A Rapid, Automated Method for the Determination of *cis* and *trans* Content of Fats and Oils by Fourier Transform Infrared Spectroscopy

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ABSTRACT: A rapid Fourier transform infrared (FTIR) method was developed to simultaneously determine percent *cis* and *trans* content of edible fats and oils. A generalized, industrial sample-handling platform/accessory was designed for handling both fats and oils and was incorporated into an FTIR spectrometer. The system was calibrated to predict the *cis* and *trans* content of edible oils by using pure triglycerides as standards and partial least squares as the chemometric approach. The efficacy of the calibration was assessed by triglyceride standard addition, by mixing of oils with varying *cis/trans* contents, and by analyzing fats and oils of known iodine value. Each of the approaches verified that the FTIR method measured the *cis* and *trans* content in a reproducible (±0.7%) manner, with the measured accuracies being 1.5% for standard addition and 2.5% for the chemically analyzed samples. Comparisons also were made to the conventional American Oil Chemists' Society (AOCS) method for the determination of *trans* isomers by IR spectroscopy. The FTIR–partial least squares approach worked well over a wide range of *trans* contents, including those between 0 and 15%. The sample-handling accessory designed for this application is robust, flexible, and easy to use, being particularly suited for quality-control applications. In addition, the analysis was automated by programming the spectrometer in Visual Basic (Windows), to provide a simple, prompt-based user interface and to allow an operator to carry out *cis/trans* analyses without any knowledge of FTIR spectroscopy. A typical analysis requires less than two minutes per sample. The derived calibration is transferable between instruments, eliminating the need for recalibration. The integrated analytical system provides a sound basis for the implementation of FTIR methods in place of a variety of AOCS wet chemical methods when analytical speed, cost, and environmental concerns are issues.
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KEY WORDS: *cis*, FTIR spectroscopy, lipid analysis, *trans*, *trans* analysis.

Edible fats and oils are, by definition, either solid or liquid at room temperature, their physical state being defined by their triglyceride makeup, relative degree and form of unsaturation (*cis* or *trans*), weight-average molecular weight or saponification number (SN), and overall fatty acid composition/distribution. Of these complex determinants that define the physical state of a triglyceride lipid system, reducing the degree of unsaturation is the most common means used by industry to convert oils into solid fats. The majority of the unsaturated fatty acids that make up edible oil triglycerides are normally found in the *cis* form. When oils are hardened by hydrogenation for their use in formulating margarine and shortenings or partially hydrogenated to stabilize oils to oxidation, there is the concurrent conversion of *cis* to *trans* double bonds. As *trans* fatty acids have a higher melting point, they also contribute to the net hardening effect. However, the presence of substantial amounts of *trans* fatty acids has become controversial due to their association with arteriosclerosis and heart disease (1). The levels of *trans* fatty acids can reach values of 40% or more in hardened fats, and legislation is being considered by the United States Food and Drug Administration to require the labeling of the *trans* content of hydrogenated fats and oils. As such, it is becoming imperative for processors to be able to rapidly determine the *trans* and/or *cis* content of processed fats and oils, so that they can better control the hydrogenation process.

The McGill FTIR Group has been working on the development of new, rapid, and accurate methods for food and edible oil analysis based on Fourier transform infrared (FTIR) spectroscopy (2). This technology is a major advance over conventional dispersive-based IR spectroscopy (3) and, being based on interferometry, provides enhanced energy throughput and a better signal-to-noise ratio, and incorporates substantial computing, chemometric, and macro-programming capabilities. As such, FTIR instruments can be pre-programmed and automated to carry out analyses on a routine basis.

Edible oils are ideal candidates for quality-control applications of FTIR spectroscopy. To date, methods have been developed for iodine value (IV) and SN (4), free fatty acids (5), and peroxide value (6), with the spectral groundwork having been laid for the comprehensive assessment of the oxidative state of oils (7). Our overall objective is to develop pre-programmed analytical systems that are capable of carrying out routine edible oil analyses in less than three minutes per samples. In our work to date, the ever-present bottleneck has been the lack of a suitable sample-handling system, as no commercial system is available that can handle both fats and oils. This

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paper describes a generalized industrial sample-handling platform, designed specifically for the analysis of fats and oils, and the development of an automated FTIR method for the rapid determination of their *cis* and *trans* contents.

