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Profile of *trans* Fatty Acids (FAs) Including *Trans* Polyunsaturated FAs in Representative Fast Food Samples

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ABSTRACT: The content of *trans* fat in foods is most commonly determined by summing the levels of individual *trans* fatty acids (FAs), analyzed as FA methyl esters (FAME) by gas chromatography. Current Official Methods of the American Oil Chemists' Society (AOCS) enable quantitation of total *trans* fat in foods but were not designed for the determination of *trans* FA isomeric compositions. In the present study, the content of *trans* fat in 32 representative fast food samples ranged from 0.1 to 3.1 g per serving, as determined according to AOCS Official Method Ce 1j-07. Further analysis of FAME using the 200 m SLB-IL111 ionic liquid column yielded quantitative results of total, *trans*, saturated, and *cis* unsaturated fat that were comparable to those of Method Ce 1j-07 and also allowed for the complementary determination of individual *trans* 18:1, *trans* 18:2, and *trans* 18:3 FA isomeric compositions under conditions suitable for routine sample analysis.

KEYWORDS: *trans fat, fast food, AOCS Official Method Ce 1j-07, gas chromatography, SP-2560 column, SLB-IL111 ionic liquid column*

INTRODUCTION

There are two main sources of *trans* fat in the food supply, those originating from partial hydrogenation of polyunsaturated oils and those found in ruminant-derived foods occurring as a result of biohydrogenation of polyunsaturated fatty acids (PUFA) by rumen bacteria. Partially hydrogenated oils are known to increase biomarkers of chronic disease risk, especially for cardiovascular disease,[1] and impair essential fatty acid (FA) metabolism.[2,3] Because of this, their daily intake is recommended to be as little as possible.[4−6] *Trans* FA isomers of linoleic acid (*cis*-9,*cis*-12 18:2) and α-linolenic acid (*cis*-9,*cis*-12,*cis*-15 18:3) also appear in the food supply as a result of the deodorization of polyunsaturated vegetable oils[7] and from fryer oils, which are maintained at elevated temperatures for extended periods of time.[8,9] Several studies have indicated that the consumption of these *trans* PUFA may have a more adverse effect on biomarkers of cardiovascular disease risk than the *trans* 18:1 FA isomers.[10−12]

The Nutrition Labeling and Education Act of 1990 (NLEA) amended the Federal Food, Drug and Cosmetic Act (FD&C Act) by adding section 403(q), which specified, with certain exceptions, that a food is considered to be misbranded unless its label or labeling bears nutrition information. The NLEA amendments to the FD&C Act included an exemption for nutrition labeling for food served in restaurants or other establishments in which food is served for immediate human consumption and/or sold for sale or use in such establishments. More recently, section 4205 of the Patient Protection and Affordable Care Act of 2010 (Affordable Care Act) (Pub.L.111−148) amended, among others, section 403(q) of the FD&C Act. As amended, section 4205 would require restaurants and similar retail food establishments (e.g., fast food restaurants) that are part of a chain with 20 or more locations and operating under the same name to provide calorie information for standard items on menus or menu boards. The FDA published a proposed rule for implementing the menu labeling provisions of the Affordable Care Act in the Federal Register on April 6, 2011.[13] Under these proposed provisions, the content of calories from any source and calories from total fat would be required declarations, and written declarations of the amounts of total fat, saturated fat, *trans* fat, cholesterol, sodium, total carbohydrates, sugars, dietary fiber, and total protein would be required upon consumer request. With these changes have come an increased interest in the composition of fast foods, particularly the content of *trans* fat, and there is current, but limited, evidence of already reduced levels of *trans* fat in some restaurant foods in the United States.[14]

The content of *trans* fat in foods is most commonly determined by summing the levels of individual *trans* FAs, analyzed as FA methyl esters (FAME), by gas chromatography (GC) according to one of several Official Methods approved by the American Oil Chemists' Society (AOCS)[15,16] or AOAC International.[17] The most recent Official Methods recommend the use of 100 m cyanopropyl polysiloxane (CPS) capillary columns (e.g., SP-2560, Supelco Inc., Bellefonte, PA; CP-Sil 88, Agilent Technologies, Inc., Wilmington, DE) that permit limited separations of FAMEs over the wide range of hydrocarbon chain lengths (i.e., 4−26 carbons). A limitation of these Official Methods is the incomplete chromatographic resolution of *cis* and *trans* 18:1, 18:2, and 18:3 FAME isomers.[18,19] The current Official Methods were designed to rapidly enable the

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quantitation of total *trans* fat in foods, and alternate chromatographic separation methodologies are recommended if the isomeric composition of *trans* FA is of interest.[16] Recent introduction of the SLB-IL111 ionic liquid capillary column (Supelco Inc.) provided analysts with a novel separation tool characterized by an extremely polar stationary phase and enhanced selectivity toward geometric and positional isomers of unsaturated FAME. Delmonte et al.[20] reported improved separations of *cis* and *trans* 18:1 FAME positional isomers, as well as other complex clusters of FAME, using the 100 m SLB-IL111 column relative to those achieved with 100 m CPS columns recommended for use with the Official Methods.[15−17] Further improved separations of most of the *cis* and *trans* FAME in milk fat could be attained by coupling two 100 m SLB-IL111 columns in-line to form a continuous 200 m column and using a combination ramped temperature and flow program.[21] These improvements in GC methodology have made possible a more accurate determination of both the content and the isomeric composition of *trans* fat in foods.

The primary objective of this study was to analyze the current levels of *trans* fat, including the content of *trans* PUFA, in representative fast food samples from U.S.-based restaurants using the AOAC 996.06 extraction and transmethylation procedure[17] with AOCS Official Method Ce 1j-07[16] for the quantitation of FAMEs. The prepared FAMEs were also analyzed using the 200 m SLB-IL111 ionic liquid column according to the recent method of Delmonte et al.[21] to evaluate whether the quantitation of total, *trans*, saturated, and *cis*-unsaturated fat would be comparable to values obtained using Method Ce 1j-07, while also allowing for the determination of *trans* 18:1, *trans* 18:2, and *trans* 18:3 FA isomeric compositions. The five fast food categories selected for this analysis were hamburgers, cheese pizza, chicken tenders/nuggets, French fries, and apple pie/turnovers.

MATERIALS AND METHODS

Materials. All chemicals and reagents were of ACS reagent grade or higher and purchased from Fisher-Scientific (Pittsburgh, PA) or Sigma-Aldrich (St. Louis, MO). FAME reference standards and the C13:0 triglyceride internal standard were purchased from Nu-Chek Prep., Inc. (Elysian, MN). Methyl phytanate was purchased from Larodan Fine Chemicals AB (Malmö, Sweden). Mixtures of FAMEs containing positional and geometric isomers of 18:1, *cis*-9,*cis*-12 18:2, and *cis*-9,*cis*-12,*cis*-15 18:3 were prepared as previously reported.[18,22]

Sample Selection. A total of 17 fast food restaurants belonging to major U.S. chains and operating in Prince George's County (MD) were identified using the Google search engine. Samples were collected between May and July, 2011, and nutritional information reported by the food establishments was recorded. The categories and number of independent samples per category were as follows: hamburgers (n = 6), cheese pizza (n = 6), chicken tenders/nuggets (n = 7), French fries (n = 7), and apple pie/turnovers (n = 6). Each food item was purchased "as served" (e.g., hamburger, bun, pickle, onions, ketchup, etc.); no additional toppings or sauces were included. The serving size was reported, in grams, by the respective fast food restaurants as the standard amount of food offered per menu item. For chicken tenders/nuggets and French fries, the target serving sizes were approximately 130 and 120 g, respectively, equivalent in most cases to a medium-sized serving. Two servings of each food item were purchased, weighed, and frozen overnight at −75 °C. The duplicate frozen food items were subsequently homogenized to a fine powder or paste using a GrindoMix GM 300 Knife Mill (Retsche, Newton, PA) and stored at −75 °C until further analysis was performed.

Extraction of Total Lipids. Total lipids were extracted in duplicate according to AOAC Official Method 996.06 with slight modifications.[17] For foods excluding dairy products and cheese, homogenized samples were thawed in a water bath (∼15−18 °C), and then, 1 g was weighed to the nearest 0.0001 g into round-bottom flasks. Three milliliters of C13:0 triglyceride internal standard solution (5 mg/mL in chloroform), 1 mL of chloroform, 150 mg of pyrogallic acid, 4 mL of ethanol, and 20 mL of 8.3 M hydrochloric acid were added, and then, flasks were sealed and placed in a shaking water bath set at 80 °C for 60 min. Teflon sleeves (SciMac, Middlesex, NJ) were used to maintain a tight seal between the glass stopper and the round-bottom flask. For pizza, 1 g was weighed to the nearest 0.0001 g into round-bottom flasks, and then, 3 mL of C13:0 triglyceride internal standard solution (5 mg/mL in chloroform), 1 mL of chloroform, 150 mg of pyrogallic acid, 4 mL of ethanol, 8 mL of H_2O, and 4 mL of 58.8% ammonium hydroxide were added. Samples were incubated in a shaking water bath set at 80 °C for 20 min, then 20 mL of 12 M hydrochloric acid was added, and samples were incubated for an additional 20 min. Following incubation, the fast food samples were filtered to remove solid particulates and transferred, along with 40 mL of ethanol, to 125 mL separatory funnels for liquid−liquid extraction. Total lipids were extracted using a 50:50 mixture of petroleum ether and anhydrous diethyl ether (2 extractions, 40 mL of 50:50 petroleum:-diethyl ether per extraction) and transferred to 100 mL round-bottom flasks, passing first through a glass funnel containing anhydrous sodium sulfate. The solvent was removed under vacuum, and the lipid extracts were quantitatively transferred (2 mL of *n*-hexanes followed twice by 2 mL of 50:50 petroleum:diethyl ether) to 16 mm × 125 mm extraction tubes for transmethylation.

