acid	(n = 7)	SEM ¹	Significance				
			– (% of total n	nethyl esters) -			
5 _{4:0}	2.09 ^b	2.24 ^a	2.23 ^a	2.22 ^a	2.10 ^b	0.041	*
6:0	2.47 ^a	2.43 ^a	2.41 ^{ab}	2.41 ^{ab}	2.24 ^b	0.050	*
×8:0	2.84 ^a	2.71 ^{ab}	2.76 ^{ab}	2.78 ^{ab}	2.54^{b}	0.063	*
10:0	10.61 ^a	9.89 ^b	9.88 ^b	9.88 ^b	9.61 ^b	0.200	*
/10:1	0.22	0.24	0.25	0.24	0.26	0.013	
/11:0	0.15	0.16	0.17	0.10	0.17	0.017	
12:0	5.49	4.89	4.96	4.84	4.83	0.198	
/12:1	0.18	0.21	0.21	0.16	0.18	0.008	
/13:0	0.15 ^a	0.18 ^a	0.16 ^a	0.10 ^b	0.15 ^a	0.016	*
14.0	10.01	9.69	9.23	9.83	10.28	0.258	
14.1	0.18	0.17	0.18	0.19	0.20	0.011	
<i>so</i> -C _{15:0}	0.12	0.12	0.13	0.15	0.13	0.010	
nteiso-C _{15:0}	0.21	0.21	0.21	0.21	0.22	0.008	
15:0	0.71	0.68	0.63	0.64	0.69	0.040	
15.1	0.10	0.12	0.10	0.11	0.08	0.012	
so-C _{16:0}	0.29 ^a	0.21 ^b	0.21 ^b	0.21 ^b	0.30 ^a	0.018	***
16:0	27.42 ^b	30.08 ^a	25.59^{b}	26.76 ^b	31.31 ^a	0.749	***
16:1	1.63	1.59	1.52	1.30	1.89	0.058	
so-C _{17:0}	0.33	0.36	0.34	0.32	0.39	0.026	
nteiso-C _{17:0}	0.42	0.43	0.37	0.40	0.46	0.035	
17:0	0.72	0.68	0.74	0.69	0.77	0.100	
17:1	0.44 ^{ab}	0.35 ^{bc}	0.43 ^{ab}	0.30 ^c	0.45 ^a	0.023	**
18:0	8.65 ^{bc}	8.00 ^c	9.66 ^{ab}	10.68 ^a	7.39 ^c	0.533	**
18:1	18.68	18.23	21.47	19.76	18.33	0.486	
18.2	3.49 ^a	3.33 ^a	3.23 ^a	3.23 ^a	2.68 ^b	0.096	***
20:0	0.15	0.15	0.16	0.16	0.15	0.027	
18:3	0.43	0.37	0.50	0.37	0.42	0.045	
C _{18:2} conj	0.76 ^{ab}	0.94 ^a	0.82 ^a	0.41 ^c	0.56 ^{bc}	0.073	* *

TABLE 1	Moons a	and significance	of	offort	of	hord	on	fatty	acid	composition	of	anat	milk
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a,b,cMeans in the same row with different superscript letters are different (P < 0.05).

¹Mean standard error.

*P < 0.05.

**P < 0.01.

***P < 0.001.

TABLE 2. Composition and tentative identification of minor (<0.1% weight) branched-chain and odd-numbered chain saturated fatty acids.