Current status of trans *analyses.* The American Oil Chemists' Society (AOCS) officially recognizes three methods (8) for the analysis of *trans* content of fats and oils, one being specific for margarine. Two of these methods employ gas chromatography (GC) (Cd 17-85 and Ce 1c-89), and the third measures isolated *trans* isomers by conventional IR spectroscopy (Cd 14-61) (8). Both gas-chromatographic methods allow for the identification of individual *trans* as well as *cis* fatty acids; however, depending on the degree of separation of the various isomers, peak overlap can lead to an underestimation of $C_{18:1t}$ (9). Overall, capillary GC is a useful primary method but is not particularly suited to routine quality-control applications in a process environment.

The AOCS IR spectroscopic method is based on measuring the peak height of the isolated *trans* band at ~10.3 μm (970 cm^{-1}) and comparing it to that measured in the spectrum of either elaidic acid (when *trans* content >15%) or methyl elaidate (when *trans* content <15%) standards. From a practical standpoint, the AOCS method has a number of limitations; i.e., it requires the use of CS_2 (volatile and unpleasant), and saponification and methylation of the oil are required for *trans* contents <15% because triglycerides exhibit a broad IR absorption band that overlaps with the *trans* band, which affects quantitation (10). Lanser and Emken (11) developed an FTIR method based on measurement of the area of the *trans* peak and demonstrated good agreement with the results obtained by capillary gas chromatography. Sleeter and Matlock (12) developed an FTIR procedure for measuring the *trans* content of oils directly in their neat form in a 100-μm KBr transmission cell, eliminating the need for CS_2. Ulberth and Haider (13) used *trans*-free methylated soybean oil mixed with methyl elaidate in combination with spectral subtraction techniques to accurately measure low *trans* contents. Partial least squares (PLS) chemometric procedures also were used; however, the analytical results were not validated.

Clearly, researchers have been addressing the issue of *trans* determination by FTIR spectroscopy; however, practical sample-handling complications, unfamiliarity of the industry with FTIR methods, and the general lack of expertise in spectroscopy has effectively stifled the routine application of FTIR spectroscopy to oil analysis. Of these impediments, sample handling is the first hurdle to overcome, as the analysis *per se* and the spectroscopic aspects can readily be automated by macro-programming.

MATERIALS AND METHODS

Sample-handling system platform. Based on our experience over the past several years in working on FTIR oil analysis, it has become evident that a rugged sample-handling platform is required if FTIR methods are to be routinely employed in quality control of fats and oils. For this purpose, a general fats and oils sample-handling prototype was designed and manufactured in cooperation with Dwight Analytical Solutions Ltd. (Toronto, Ontario, Canada), amenable for "on line" or laboratory use. Figure 1 illustrates the basic FTIR spectrometer, the computer that controls the spectrometer, a temperature controller and sample-handling accessory inlet, and control valve. Figure 2 illustrates a side view of the accessory within the sample compartment of the spectrometer and shows the cell and its housing, the latter being composed of a temperature-control block and a removable cell insert. The insert allows for ready removal of the cell to take an open beam background or to interchange cells if a change in cell configuration (i.e., window, pathlength) is required for a particular analysis. The cell insert (not shown) is designed with a spring-loaded face plate, which makes it readily demountable, allowing one to change windows or spacers as needed. Figure 3 presents a schematic diagram of the flow path through the cell and its housing, showing the inlet line, a three-way valve used to direct the oil flow, and an outlet line that empties into a collection vessel (not shown), which in turn is connected to a vacuum line, used to aspirate the sample through the IR transmission flow cell (Dwight Analytical Solutions Ltd., Toronto, Ontario). All components of the accessory are heated (usually to 80 ± 0.2°C) so that fats will be in their liquid form and will flow without crystallization in the lines or cell. The system includes a bypass line to flush out the bulk of the previous sample, avoiding large sample volumes passing through the IR cell and minimizing sample