Transmethylation. Transmethylation of extracted food lipids (∼150 mg) was carried out according to AOAC Official Method 996.06 using fresh boron trifluoride (7% in methanol). FAMEs were stored in 2 mL silanized, amber-colored autosampler vials with Teflon-lined caps.

Instrumentation. FAMEs were analyzed according to AOCS Official Method Ce 1j-07 on a 6890N gas chromatograph (Agilent Technologies, Inc.) equipped with a flame ionization detector and the SP-2560 CPS column (100 m × 0.25 mm i.d., 0.20 μm film; Supelco Inc.). GC conditions were as follows: The oven was maintained at 180 °C for 32 min and then ramped at 20 °C/min to 215 °C and held for 31.25 min. Hydrogen was the carrier gas at a constant flow of 1 mL/min and a linear velocity of 26 cm/s. The detector air flow was 400 mL/min, and nitrogen make up gas flow was 33.0 mL/min. The injection port and detector temperatures were 235 and 325 °C, respectively. GC conditions reported in Delmonte et al.[21] were as follows: The 200 m SLB-IL111 ionic liquid capillary column was formed by coupling two 100 m SLB-IL111 columns (100 m × 0.25 mm i.d., 0.20 μm film; Supelco Inc.) using an Ultimate Union (Agilent Technologies, Inc.). The GC oven was maintained at 170 °C for 50 min, then ramped at 6 °C/min to 185 °C, and held for 50 min. A 5 min re-equilibration time was used between injections. Hydrogen was the carrier gas with the following ramped flow program: 1.6 mL/min maintained for 35 min, then ramped at 0.30 mL/min/min to 3.0 mL/min and held for 40 min. The detector constant make up gas plus column eluent flow was set at 30 mL/min, hydrogen flow was set at 30 mL/min, and air was set at 400 mL/min. The injection port and detector temperatures were 300 and 250 °C, respectively. The duration of a typical analysis was 65 min with the SP-2560 column and 102.5 min with the 200 m SLB-IL111 column. For both columns, the split ratio was 100:1, and the typical injection volume was 0.3 μL. Chromatograms were processed using ChemStation Rev. B.02.01-SR1 software (Agilent Technologies, Inc.). Individual FAMEs were identified by retention time compared with known reference standards and using available literature.[16,18,21,23,24] Unknown peaks were analyzed by covalent adduct chemical ionization tandem mass spectrometry[25] on a 240 MS ion trap mass spectrometer coupled to a 7890A gas chromatograph (Agilent Technologies, Inc.). Operating conditions were similar to those described for Method Ce 1j-07, except that helium was the carrier gas. Mass spectrometry analysis was not performed with the SLB-IL111 column because flow rates exceeded the capacity of the ion trap mass spectrometer.

Calculations. Calculations were performed according to AOCS Official Method Ce 1j-07.[16] Theoretical flame ionization detector

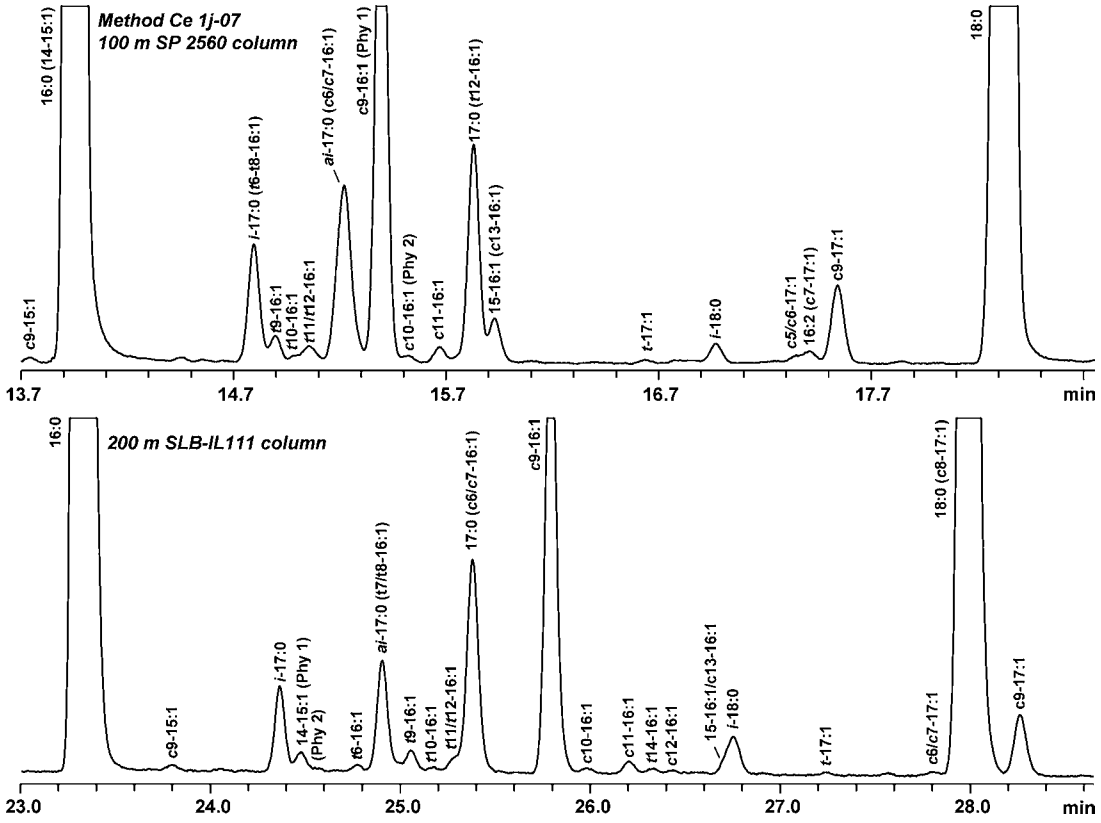

Figure 1. Partial gas chromatograms of the 16:0 to 18:0 FAME region of a typical pizza sample analyzed using the 100 m SP-2560 CPS column according to AOCS Official Method Ce 1j-07 (top chromatogram) and the 200 m SLB-IL111 ionic liquid column according to Delmonte et al.[21] (bottom chromatogram). FAMEs labeled in parentheses were considered a minor component of the coeluting peak and were not quantified separately. Phy, methyl phytanate.

correction factors reported in AOCS Official Method Ce 1 h-05[15] were applied to individual FAMEs. The total fat was calculated based on the sum of all known FA and expressed as triacylglycerol equivalents, whereas the sums of saturated FA (SFA), *cis*-monounsaturated FA (MUFA), *cis*-PUFA, and *trans* FA were expressed as FA equivalents. A relative standard deviation of 3.0 or less for total fat between the two independent extractions of one sample was considered acceptable. The content of *trans* fat was calculated based on the sum of all FAs containing one of more isolated (i.e., nonconjugated) double bonds in the *trans* configuration.

Statistical Analysis. Statistical analysis was performed using JMP (Version 9.0.2, SAS Institute Inc., Cary, NC). One-way ANOVA was used to determine the effect of analytical method on the mean content of total, saturated, *trans*, and *cis*-unsaturated fat in individual fast food samples. When a significant difference was detected ($P < 0.05$), Student's *t* test was used to compare mean values ($\alpha = 0.05$).

RESULTS

Sample Characteristics. Most restaurants reported the composition of frying oil on their Web site or in their nutrition information documents. Use of an oil blend (e.g., canola, soybean, cottonseed, sunflower, and/or corn) was most commonly reported. Two restaurants reported using peanut oil to prepare fried foods; another reported the use of shortening, containing partially hydrogenated beef tallow and partially hydrogenated soybean oil. For one restaurant, the nutrition information document was specific for one city, only, and declared the use of partially hydrogenated soybean oil in the remaining U.S. locations. The *trans* 18:1 FA composition of those samples was consistent with the use of partially hydrogenated oils.[26] For pizza, one restaurant reported their nutrition information on a per slice basis. With the expectation that the size of a slice of pizza could vary within a whole pizza, four slices were included in the analysis to obtain a better representation of the mean weight of one slice. For the remaining pizza samples, the two servings were selected from a medium cheese pizza.