Carbon number	Identification	x	SD
		(mg/g	
		0	esters)
5	2-Methylbutanoate ^a	0.005	0.0008
5	3-Methylbutanoate ^b	0.023	0.0025
6	2-Ethylbutanoate ^a	0.004	0.0005
5	Pentanoate (C _{5:0}) ^b	0.134	0.0102
6	3-Methylpentanoate ^a	0.025	0.0030
6	4-Methylpentanoate ^a	0.017	0.0018
8	2-Ethylhexanoate ^a	0.007	0.0014
7	4-Methylhexanoate ^a	0.050	0.0058
7	Heptanoate (C _{7:0}) ^b	0.363	0.0326
8	Methylheptanoate ^a	0.026	0.0030
9	2,4-Dimethylheptanoate ^a	0.051	0.0052
8	Methylheptanoate ^a	0.028	0.0034
9	4-Methyloctanoate ^b	0.391	0.0454
9	6-Methyloctanoate ^a	0.041	0.0063
9	Nonanoate $(C_{9:0})_{b}$	0.915	0.0430
10	4-Ethyloctanoate ^a	0.227	0.0218
10	4-Methylnonanoate ^a	0.016	0.0025
10	8-Methylnonanoate ^a	0.036	0.0042
11	Methyldecanoate ^b	0.660	0.0535
11	Methyldecanoatea	0.016	0.0018
11	Methyldecanoatea	0.027	0.0038
12	2-Ethyldecanoate ^a	0.322	0.3958
11	Undecanoate (C _{11:0}) ^b	0.233	0.0169
12	Dimethyldecanoateb	0.310	0.0366
13	4-Methyldodecanoate ^b	0.135	0.0313
14	12-Methyltridecanoate (<i>iso</i>) ^b	0.765	0.0647
	11-Methyltridecanoate		
14	(anteiso) b	0.370	0.0762
15	Methyltetradecanoatea	0.348	0.0547
15	Methyltetradecanoatea	0.230	0.0560
	13-Methylpentadecanoate		
16	(anteiso) ^b	0.131	0.0292
17	Methylhexadecanoateb	0.250	0.0437
17	Methylhexadecanoate ^b	0.243	0.0733
17	Methylhexadecanoate ^b	0.194	0.0254
	16-Methylheptadecanoate		
18	(<i>iso</i>) ^b	0.578	0.0182
19	Nonadecanoate $(C_{19:0})^{b}$	0.161	0.0192
21	Methyleicosanoate ^b	0.322	0.0714
		5.044	5.0.2.2

^aIdentified by comparison with data from the literature (7, 13). ^bIdentified by mass spectral analysis.

are instrumental in determining the characteristic flavor of dairy foods.

Trans Fatty Acid Content in Caprine Milk Fat

The mean *trans*- $C_{18:1}$ content (Table 3) of caprine milk fat was 2.12%, which was lower than the 3.8% reported for bovine milk fat (17, 21). Information is not available on the caprine milk content of *trans*- $C_{18:1}$, but, from the data obtained by Bickerstaffe et al. (1), it can be deduced that about 12% of the geometrical isomers of $C_{18:1}$ of caprine milk was of *trans*

TABLE 3. Total content of $trans\-C_{16:1}$ and $trans\-C_{18:1}$ in caprine milk fat and proportions of the different peaks of the gas-chromatographic profiles of $trans\-C_{18:1}$ methyl esters isolated by $AgNO_3\-TLC$.

Item	Identification	$\overline{\mathbf{X}}$	SD
		(% of total methyl esters)	
Total trans-C ₁₆₋₁		0.16	0.013
Total trans-C _{18:1}		2.12	0.237
Peak 1	trans-6 to trans-9	5.11	0.646
Peak 2	trans-10 + trans-11	54.19	2.292
Peak 3	trans-12	8.58	0.676
Peak 4	trans-13 + trans-14	17.36	0.942
Peak 5	trans-15	6.08	0.407
Peak 6	trans-16	8.67	0.357

configuration, a proportion similar to that determined in the present work (11.0%). A slightly higher *trans*- $C_{18:1}$ content (2.68%, the minimum value being 1.75%) was found in caprine cheese by Wolf (22).