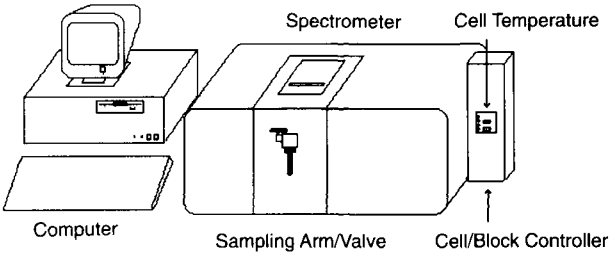

FIG. 1. A schematic diagram of the Fourier transform infrared spectrometer, personal computer, temperature-control unit, and sample compartment with oil sample inlet and valve.

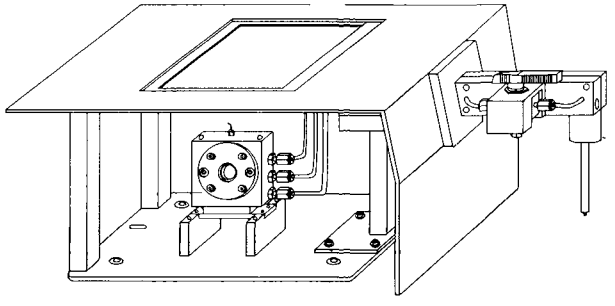

FIG. 2. Oil analysis accessory (side view), illustrating the cell, heated inlet and outlet lines.

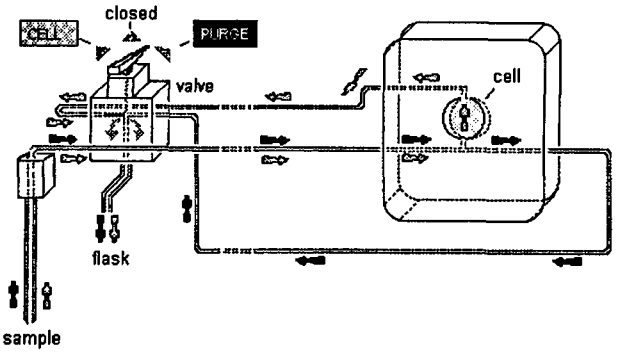

FIG. 3. A schematic diagram of the cell and flow pattern through the system.

cross-contamination. As configured, a fat or oil sample is either heated in the test tube block or pre-warmed in a microwave oven, presented to the input line and aspirated into the cell using the three-way valve. The system as configured was used throughout this study.

IR spectroscopy. IR spectroscopy was carried out with a Nicolet "Impact 400" FTIR spectrometer interfaced to a 486/33MHz PC, which operated under Windows®-based Nicolet Omnic 2.1® software (Nicolet Instrument Corp., Madison, WI) (14). The instrument and sample compartment were purged with dry air from a Balston dryer (Balston, Lexington, MA) to minimize water vapor and CO_2 interferences. The spectrometer was equipped with the sample-handling accessory described previously, operated at 80 ± 0.2°C. A cell with NaCl windows and a nominal pathlength of 25 μm was placed in the cell insert.

Reagent-grade triglyceride standards (purity >98%) tricaproin (C_6), tricaprylin (C_8), tricaprin (C_{10}), trilaurin (C_{12}), tripalmitin (C_{16}), tripalmitolein ($C_{16:1c}$), tristearin (C_{18}), triolein ($C_{18:1c}$), trielaidin ($C_{18:1t}$), trilinolein ($C_{18:2c}$), trilinolelaidin ($C_{18:2t}$), trilinolenin ($C_{18:3c}$), and trierucin ($C_{22:1c}$) were obtained from Sigma Chemical Company (St. Louis, MO). These standards were selected to cover a range of molecular weights (SN) and degrees of unsaturation, including both *cis* and *trans* isomers. The *cis* and *trans* contents of the standards were expressed as % triolein and % trielaidin, with $C_{18:1c}$ and $C_{18:1t}$ representing 100% *cis* and *trans*, respectively, and with $C_{18:2c}$, $C_{18:3c}$, and $C_{18:2t}$ having values of 200% *cis*, 300% *cis*, and 200% *trans*, respectively.