Chromatographic Separations of FAME from Representative Fast Food Samples. The stability of the 200 m SLB-IL111 ionic liquid column was verified by the reproducibility of replicate analyses of a FAME reference standard mixture over a several day period. Partial gas chromatograms of FAME from a typical pizza sample, highlighting the most important *trans* FA-containing regions, achieved with the SP-2560 and 200 m SLB-IL111 capillary columns are presented in Figures 1−3. Both sets of chromatographic conditions led to several coeluting peaks of FAME (described below). Where possible, separations achieved with one column were used to determine the contribution of individual FAME in the corresponding coeluting peak on the other column. In other cases, one of the coeluting FAME was considered a minor component, and the contribution of its peak area was excluded from the final quantitation.

Figure 1 presents the separation of FAME in the chromatographic region from 16:0 to 18:0. Separations achieved with the SLB-IL111 column permitted the baseline integration of several 16:1 isomers that coeluted with the larger-abundance branched and straight chain 17:0 FAME isomers on the SP-2560 column. Resolution of *cis*-6/*cis*-7 16:1 was difficult because these isomers coeluted with *anteiso*-17:0 on the SP-2560 column and with *trans*-11/*trans*-12 16:1 and 17:0 on the SLB-IL111 column. Similarly, 15−16:1, the C16 FAME with a terminal

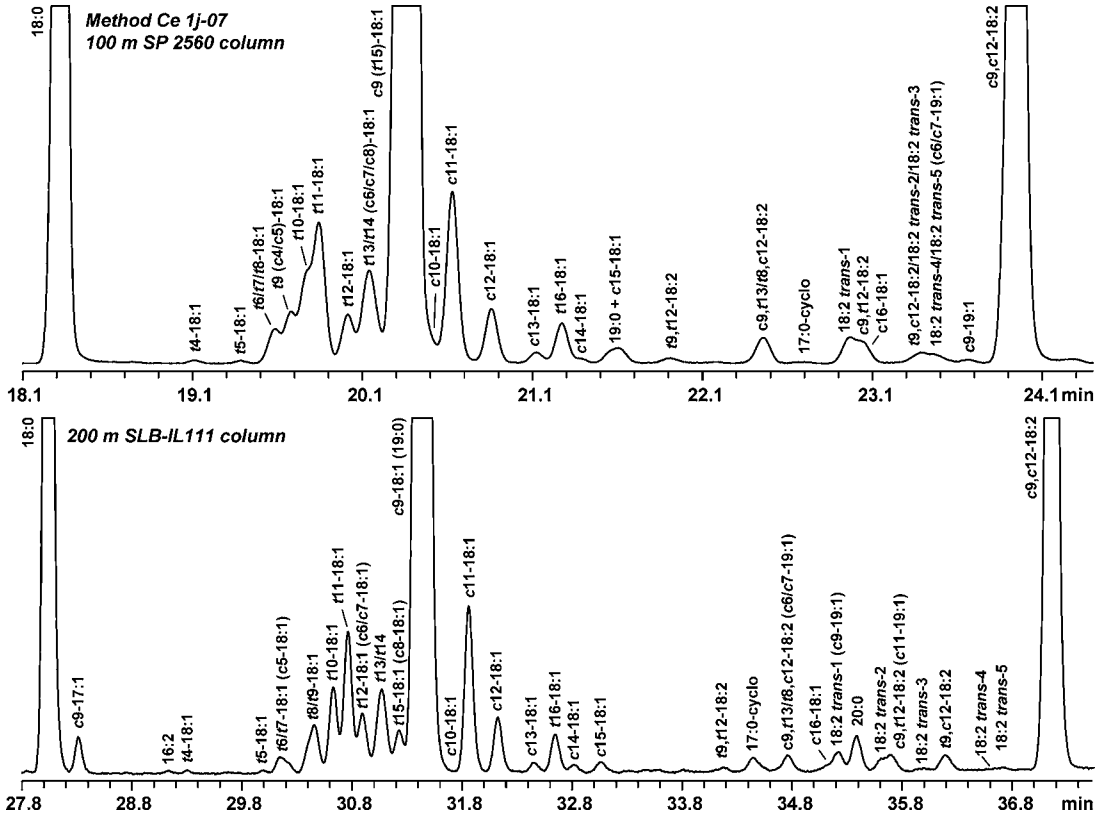

Figure 2. Partial gas chromatograms of the 18:0 to *cis*-9,*cis*-12 18:2 FAME region of a typical pizza sample analyzed using the 100 m SP-2560 CPS column according to Method Ce 1j-07 (top chromatogram) and the 200 m SLB-IL111 ionic liquid column according to Delmonte et al.[21] (bottom chromatogram). FAMEs labeled in parentheses were considered a minor component of the coeluting peak and were not quantified separately. Five 18:2 FAME were left as unidentified (labeled as 18:2 *trans*-1−18:2 *trans*-5) due to the coelution of more than one FAME in a single peak. The 18:2 *trans*-3−*trans*-5 FAMEs were detected only in chicken tenders/nuggets-4 and French fries-4.

double bond, coeluted with *cis*-13 16:1 on the SP-2560 column and with *iso*-18:0 on the SLB-IL111 column. Analysis at 160 °C isothermal temperature on the SLB-IL111 led to the determination that *cis*-6/*cis*-7 16:1 and 15−16:1 contributed 0.08 and 0.04% of total FA, respectively, in the typical pizza sample; thus, the contribution of these minor FA was excluded in the final analysis.

Chromatographic separations achieved with the SLB-IL111 column showed marked improvements in the resolution of *cis* and *trans* 18:1 FAME positional isomers as compared with separations achieved with the SP-2560 column, especially among the *trans*-6 to *trans*-11 FAME positional isomers and between *trans*-15 and *cis*-9 18:1 (Figure 2). The SLB-IL111 column also enabled a greater separation of 18:2 FAME eluting before *cis*-9,*cis*-12 18:2, especially for samples with large abundances of unidentified 18:2 FAME (e.g., chicken tenders/nuggets-4, French fries-4). Partial coelutions of 18:1, 18:2, 19:1, and 20:0 FAME in the 18:2 region of the SLB-IL111 chromatogram were still apparent.

CACI-MS was used to verify the presence of certain low-abundance FAMEs (e.g., 16:2; *cis*-9 17:1) and to exclude certain peaks as being FAMEs. Several methylene and non-methylene interrupted *trans* 18:2 FAME were identified; however, the positions and geometry of the double bonds were not determined because the SP-2560 column was incapable of separating these coeluting FAME under conditions described in Official Method Ce 1j-07. Therefore, no novel mass spectra are presented.

Figure 3 presents the chromatographic separation of FAME eluting between *cis*-9,*cis*-12 18:2 and the conjugated linoleic acid (CLA) isomers. Two major improvements achieved with the SLB-IL111 column, relative to the SP-2560 column, were the separation of *cis*-9 20:1 and *trans*-9,*cis*-12,*cis*-15 18:3 and the separation of *trans*-7,*cis*-9 CLA and *cis*-9,*trans*-11 CLA, which has also been previously reported with the 100 m SLB-IL111 column.[20] Hamburger and pizza samples showed a partial coelution of *cis*-9,*trans*-11 CLA and *cis*-11,*cis*-14 20:2 under conditions achieved with the SLB-IL111 column. In this case, values obtained with the SP-2560 column were used to determine the contribution of individual FAME in the coeluting peak on the SLB-IL111 column, while also accounting for the coelution of *trans*-7,*cis*-9 CLA with *cis*-9,*trans*-11 CLA.

Determination of Total, *trans*, Saturated, and Unsaturated Fats in Representative Fast Food Samples. Table 1 compares the content of total, saturated, and *cis*-unsaturated fat in fast food samples determined using the SP-2560 and SLB-IL111 capillary columns. Overall, very few significant differences were detected for pairwise comparisons of fat and FA content between the two columns, and those were largely due to the small standard deviations between replicates. Absolute differences in the mean levels of total, saturated, and *cis*-unsaturated fat, as determined using the two columns, were <4% (data not presented). In Table 2, the contents of total *trans* FA and *trans* FA with different degrees of unsaturation (i.e., MUFA, 18:2, 18:3) are presented. There was a tendency for higher levels of *trans* FA using the SLB-IL111 column. The exception was for samples with a *trans* 18:2 FA