The profiles of trans-C_{18:1} isomers are illustrated in Figure 1, and the proportions of partially resolved individual trans isomers are shown in Table 3. Peaks were identified from data reported by others (13, 22). The trans-11- $C_{18:1}$ (vaccenic acid) was found to be the most important component of trans-C_{18:1} in caprine milk (1), but, in the given experimental conditions, the *trans*-10- and *trans*-11- $C_{18:1}$ isomers were not completely separated (see Figure 1). The proportion of both isomers was 54.2%, which was higher than reported by Bickerstaffe et al. (1) for caprine milk (36%) and by Wolf (22) for caprine cheese (44.9%) but was comparable with the 48 to 58% reported for bovine milk fat (17, 21). The proportions of trans-12- to trans-16-C_{18:1} isomers were similar to those reported for caprine (1) and also for bovine (17, 22) milk fat. Elaidic acid (trans-9-C_{18:1}) has been the basis for the most clinical nutritional studies that have related the intake of *trans* fatty acids to an increased risk of coronary heart disease. According to Bickerstaffe et al. (1), the elaidic acid is the most important isomer of trans-6-C_{18:1} to trans-9-C_{18:1} group in caprine milk; in the current work, the proportion of these isomers was 5.1%, a value considerably lower than previous data reported for caprine [12% (1)] and bovine [11.6% (17), 8.8% (21)] milk.

Figure 2 shows partial gas chromatograms (the zone between C_{16} and C_{18} FAME) of the saturated plus *trans*-monoenic, *trans*-monoenoic, and saturated FAME fractions separated by AgNO₃-TLC. *Trans*- $C_{16:1}$ isomers were present in caprine milk fat and could not be individually identified when the total

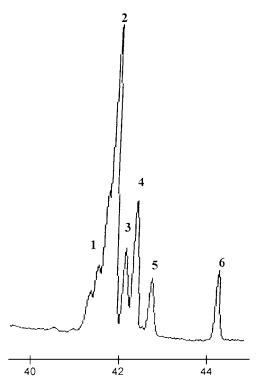


Figure 1. Partial gas chromatographic profile of $trans-C_{18:1}$ methyl esters isolated by AgNO₃-TLC. Peak numbers are detailed in Table 3.

FAME or the saturated plus *trans*-monoenoic acids fraction were analyzed because the *trans*- $C_{16:1}$ peaks coelute with the *iso*- $C_{17:0}$ and $C_{17:0}$ acids. Quantification of the individual *trans*- $C_{16:1}$ fatty acids is possible after AgNO₃-TLC fractionation. The total *trans*- $C_{16:1}$ acid content determined in this study was 0.16%. Although no comparable data have been found for caprine milk, this value was similar to the values reported for bovine milk: 0.13% (15), 0.17% (9), 0.18 (13) and was slightly lower than the value reported for caprine cheese: 0.18% (22).

The data of this study, generated from the milk of five goat herds of the same region, showed significant differences, mainly in the content of long-chain fatty acids. The results also indicated that caprine milk fat contains a very high number of minor BCFA, mostly monomethylates, that have potential implications for the flavor of the dairy products. The content of *trans*- $C_{18:1}$ fatty acids in caprine milk fat presented lower values than those reported for bovine milk fat; however, the proportion of the different *trans*- $C_{18:1}$ isomers and the *trans*- $C_{16:1}$ content were comparable in both species.

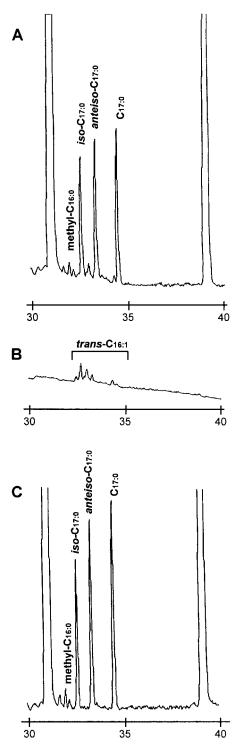


Figure 2. Partial gas chromatographic profiles of *trans* unsaturated plus saturated (A), *trans* unsaturated (B), and saturated (C) fatty acid methyl esters fractions isolated by AgNO₃-TLC.

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