Calibration standards were loaded into the cell with a microsyringe because the standards were expensive and available only in limited quantity. The same procedure was employed for samples prepared by standard addition. For the analysis of the other samples, sample handling consisted of warming the sample in a microwave oven to within 5°C of operating temperature and aspirating the sample into the cell (~5 mL to flush the bypass and 2 mL to load the cell), recording its spectrum (128 scans, resolution of 4 cm^{-1} at a gain of 1.0), evacuating the sample, and loading the subsequent sample in the same manner. The cell was only cleaned prior to and at the end of an analytical run with isooctane. Spectra were collected under program control by using 512 scans for calibration and 32 scans for analysis.

PLS calibrations were derived for *cis* content, *trans* content, IV, and SN; the basic principles of the calibration have been described in previous publications (4–7). Spectral mixtures generated from the spectra of the 13 base triglycerides to obtain a wider range of *cis* and *trans* contents than were available from the unsaturated triglyceride standards also were included in the calibrations. The PLS calibrations were validated by the "leave one out" cross validation procedure and incorporated into a master program written in Visual Basic, which drove the spectrometer, performed all calculations required and printed the results. The program also included a Windows-based operator interface, which prompted the user on how to proceed with the analysis.

For the analysis of unknowns, CanAmera Foods Ltd. (Toronto, Canada) supplied a selection of vegetable oils, shortenings, and other hydrogenated fats, pre-analyzed for IV by the standard AOCS chemical method. These samples were analyzed by FTIR under program control to provide *cis* and *trans* data directly. The total unsaturation (Σ*cis* + *trans*) was related to IV. Quantitative standard addition also was carried out on a micro-scale by adding pure *cis* and *trans* unsaturated triglycerides to selected fats and oils to determine whether the calibration responded quantitatively to precise gravimetric changes. For comparative purposes, the instrument also was calibrated by a simple modification of the AOCS method, with pure trielaidin as a standard (100% *trans*) by measuring the peak height at 971 cm^{-1} (10.3 μm) relative to a baseline drawn between 995 cm^{-1} (10.05 mm) and 937 cm^{-1} (10.67 μm). This is effectively a parallel form of the AOCS method, but is suitable for oils in their neat form.

RESULTS AND DISCUSSION

Figure 4 presents the overlaid spectra of six triglycerides ($C_{18:0}$, $C_{18:1c}$, $C_{18:2c}$, $C_{18:3c}$, $C_{18:1t}$, and $C_{18:2t}$) and illustrates the *cis* and *trans* absorption bands. *Trans* fatty acids exhibit a characteristic absorption at ~970 cm^{-1}, whereas *cis* fatty acids are characterized by a distinctive band at ~3010 cm^{-1}, due to the CH double bond stretching absorption. *Trans* fatty acids also exhibit a weaker CH stretching absorption at ~3025 cm^{-1}. Figure 5 presents a more detailed view of the variability introduced in the *cis* and *trans* regions when triglycerides are mixed spectrally. This figure illustrates some of the difficulties that one would encounter by using a simple peak height or area measurement to determine the *cis* or *trans* content of an oil. First, the extinction coefficient of the characteristic *trans* band for $C_{18:2t}$ is ~1.7 times that for $C_{18:1t}$ rather than 2.0 (Fig. 4). Furthermore, there is a slight shift in the peak maximum on going from $C_{18:1t}$ (966.3 cm^{-1}) to $C_{18:2t}$ (967.8 cm^{-1}), as illustrated in Figure 5B. Thus, in samples containing $C_{18:2t}$, the overall *trans* content would be overestimated by the peak height method. Similar effects are ob-