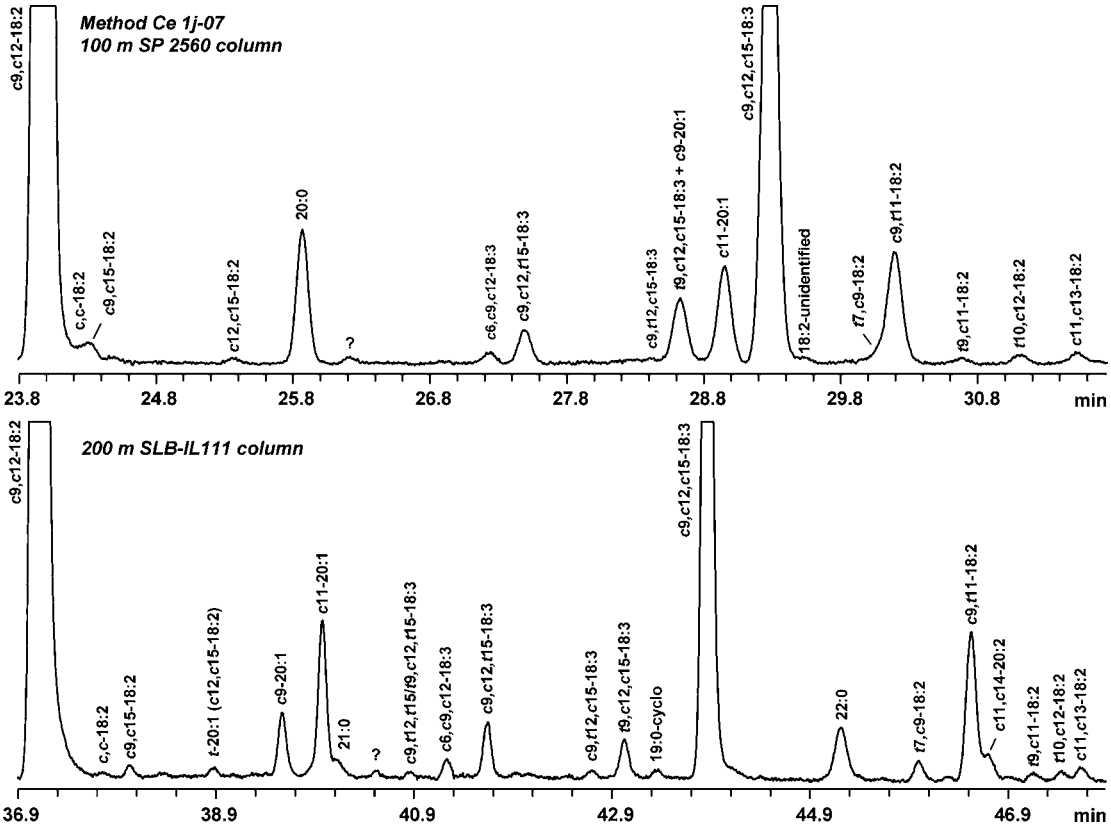

Figure 3. Partial gas chromatograms of the *cis*-9,*cis*-12 18:2 to CLA FAME region of a typical pizza sample analyzed using the 100 m SP-2560 CPS column according to Method Ce 1j-07 (top chromatogram) and the 200 m SLB-IL111 ionic liquid column according to Delmonte et al.[21] (bottom chromatogram). FAMEs labeled in parentheses were considered a minor component of the coeluting peak and were not quantified separately.

content >0.5% of total FA, which tended to have significantly higher levels when analyzed on the SP-2560 column (e.g., Pizza-1, 3−5). The content of *trans*-11 18:1, as determined using the SLB-IL111 column, ranged from 0.03 to 2.28% of total FA and was greatest for chicken tenders/nuggets-4 and French fries-4, both of which had the highest total *trans* FA contents (Table 2).

Table 3 presents a sample-by-sample comparison of the content of individual *trans* FA in four fast food samples determined using the SP-2560 and SLB-IL111 columns. Separations with the SLB-IL111 column permitted the quantitation of six *trans* FA, including *trans*-9 14:1, *trans*-6 16:1, and *trans*-11/*trans*-12 16:1, all of which coeluted with non-*trans* FAME on the SP-2560 column. The quantitation of most *trans* 18:1 FAME positional isomers was achievable with the SLB-IL111 column, while on the SP-2560 column *trans*-9, *trans*-10, and *trans*-11 18:1 coeluted. Separations with the SLB-IL111 column also permitted the resolution of several unidentified 18:2 FAME that coeluted with *cis*-9,*trans*-12 and *trans*-9,*cis*-12 18:2 on the SP-2560 column.

***trans* FA Content of Representative Fast Food Samples.** Table 4 presents the grams per serving of total, *trans*, saturated, and *cis*-unsaturated fat in the 32 fast food samples analyzed on the SP-2560 column. The mean content of *trans* fat in the five fast food categories is illustrated in Figure 4. In hamburgers and pizza samples, the mean levels of *trans* fat were relatively constant at 4.4 ± 1.2 and 4.3 ± 0.7% total FA, respectively (Table 2); thus, samples with the greatest *trans* fat content were also those with the largest serving size. For chicken tenders/nuggets-4 and French fries-4, the content of *trans* fat was 2.3 and 3.1 g per serving, respectively.

The content of *trans* MUFA, *trans* 18:2, and *trans* 18:3 FA in the five fast food categories, as determined using the SLB-IL111 column and expressed as a proportion of total FA, is illustrated in Figure 5. The ruminant-derived samples (i.e., hamburgers and pizza) showed a similar pattern of *trans* FA content with the *trans* MUFA, *trans* 18:2, and *trans* 18:3 FA comprising approximately 3.7, 0.5, and <0.1% of total FA, respectively. For chicken tenders/nuggets, French fries, and apple pie/turnovers, the content of *trans* FA was more variable than that for the ruminant-derived samples, which could be attributed to differences in the content of total *trans* FA within a food category and differences in the content of *trans* MUFA, *trans* 18:2, and *trans* 18:3 FA among individual samples (Table 2).

■ DISCUSSION

In 2006, Stender and colleagues reported a large variability in the content of *trans* fat in chicken nuggets and French fries (<1−24 g of *trans* fat per serving) purchased from fast food restaurants around the world, including several locations in the United States.[27] Currently in the United States, although few systematic investigations have been carried out, evidence does suggest that the levels of *trans* fat in restaurant foods have begun to decline.[14] The present study was designed to provide a snapshot representation of the content of *trans* fat in commonly selected fast food items purchased in Prince George's County (MD) in May−July, 2011. Our results indicated that the current levels ranged from 0.0 to 3.1 g of *trans* fat per serving. Foods with the highest levels had a *trans* fat content that

Table 1. Content of Total, Saturated (SFA), and *cis*-Unsaturated (MUFA, PUFA) Fat in Fast Food Samples Analyzed Using the SP-2560 and 200 m SLB-IL111 Capillary Columns[a]

	total fat (% weight)		SFA (% total FA)		MUFA (% total FA)		PUFA (% total FA)	
sample	SP-2560	SLB-IL111	SP-2560	SLB-IL111	SP-2560	SLB-IL111	SP-2560	SLB-IL111
hamburger-1	14.94 ± 0.15	15.17 ± 0.18	46.64 ± 0.01 A	45.96 ± 0.00 B	41.89±0.02 B	42.38±0.00 A	6.30±0.00 A	6.25±0.01 B
hamburger-2	9.52 ± 0.03 B	9.84 ± 0.03 A	37.50 ± 0.07 A	36.67 ± 0.08 B	40.15 ± 0.31	40.73 ± 0.35	16.66 ± 0.23	16.71 ± 0.22
hamburger-3	10.46 ± 0.00 B	10.62 ± 0.01 A	41.20 ± 0.03 A	40.62 ± 0.02 B	38.75 ± 0.09	39.09 ± 0.11	15.03 ± 0.04	15.02 ± 0.09
hamburger-4	7.70 ± 0.12	7.96 ± 0.15	40.25 ± 0.11 A	39.62 ± 0.10 B	37.17 ± 0.00 B	37.56 ± 0.03 A	16.49 ± 0.12	16.50 ± 0.10
hamburger-5	13.65 ± 0.02 B	14.16 ± 0.00 A	28.83 ± 0.08 A	28.28 ± 0.10 B	31.78 ± 0.12	32.01 ± 0.13	36.41 ± 0.03	36.45 ± 0.03
hamburger-6	10.83 ± 0.01 B	11.00 ± 0.00 A	37.65 ± 0.39	37.26 ± 0.43	34.45 ± 0.02 B	34.63 ± 0.01 A	24.33 ± 0.44	24.21 ± 0.46
pizza-1	7.84 ± 0.12	7.84 ± 0.10	55.92 ± 0.13	55.89 ± 0.15	25.29 ± 0.07	25.06 ± 0.14	12.71 ± 0.00	12.71 ± 0.01
pizza-2	8.77 ± 0.03	8.75 ± 0.01	55.08 ± 0.14	55.32 ± 0.13	24.36 ± 0.02	23.87 ± 0.22	15.81 ± 0.10	15.72 ± 0.10
pizza-3	6.08 ± 0.04	6.08 ± 0.06	57.24 ± 0.11	57.20 ± 0.08	28.09 ± 0.06	27.97 ± 0.07	10.28 ± 0.02 A	10.11 ± 0.04 B
pizza-4	8.64 ± 0.04	8.67 ± 0.05	49.72 ± 0.18	49.56 ± 0.07	23.43 ± 0.14	23.30 ± 0.04	22.98 ± 0.02	22.93 ± 0.03
pizza-5	9.94 ± 0.04	9.96 ± 0.03	54.55 ± 0.32	54.51 ± 0.22	31.04 ± 0.18	31.02 ± 0.18	10.23 ± 0.03	10.11 ± 0.06
pizza-6	8.86 ± 0.00	8.87 ± 0.01	48.79 ± 0.08	48.79 ± 0.10	26.39 ± 0.07 A	26.16 ± 0.03 B	20.71 ± 0.05	20.64 ± 0.04
chick. tenders-1	7.08 ± 0.02 B	7.22 ± 0.02 A	21.58 ± 0.00 B	21.72 ± 0.02 A	54.73 ± 0.01 A	54.61 ± 0.03 B	22.98 ± 0.00 A	22.93 ± 0.01 B
chick. tenders-2	10.30 ± 0.07	10.45 ± 0.07	17.96 ± 0.02	18.07 ± 0.05	49.76 ± 0.01	49.68 ± 0.05	30.59 ± 0.00 A	30.54 ± 0.00 B
chick. tenders-3	20.66 ± 0.32	20.94 ± 0.31	21.04 ± 0.04	21.07 ± 0.03	29.49 ± 0.00 A	29.40 ± 0.02 B	48.61 ± 0.04	48.67 ± 0.00
chick. tenders-4	12.82 ± 0.04 B	13.13 ± 0.01 A	20.67 ± 0.03 A	20.31 ± 0.02 B	32.27 ± 0.02	32.23 ± 0.02	35.37 ± 0.03 B	35.49 ± 0.02 A
chick. tenders-5	21.32 ± 0.58	21.40 ± 0.54	14.59 ± 0.03	14.70 ± 0.05	48.16 ± 0.04	48.08 ± 0.04	36.37 ± 0.00	36.37 ± 0.00
chick. tenders-6	20.75 ± 0.02 B	21.07 ± 0.01 A	23.86 ± 0.03	23.88 ± 0.01	31.66 ± 0.04	31.55 ± 0.03	43.35 ± 0.07	43.43 ± 0.04
chick. tenders-7	13.37 ± 0.12	13.54 ± 0.15	46.13 ± 0.10	45.82 ± 0.06	39.36 ± 0.00 B	39.59 ± 0.01 A	8.26 ± 0.03	8.23 ± 0.08
french fries-1	15.52 ± 0.28	15.79 ± 0.30	10.74 ± 0.13	10.89 ± 0.13	63.99 ± 0.11	63.83 ± 0.10	24.63 ± 0.04	24.58 ± 0.05
french fries-2	13.99 ± 0.23	14.22 ± 0.22	12.96 ± 0.55	13.08 ± 0.55	50.71 ± 0.01 A	50.54 ± 0.03 B	35.71 ± 0.54	35.67 ± 0.53
french fries-3	12.78 ± 0.11	13.00 ± 0.12	19.17 ± 0.00 B	19.36 ± 0.05 A	57.49 ± 0.02 A	57.33 ± 0.04 B	22.84 ± 0.02 A	22.74 ± 0.01 B
french fries-4	17.25 ± 0.33	17.50 ± 0.43	20.72 ± 0.14	20.44 ± 0.02	33.65 ± 0.22	33.78 ± 0.02	33.12 ± 0.17	33.09 ± 0.05
french fries-5	12.14 ± 0.07	12.31 ± 0.11	19.74 ± 0.01 B	19.82 ± 0.01 A	23.92 ± 0.01	23.93 ± 0.03	55.12 ± 0.01 A	54.97 ± 0.02 B
french fries-6	14.34 ± 0.12	14.83 ± 0.17	18.48 ± 0.01 A	18.40 ± 0.01 B	25.48 ± 0.00	25.51 ± 0.02	55.14 ± 0.02	55.14 ± 0.00
french fries-7	16.47 ± 0.14	16.71 ± 0.13	44.80 ± 0.08	44.50 ± 0.15	40.35 ± 0.11	40.56 ± 0.08	8.09 ± 0.02	8.04 ± 0.02
apple pie-1	14.89 ± 0.33	15.07 ± 0.36	59.84 ± 0.04 A	59.52 ± 0.06 B	31.34 ± 0.01	31.55 ± 0.09	8.43 ± 0.05	8.51 ± 0.04
apple pie-2	9.83 ± 0.26	9.88 ± 0.19	41.65 ± 0.32	41.56 ± 0.31	34.54 ± 0.13	34.52 ± 0.15	22.97 ± 0.45	23.06 ± 0.40
apple pie-3	14.83 ± 0.04 B	15.03 ± 0.01 A	53.01 ± 0.23	52.92 ± 0.25	30.11 ± 0.19	30.11 ± 0.19	15.46 ± 0.04	15.55 ± 0.08
apple pie-4	16.07 ± 0.01 B	16.31 ± 0.01 A	27.03 ± 0.02	26.99 ± 0.04	32.97 ± 0.01	32.90 ± 0.02	38.25 ± 0.02	38.30 ± 0.01
apple pie-5	11.75 ± 0.45	11.92 ± 0.30	43.86 ± 0.17	43.77 ± 0.11	37.58 ± 0.17	37.54 ± 0.12	17.82 ± 0.03	17.84 ± 0.00
apple pie-6	14.00 ± 0.05	14.15 ± 0.11	39.81 ± 0.10	39.68 ± 0.07	35.61 ± 0.04	35.61 ± 0.12	19.73 ± 0.15	19.73 ± 0.17

[a]Chromatographic conditions for the 100 m SP-2560 CPS column were according to AOCS Official Method Ce 1j-07 and for the 200 m SLB-IL111 column were according to Delmonte et al.[21] Values represent the means ± SDs of two independent extracts for each food item. A single analysis of FAME by GC on each of the GC columns was performed. One-way ANOVA was used to determine the effect of analytical method on the content of total, saturated, and *cis*-unsaturated fat; mean comparisons were performed using Student's *t* test, $\alpha = 0.05$. Online letters within a column indicate significant differences.

was nearly 1.5× the recommended maximum daily intake for *trans* fat.[28,29]

The present study evaluated the quantitation of *trans* fat in restaurant foods using the 200 m SLB-IL111 ionic liquid column. This column, characterized by its extremely polar stationary phase, was selected based on previous reports of improved separations of FAME from milk fat, especially positional isomers of *trans* 18:1 FAME, relative to those achieved with the 100 m SLB-IL111 column and conventional 100 m CPS columns.[20,21] To the best of our knowledge, we are aware of no study that has yet compared the quantitation of *trans* fat using the 200 m SLB-IL111 column relative to that of 100 m CPS columns recommended for use with the Official Methods.[15−17] However, Richter and colleagues previously reported the use of a 200 m CP7421 CPS column for the quantitation of *trans* fat in conventional and restaurant foods,[30] indicating that the use of 200 m column lengths is becoming more routine in the research setting.

According to AOAC Official Method 996.06, the quantitation of *trans* fat from partially hydrogenated oils involves the summation of FAME peak areas in regions of the chromatogram where non-*trans* FAME are known to coelute.[17] This rationale was based on the expectation that in partially hydrogenated oils the content of non-*trans* FAME would be negligible relative to the greater abundance *trans* 18:1 and *trans* 18:2 FAME. In the present study, Method Ce 1j-07 was applied for the quantitation of FAME because the chromatographic conditions were designed to improve the separation of unsaturated FAME relative to those of Method 996.06. We speculated that the quantitation of FAME using the 200 m SLB-IL111 ionic liquid column would enable a more accurate determination of the content of *trans* fat due to further improvements in the separation of unsaturated FAMEs. However, quantitative analysis of *trans* fat with the 200 m SLB-IL111 column was, in fact, comparable to that of Method Ce 1j-07, confirming the validity of Method Ce 1j-07 for the determination of total *trans* fat in foods.

In 2003, the FDA published a final rule requiring a mandatory declaration of the content of *trans* fat on the Nutrition Facts label of conventional foods and dietary supplements.[31] Under provisions of this rule, all FAs containing one or more isolated (i.e., nonconjugated) *trans* double bonds were included

Table 2. Content of *trans* Fat (% Total FA) in Fast Food Samples Analyzed Using the SP-2560 and 200 m SLB-IL111 Capillary Columns[a]