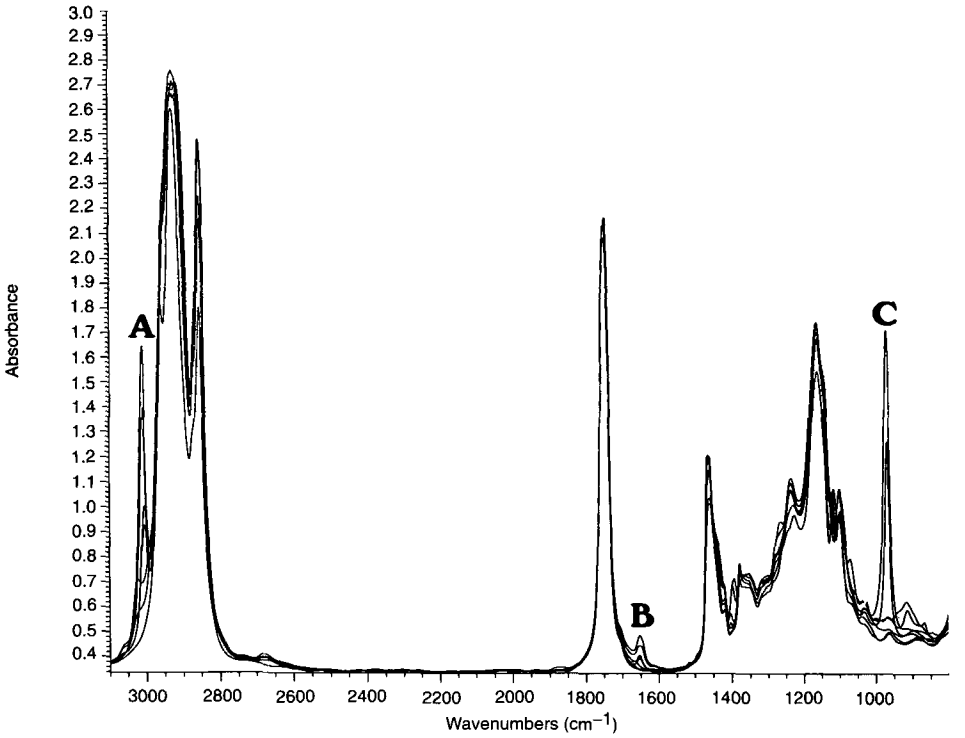

FIG. 4. Overlaid spectra of $C_{18:0}$, $C_{18:1c}$, $C_{18:2c}$, $C_{18:3c}$, $C_{18:1t}$, and $C_{18:2t}$, illustrating the major bands of interest related to *cis* and *trans* analysis in edible oils: (A) *cis* C-H stretching absorption; (B) C=C stretching absorption; (C) *trans* C=C-H bending absorption.

served for the *cis* band (Fig. 5A), which shifts from 3004.8 cm^{-1} for $C_{18:1c}$ to 3009.8 cm^{-1} for $C_{18:2c}$, and 3011.4 cm^{-1} for $C_{18:3c}$. Secondly, in the CH stretching region, the *cis* band partially overlaps the *trans* band, while in the *trans* region, triglyceride contributions interfere with the measurement of the peak height of the *trans* band, and these contributions vary with SN (molecular weight).

PLS is the chemometric method of choice when such interferences are present. The power of PLS is based on its ability to make use of spectral information from broad spectral regions, rather than peak height or peak area measurements, and to mathematically correlate spectral changes to changes in the concentration of a component of interest, while simultaneously accounting for other spectral contributions that may perturb the spectrum (15). As such, a PLS calibration model is capable of delivering accurate and reproducible results as long as the calibration spectra contain enough information that is representative of both the component of interest and the nonrelated spectral variations associated with the samples to be analyzed. PLS calibration models were developed based on a calibration set that included both $C_{18:1t}$ and $C_{18:2t}$ and five *cis* isomers ($C_{16:1c}$, $C_{18:1c}$, $C_{18:2c}$, $C_{18:3c}$, and $C_{22:1c}$) as well as six saturated triglycerides that covered a broad range of SNs. This approach allows the spectral shifts between the different *cis* and *trans* forms to be accounted for in the calibrations. Furthermore, as the PLS procedure makes use of broad spectral regions in determining *cis* and *trans* contents, the spectral contribution of the weak *trans* (3025 cm^{-1}) band in the *cis* region is included, as is that of a weak *cis* (913 cm^{-1}) band in the *trans* region. Last, but not least, by incorporating the effect of SN by including saturated triglycerides in the calibration set, the PLS model accounts for the contributions of the underlying triglyceride absorptions that lead to errors in the AOCS method. Our initial calibrations were based on some 70 spectra in all, the 13 base triglyceride spectra plus 57 spectral mixtures produced by co-adding various proportions of the base spectra to provide a wider range of *cis* and *trans* values. The additional information generated from the co-added spectra was found to be redundant in improving the calibration, and the 13 base standards sufficed. However, the co-added spectra were useful for comparative, illustrative, and interpretational purposes. Excellent calibrations were obtained for *cis* and *trans* determinations as well as for SN and IV (not discussed). These calibrations were validated by the "leave one out" cross-validation technique, with the mean errors obtained from the cross validation being 0.10 and 0.05% for *cis* and *trans*, respectively.