	total *trans* FA		*trans* MUFA		*trans*-11 18:1	*trans* 18:2 FA		*trans* 18:3 FA	
sample	SP-2560	SLB-IL111	SP-2560	SLB-IL111	SLB-IL111	SP-2560	SLB-IL111	SP-2560	SLB-IL111
hamburger-1	4.81 ± 0.03 B	5.00 ± 0.01 A	4.21 ± 0.02 B	4.38 ± 0.00 A	0.96 ± 0.01	0.59 ± 0.01	0.59 ± 0.01	0.00 ± 0.00 B	0.04 ± 0.00 A
hamburger-2	5.35 ± 0.03 B	5.57 ± 0.07 A	4.82 ± 0.04	5.02 ± 0.07	0.63 ± 0.00	0.49 ± 0.01	0.46 ± 0.01	0.05 ± 0.00 B	0.10 ± 0.00 A
hamburger-3	4.59 ± 0.02 B	4.82 ± 0.01 A	4.06 ± 0.03 B	4.24 ± 0.00 A	1.13 ± 0.01	0.51 ± 0.00	0.52 ± 0.00	0.02 ± 0.00 B	0.06 ± 0.00 A
hamburger-4	5.70 ± 0.02 B	5.92 ± 0.03 A	5.18 ± 0.03 B	5.37 ± 0.03 A	0.91 ± 0.00	0.49 ± 0.01	0.49 ± 0.00	0.03 ± 0.00 B	0.05 ± 0.00 A
hamburger-5	2.79 ± 0.01 B	3.02 ± 0.00 A	2.12 ± 0.01 B	2.22 ± 0.01 A	0.50 ± 0.01	0.46 ± 0.00	0.45 ± 0.01	0.20 ± 0.00 B	0.34 ± 0.00 A
hamburger-6	3.23 ± 0.03 B	3.52 ± 0.01 A	2.51 ± 0.04 B	2.69 ± 0.02 A	1.29 ± 0.01	0.60 ± 0.01	0.62 ± 0.00	0.12 ± 0.01 B	0.22 ± 0.00 A
pizza-1	5.69 ± 0.06 B	5.90 ± 0.02 A	4.91 ± 0.02 B	5.22 ± 0.02 A	1.37 ± 0.01	0.74 ± 0.03 A	0.63 ± 0.01 B	0.04 ± 0.00 B	0.05 ± 0.00 A
pizza-2	4.49 ± 0.02 B	4.78 ± 0.00 A	3.87 ± 0.00 B	4.17 ± 0.03 A	1.02 ± 0.01	0.57 ± 0.02	0.53 ± 0.01	0.05 ± 0.00	0.07 ± 0.02
pizza-3	4.12 ± 0.02 B	4.43 ± 0.03 A	3.50 ± 0.02 B	3.85 ± 0.05 A	0.98 ± 0.00	0.61 ± 0.01 A	0.56 ± 0.02 B	ND B	0.02 ± 0.00 A
pizza-4	3.65 ± 0.03 B	3.94 ± 0.02 A	2.97 ± 0.03 B	3.23 ± 0.02 A	0.74 ± 0.01	0.57 ± 0.00 A	0.54 ± 0.00 B	0.12 ± 0.00 B	0.17 ± 0.00 A
pizza-5	3.93 ± 0.11	4.06 ± 0.02	3.18 ± 0.11	3.36 ± 0.02	0.81 ± 0.01	0.68 ± 0.00 A	0.57 ± 0.00 B	0.08 ± 0.00 B	0.13 ± 0.01 A
pizza-6	3.86 ± 0.04 B	4.12 ± 0.02 A	3.18 ± 0.01 B	3.47 ± 0.01 A	0.84 ± 0.01	0.58 ± 0.05	0.51 ± 0.01	0.10 ± 0.00 B	0.14 ± 0.00 A
chick. tenders-1	0.71 ± 0.00 B	0.74 ± 0.00 A	0.32 ± 0.00 A	0.31 ± 0.00 B	0.03 ± 0.00	0.34 ± 0.00 B	0.39 ± 0.00 A	0.05 ± 0.00 A	0.03 ± 0.00 B
chick. tenders-2	1.69 ± 0.00	1.71 ± 0.00	0.60 ± 0.00 A	0.56 ± 0.00 B	0.10 ± 0.00	0.32 ± 0.00 B	0.38 ± 0.00 A	0.78 ± 0.00	0.77 ± 0.00
chick. tenders-3	0.87 ± 0.00	0.84 ± 0.01	0.22 ± 0.00	0.20 ± 0.01	0.03 ± 0.00	0.46 ± 0.00	0.47 ± 0.00	0.18 ± 0.00	0.18 ± 0.00
chick. tenders-4	11.69 ± 0.01 B	11.88 ± 0.02 A	8.45 ± 0.03 B	8.68 ± 0.01 A	2.15 ± 0.00	2.64 ± 0.01	2.59 ± 0.02	0.61 ± 0.00	0.61 ± 0.00
chick. tenders-5	0.89 ± 0.01 A	0.84 ± 0.01 B	0.31 ± 0.01 A	0.22 ± 0.01 B	0.02 ± 0.00	0.30 ± 0.00 B	0.33 ± 0.00 A	0.28 ± 0.00 B	0.29 ± 0.00 A
chick. tenders-6	1.12 ± 0.00	1.12 ± 0.01	0.32 ± 0.00 A	0.30 ± 0.00 B	0.02 ± 0.00	0.39 ± 0.01 B	0.42 ± 0.01 A	0.41 ± 0.00 A	0.40 ± 0.00 B
chick. tenders-7	6.05 ± 0.07	6.15 ± 0.01	5.54 ± 0.07	5.67 ± 0.01	1.01 ± 0.01	0.51 ± 0.00 A	0.46 ± 0.01 B	0.00 ± 0.00 B	0.03 ± 0.00 A
french fries-1	0.63 ± 0.02	0.70 ± 0.02	0.11 ± 0.01	0.10 ± 0.01	ND	0.18 ± 0.00 B	0.25 ± 0.00 A	0.34 ± 0.00	0.35 ± 0.01
french fries-2	0.63 ± 0.01 B	0.70 ± 0.00 A	0.14 ± 0.01	0.13 ± 0.01	0.01 ± 0.00	0.21 ± 0.00 B	0.27 ± 0.01 A	0.28 ± 0.00 B	0.30 ± 0.00 A
french fries-3	0.51 ± 0.00 B	0.56 ± 0.00 A	0.07 ± 0.00	0.07 ± 0.00	ND	0.43 ± 0.00 B	0.47 ± 0.00 A	0.01 ± 0.00 B	0.02 ± 0.00 A
french fries-4	12.51 ± 0.53	12.62 ± 0.02	8.87 ± 0.04 B	9.20 ± 0.04 A	2.28 ± 0.00	3.05 ± 0.56	2.79 ± 0.01	0.59 ± 0.00 B	0.63 ± 0.00 A
french fries-5	1.21 ± 0.01	1.25 ± 0.02	0.15 ± 0.00 A	0.12 ± 0.01 B	0.01 ± 0.00	0.51 ± 0.01 B	0.54 ± 0.00 A	0.56 ± 0.00	0.59 ± 0.01
french fries-6	0.91 ± 0.01	0.92 ± 0.00	0.11 ± 0.01	0.09 ± 0.00	ND	0.53 ± 0.00	0.54 ± 0.01	0.27 ± 0.00 B	0.29 ± 0.00 A
french fries-7	6.54 ± 0.04	6.64 ± 0.05	5.99 ± 0.00 B	6.07 ± 0.02 A	1.08 ± 0.00	0.51 ± 0.04	0.51 ± 0.01	0.04 ± 0.00	0.06 ± 0.01
apple pie-1	0.39 ± 0.00 B	0.42 ± 0.00 A	0.12 ± 0.01	0.14 ± 0.00	0.01 ± 0.00	0.22 ± 0.00 B	0.24 ± 0.00 A	0.05 ± 0.00	0.05 ± 0.00
apple pie-2	0.84 ± 0.00	0.85 ± 0.06	0.20 ± 0.01	0.23 ± 0.01	0.03 ± 0.00	0.43 ± 0.01	0.41 ± 0.06	0.21 ± 0.00	0.22 ± 0.00
apple pie-3	1.42 ± 0.00	1.41 ± 0.02	0.93 ± 0.00	0.89 ± 0.02	0.17 ± 0.01	0.37 ± 0.00 B	0.40 ± 0.00 A	0.11 ± 0.00 B	0.13 ± 0.00 A
apple pie-4	1.75 ± 0.01	1.78 ± 0.01	0.85 ± 0.00	0.85 ± 0.01	0.15 ± 0.00	0.40 ± 0.01 B	0.44 ± 0.00 A	0.50 ± 0.00 A	0.48 ± 0.00 B
apple pie-5	0.74 ± 0.03	0.84 ± 0.02	0.24 ± 0.02 B	0.34 ± 0.02 A	0.03 ± 0.00	0.37 ± 0.01	0.38 ± 0.00	0.13 ± 0.00	0.12 ± 0.00
apple pie-6	4.73 ± 0.02 B	4.81 ± 0.02 A	3.97 ± 0.02	3.99 ± 0.00	0.74 ± 0.01	0.57 ± 0.00 A	0.53 ± 0.01 B	0.19 ± 0.00 B	0.29 ± 0.01 A

[a]Chromatographic conditions for the 100 m SP-2560 CPS column were according to AOCS Official Method Ce 1j-07 and for the 200 m SLB-IL111 column were according to Delmonte et al. [21] Values represent the means ± SDs of two independent extracts for each food item. A single analysis of FAME by GC on each of the GC columns was performed. One-way ANOVA was used to determine the effect of analytical method on the content of *trans* fat; mean comparisons were performed using Student's *t* test, $\alpha = 0.05$. Online letters within a column indicate significant differences.