Analyses were initially carried out on commercially available fats and vegetable oils to determine the efficacy of the sample-handling system and its reproducibility. The sample-handling prototype proved to be functional, rugged, reliable, and easy to use, effectively overcoming all the common sample-handling problems encountered in our previous work. At 80°C, even hardened fats were liquid and flowed well through the system. Flow through both the bypass line and the cell was smooth, and no cell loading or cross-contamination problems were encountered. Table 1 provides an assessment of analytical reproducibility (16) in terms of the mean difference

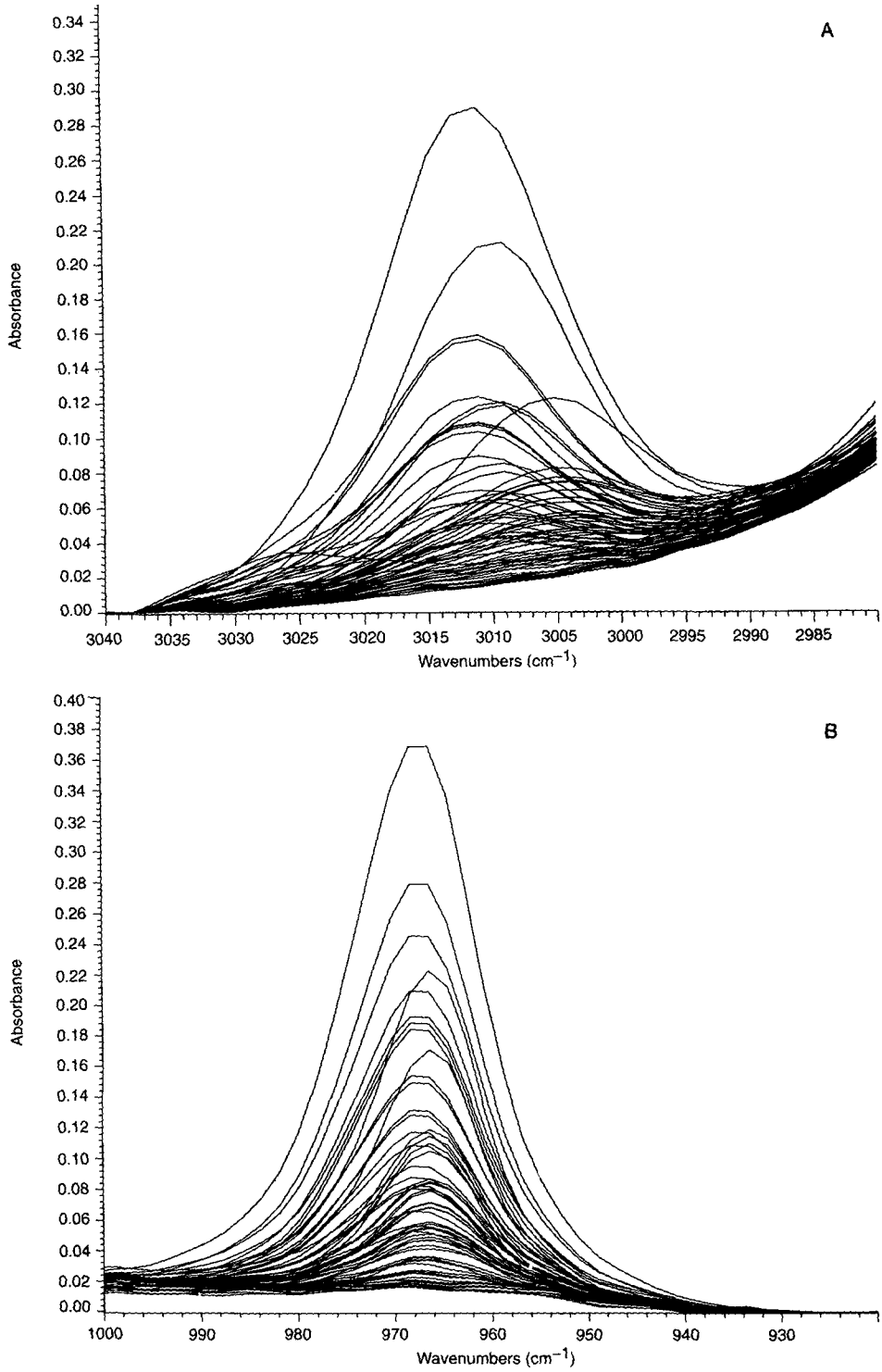

FIG. 5. Detail of the *cis* and *trans* regions of co-added spectra, illustrating the spectral variability in these regions.

(MD_r) and the standard deviation of the difference (SDD_r) for *cis*, *trans*, IV, and SN predictions for 20 sample pairs run one week apart. Hence, consistent results were obtained in terms of the overall means, regardless of the sample, with the variability around the mean difference being about ±0.25% for the *cis* and *trans* data. This type of consistency was the norm and could be maintained over time.

Subsequent experiments involved the standard addition of *cis* and *trans* triglycerides in mg amounts to small amounts of selected oils to determine if appropriate quantitative changes were predicted, in effect a measure of the accuracy of the analysis. Table 2 presents the data obtained in terms of accuracy (MD_a and SDD_a) for the standard addition experiments. The standard addition results indicate that, on average, the predic-

TABLE 1
The Mean Difference (MD_r) and the SD of the Difference (SDD_r) for 20 Sample Pairs of Various Oils Run One Week Apart

Statistic/sample	*cis*	*trans*	IV[a]	SN[a]
MD_r	−0.69	0.14	−0.77	−0.19
SDD_r	0.23	0.23	0.52	0.22