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Table 3. Content of Individual *trans* FA (% Total FA) in Representative Fast Food Samples Analyzed Using the SP-2560 and 200 m SLB-IL111 Columns[a]

	hamburger 3-1		pizza 5-1		chicken tenders 4-1		french fries 7-1	
trans FA (% total FA)	SP-2560	SLB-IL111	SP-2560	SLB-IL111	SP-2560	SLB-IL111	SP-2560	SLB-IL111
*t*11−14:1	CE	ND	CE	0.06	CE	ND	CE	0.01
*t*6/*t*7−15:1	ND	ND	ND	0.01	0.02	0.02	ND	0.01
*t*6−16:1	CE	0.02	ND	0.01	CE	ND	CE	0.03
*t*9−16:1	0.03	0.03	0.05	0.05	ND	ND	0.02	0.02
*t*11/*t*12−16:1	0.04	0.04	0.05	0.04	ND	ND	0.04	0.03
*t*6−16:1	CE	0.01	ND	0.01	CE	ND	CE	0.02
t-17:1	ND	ND	ND	0.01	ND	ND	ND	ND
*t*4−18:1	0.01	ND	0.02	0.02	0.02	0.02	0.01	0.02
*t*5−18:1	0.03	ND	0.01	0.01	0.04	0.03	0.06	0.02
*t*6/*t*7--18:1	0.28	0.12	0.19	0.14	0.90	0.26	0.43	0.19
*t*8/*t*9--18:1	*	0.49	0.26	0.35	*	1.46	*	0.73
*t*10−18:1	3.05*	1.62	1.32*	0.47	5.35*	2.32	4.53*	3.01
*t*11−18:1	*	1.12	*	0.81	*	2.15	*	1.08
*t*12−18:1	0.14	0.22	0.28	0.37	0.76	0.93	0.16	0.31
*t*13/*t*14−18:1	0.33	0.29	0.67	0.57	1.21	1.01	0.51	0.34
*t*15−18:1	CE	0.14	CE	0.20	CE	0.23	CE	0.13
*t*16−18:1	0.17	0.12	0.25	0.23	0.17	0.14	0.23	0.12
*t*9,*t*12−18:2	0.03	0.02	0.04	0.03	0.05	0.05	0.05	0.02
*c*9,*t*12−18:2	0.08	0.10	0.14	0.09	0.84 •	0.73	0.08	0.10
*t*9,*c*12−18:2	0.03	0.10	0.09	0.11	0.75 ‡	0.73	0.05	0.10
*c*9,*t*13/*t*8,*c*12−18:2	0.13	0.10	0.18	0.11	0.72	0.54	0.12	0.09
18:2 *trans*-1	0.10	0.15	0.16	0.15	•	0.24	0.10	0.14
18:2 *trans*-2	0.13	0.05	0.06	0.07	‡	0.03	0.13	0.05
18:2 *trans*-3	ND	ND	ND	ND	‡	0.04	ND	ND
18:2 *trans*-4	ND	ND	ND	ND	0.22	0.08	ND	ND
18:2 *trans*-5	ND	ND	ND	ND	0.06	0.15	ND	ND
*c*9,*c*12,*t*15−18:3	0.02	0.03	0.06	0.07	0.28	0.27	0.03	0.01
*c*9,*t*12,*c*15−18:3	ND	0.02	ND	0.01	0.07	0.09	ND	0.01
*t*9,*c*12,*c*15−18:3	ND	ND	0.02	0.06	0.21	0.21	0.01	0.03
*c*9,*t*12,*t*15/*t*9,*c*12,*t*15−18:3	ND	0.02	ND	ND	0.05	0.04	ND	0.01
t-20:1	ND	ND	ND	ND	ND	0.11	ND	0.01
∑ *trans* MUFA	4.08	4.24	3.10	3.37	8.46	8.68	5.99	6.05
∑ *trans* 18:2	0.51	0.52	0.67	0.57	2.63	2.57	0.54	0.49
∑ *trans* 18:3	0.02	0.06	0.08	0.13	0.60	0.61	0.04	0.06
sum	4.61	4.82	3.85	4.07	11.70	11.86	6.57	6.61

[a]Chromatographic conditions for the 100 m SP-2560 CPS column were according to AOCS Official Method Ce 1j-07 and for the 200 m SLB-IL111 column were according to Delmonte et al.[21] Values represent the content of *trans* FA (% total FA) in a single fast food sample. Values in a column sharing a common symbol indicate a coelution of FA. CE, the *trans* FA, if present in the sample, coeluted with a non-*trans* FA; ND, not detected; NID, nonidentified.

regardless of origin. In ruminant-derived foods, especially milk fat, *trans*-11 18:1 is the major *trans* 18:1 FA, and its potential health benefits in humans are related to its role as a precursor for *cis*-9,*trans*-11 18:2, a CLA isomer shown to provide anti-carcinogenic, antiatherogenic, and anti-inflammatory effects in studies with animal models.[32] *Trans*-10 is another notable *trans* 18:1 FA, and its presence at high levels in milk and meat, equal to or greater than that of *trans*-11 18:1, is indicative of alternate biohydrogenation pathways when certain diets are offered to ruminants.[33−35] Unlike *trans*-11, *trans*-10 18:1 is not known to provide any beneficial health effects in humans. The current Official Methods offer a limited capacity for separating, and therefore accurately quantitating, positional isomers of *trans* 18:1 FAME.[18,19,23,36] In the present study, separations achieved with the SP-2560 column led to the coelution of *trans*-9−*trans*-11 18:1, while on the 200 m SLB-IL111 column, most of the *trans* 18:1 FAME positional isomers, including *trans*-10 and *trans*-11, were resolved. Additionally, separation of *trans*-15 and *cis*-9 18:1 has not been previously achieved with any of the Official Methods. The higher values for *trans* MUFA achieved using the 200 m SLB-IL111 column could be attributed, in part, to this novel quantitation of *trans*-15 18:1.

Often times, the reference to *trans* fat in foods is synonymous with the content of *trans* 18:1 FA, a simplification that overlooks the contribution of *trans* 18:2 and *trans* 18:3 FA to the content of total *trans* fat. The deodorization step in the processing of food oils causes the isomerization of double bonds in all-*cis* PUFA, the natural form of most PUFA in vegetable oils, leading to the formation of mono-*trans* and di-*trans* PUFA. The content of these *trans* PUFA may reach as much as 3.5% of total FA in food oils.[7] In the present study, the content of *trans* MUFA, *trans* 18:2, and *trans* 18:3 FA differed by food category. For ruminant-derived foods, *trans* MUFA, largely *trans* 18:1 FA, were the predominant *trans* FA class (4.4% of total FA), and the combined content of *trans* 18:2 and *trans* 18:3 FA was relatively minor (<1% of total FA). For many of the chicken

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Table 4. Grams Per Serving of Total, *trans*, Saturated, and *cis*-Unsaturated Fat in Representative Fast Food Samples[a]

sample	serving size (g)	total fat (g)	*trans* fat (g)[b]	SFA (g)	MUFA (g)	PUFA (g)
hamburger-1	186 ± 2	27.85 ± 0.05	1.28 ± 0.01	12.41 ± 0.02	11.14 ± 0.02	1.68 ± 0.00
hamburger-2	221 ± 3	21.02 ± 0.21	1.08 ± 0.02	7.53 ± 0.09	8.06 ± 0.02	3.35 ± 0.08
hamburger-3	107 ± 2	11.22 ± 0.19	0.49 ± 0.01	4.42 ± 0.08	4.16 ± 0.06	1.61 ± 0.03
hamburger-4	87 ± 0	6.72 ± 0.08	0.37 ± 0.01	2.58 ± 0.04	2.39 ± 0.03	1.06 ± 0.01
hamburger-5	165 ± 1	22.47 ± 0.11	0.60 ± 0.01	6.19 ± 0.05	6.83 ± 0.01	7.82 ± 0.05
hamburger-6	135 ± 2	14.65 ± 0.24	0.45 ± 0.00	5.27 ± 0.03	4.82 ± 0.08	3.41 ± 0.12
pizza-1	188 ± 2	14.73 ± 0.39	0.80 ± 0.03	7.85 ± 0.19	3.55 ± 0.10	1.78 ± 0.05
pizza-2	94 ± 8	8.20 ± 0.70	0.35 ± 0.03	4.31 ± 0.36	1.90 ± 0.16	1.24 ± 0.11
pizza-3	71 ± 3	4.33 ± 0.23	0.17 ± 0.01	2.36 ± 0.13	1.16 ± 0.06	0.42 ± 0.02
pizza-4	58 ± 2	4.98 ± 0.21	0.17 ± 0.01	2.36 ± 0.09	1.11 ± 0.05	1.09 ± 0.05
pizza-5	76 ± 10	7.52 ± 0.99	0.28 ± 0.03	3.91 ± 0.54	2.23 ± 0.28	0.73 ± 0.09
pizza-6	75 ± 5	6.67 ± 0.40	0.25 ± 0.01	3.10 ± 0.18	1.68 ± 0.11	1.32 ± 0.08
chicken tenders-1	127 ± 9	8.99 ± 0.61	0.06 ± 0.00	1.86 ± 0.12	4.71 ± 0.32	1.98 ± 0.13
chicken tenders-2	143 ± 6	14.72 ± 0.73	0.24 ± 0.01	2.53 ± 0.13	7.01 ± 0.35	4.31 ± 0.21
chicken tenders-3	120 ± 6	24.87 ± 0.95	0.21 ± 0.01	5.00 ± 0.20	7.01 ± 0.27	11.56 ± 0.43
chicken tenders-4	158 ± 1	20.23 ± 0.24	2.26 ± 0.02	4.00 ± 0.05	6.24 ± 0.08	6.84 ± 0.07
chicken tenders-5	109 ± 22	23.35 ± 5.31	0.20 ± 0.05	3.26 ± 0.75	10.76 ± 2.44	8.13 ± 1.85
chicken tenders-6	128 ± 4	26.53 ± 0.85	0.28 ± 0.01	6.05 ± 0.20	8.03 ± 0.27	11.00 ± 0.33
chicken tenders-7	153 ± 16	20.52 ± 2.33	1.18 ± 0.12	9.05 ± 1.05	7.72 ± 0.87	1.62 ± 0.18
french fries-1	115 ± 8	17.87 ± 1.50	0.11 ± 0.01	1.84 ± 0.13	10.94 ± 0.94	4.21 ± 0.36
french fries-2	106 ± 14	14.84 ± 2.19	0.09 ± 0.01	1.85 ± 0.35	7.20 ± 1.06	5.06 ± 0.67
french fries-3	233 ± 90	29.84 ± 11.70	0.14 ± 0.06	5.48 ± 2.15	16.42 ± 6.43	6.52 ± 2.56
french fries-4	150 ± 17	25.88 ± 2.38	3.10 ± 0.42	5.13 ± 0.44	8.33 ± 0.71	8.20 ± 0.71
french fries-5	141 ± 10	17.14 ± 1.13	0.20 ± 0.01	3.24 ± 0.22	3.92 ± 0.26	9.04 ± 0.59
french fries-6	115 ± 7	16.47 ± 0.87	0.14 ± 0.01	2.91 ± 0.15	4.01 ± 0.21	8.68 ± 0.45
french fries-7	83 ± 6	13.67 ± 0.94	0.85 ± 0.05	5.85 ± 0.39	5.27 ± 0.38	1.06 ± 0.07
apple pie-1	71.3 ± 0.1	10.62 ± 0.22	0.04 ± 0.00	6.06 ± 0.13	3.18 ± 0.07	0.85 ± 0.01
apple pie-2	121 ± 4	11.84 ± 0.08	0.01 ± 0.00	4.71 ± 0.00	3.91 ± 0.01	2.60 ± 0.07
apple pie-3	86 ± 0	12.73 ± 0.03	0.17 ± 0.00	6.45 ± 0.01	3.66 ± 0.03	1.88 ± 0.01
apple pie-4	163 ± 5	26.24 ± 0.77	0.44 ± 0.02	6.78 ± 0.19	8.27 ± 0.24	9.60 ± 0.29
apple pie-5	171 ± 22	20.09 ± 3.37	0.14 ± 0.02	8.41 ± 1.38	7.21 ± 1.24	3.42 ± 0.58
apple pie-6	86 ± 1	12.04 ± 0.22	0.54 ± 0.01	4.58 ± 0.10	4.10 ± 0.08	2.27 ± 0.02
avg. hamburger	150 ± 50	17.32 ± 7.84	0.71 ± 0.37	6.40 ± 3.38	6.23 ± 3.13	3.15 ± 2.48
avg. pizza	94 ± 48	7.74 ± 3.73	0.34 ± 0.24	3.98 ± 2.05	1.94 ± 0.90	1.10 ± 0.47
avg. chick. tenders	134 ± 18	19.89 ± 6.15	0.63 ± 0.81	4.54 ± 2.45	7.36 ± 1.86	6.49 ± 4.03
avg. french fries	135 ± 49	19.39 ± 6.07	0.66 ± 1.11	3.75 ± 1.71	8.01 ± 4.48	6.11 ± 2.89
avg. apple pie	116 ± 43	15.59 ± 6.21	0.24 ± 0.20	6.17 ± 1.42	5.05 ± 2.13	3.44 ± 3.13

[a]Values represent the means ± SDs of two independent extractions for each sample analyzed on the 100 m SP-2560 CPS column according Method Ce 1j-07. [b]*trans* Fat was calculated based on the sum of all FA containing one or more isolated (i.e., nonconjugated) *trans* double bonds.

tenders/nuggets, French fries, and apple pie/turnover samples, the ratio of *trans* MUFA to the sum of *trans* 18:2 plus *trans* 18:3 FA was less than 1.0, reflecting the typical content of *trans* PUFA and low levels of *trans* MUFA in oils that have undergone deodorization but not partial hydrogenation.[7] In other samples, the ratio of *trans* MUFA to the sum of *trans* 18:2 plus *trans* 18:3 FA was greater than 1.0 and could be explained by the use of partially hydrogenated oil in the preparation of these foods. These results reveal the importance of the ratio of *trans* MUFA to *trans* 18:2 plus *trans* 18:3 FA for identifying partially hydrogenated oils in nonruminant-derived foods and emphasize the advantage of the 200 m SLB-IL111 ionic liquid column in making these determinations.

The separation of 18:2 FAME by GC methods has been problematic, especially for partially hydrogenated vegetable oils[36] and ruminant-derived samples[23,37] that contain a range of methylene- and non-methylene-interrupted *cis* and *trans* 18:2 FA isomers. These 18:2 FAME show coelutions among themselves and with other FAME, including *cis*-16 18:1, *cis* and *trans* 19:1 FA, and cyclic FAME, when analyzed on 100 m CPS columns. In the present study, separations achieved with the 200 m SLB-IL111 column resulted in significantly higher levels of *trans* 18:2 FA when samples contained less than 0.5% *trans* 18:2 FA. However, when the content of *trans* 18:2 FA was greater than 0.5% of total FA, analysis according to Method Ce 1j-07 produced significantly higher values. This overestimation in *trans* fat content may be explained by the coelution of *trans* 18:2 FAME with other non-*trans* FAME on the SP-2560 column, while on the 200 m SLB-IL111 column, these FAME isomers could be still be resolved.

Overall, results from the present study confirm the importance of Method Ce 1j-07 for the quantitation of total *trans* fat in foods and highlight the advantage of the 200 m SLB-IL111 ionic liquid column for the complementary determination of the content and isomeric composition of *trans* MUFA and *trans* PUFA. Two major advantages of the 200 m SLB-IL111 column, relative to the SP-2560 column, were the improved separation of positional isomers of *trans* 18:1 FAME and the resolution of *trans* 18:2 and *trans* 18:3 FAME isomers. As the restaurant industry continues to explore reformulation options for oils used in the production and preparation of fast

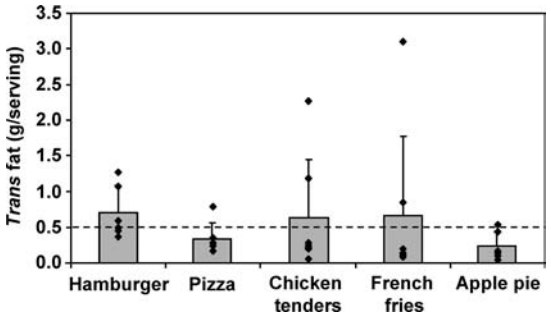

Figure 4. Grams per serving levels of *trans* fat in the five fast food categories. Bars represent the means ± SDs for all determinations within a food category, analyzed on the 100 m SP-2560 CPS column according to Method Ce 1j-07. Individual points represent the mean of two determinations for each food item (n = 6–7 per food category). The cutoff value of 0.5 g per serving, below which conventional foods may be labeled as containing 0 g of *trans* fat, is depicted by a dashed line.

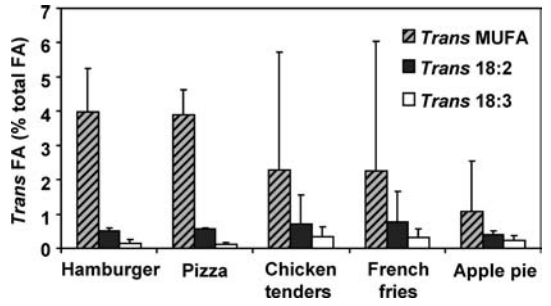

Figure 5. Content of *trans* MUFA, *trans* 18:2, and *trans* 18:3 FA as a percentage of total FA in each of the five fast food categories. Values represent the means ± SDs for samples within each category (n = 6–7 per food category), analyzed on the 200 m SLB-IL111 according to Delmonte et al.[21] The content of *trans* MUFA was based on the sum of 14:1, 15:1, 16:1, 17:1, 18:1, and 20:1 *trans* FA.

food items, with particular interest in removing high levels of *trans* fat from these foods, the importance of the contribution of *trans* 18:2 and *trans* 18:3 FA to the total *trans* fat content and the need for the accurate quantitation of these *trans* PUFA will be increasingly important. Assessment of the content of *trans* PUFA in foods is important considering their adverse effects on cardiovascular disease risk factors and PUFA biosynthesis.

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Notes

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[§]Retired.

ABBREVIATIONS USED

AOCS, American Oil Chemists' Society; CLA, conjugated linoleic acid; CPS, cyanopropyl polysiloxane; FA, fatty acid; FAME, FA methyl ester; GC, gas chromatography; MUFA, monounsaturated FA; NLEA, Nutrition Labeling and Education Act; Phy, methyl phytanate; PUFA, polyunsaturated FA; SFA, saturated FA

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