[a]For information only, not discussed in text; IV, iodine value; SN, saponification number.

TABLE 2
MD_a and SDD_a for 15 Samples[a]

Statistic/sample	*cis*	*trans*	IV[b]	SN[b]
MD_a	0.30	−0.69	0.48	0.06
SDD_a	1.31	1.25	1.61	0.53

[a]The Fourier transform infrared predictions compared to calculated change expected in the values based on the standard addition of pure *cis/trans* triglycerides.
[b]For information only, not discussed in text. Abbreviations as in Table 1. MD_a and SDD_a, mean difference and SD of difference in terms of accuracy.

tions matched the calculated values quite well. Subsequently, similar experiments were carried out by mixing selected oils in bulk in gravimetrically precise proportions to eliminate the sources of variability associated with weighing and mixing on the small scale required for standard addition and loading the cell with a microsyringe. Similar results were obtained (Table 2), indicating that the standard addition results were representative of the performance one could expect from FTIR analysis. Hence, the PLS calibration was shown to respond quantitatively to the direct addition of *cis* and *trans* moieties to diverse oils, as well as to the mixing of different fats and oils. These results imply that the spectra of only 13 pure triglycerides provide sufficient information to quantitate complex triglyceride systems in terms of their *cis* and *trans* content.

To further determine the universality of the calibration, additional FTIR analyses were carried out on 31 fats and oils, supplied and independently analyzed for their IV by CanAmera Foods Ltd. The resulting *cis* and *trans* predictions are tabulated in Table 3 along with their $\Sigma cis + trans$ plus their chemical IV. Because specific *cis* and *trans* data were not available for these samples, the results were analyzed by using IV as a basis for assessing the predictions. IV represents the sum of the *cis* and *trans* double bond contributions, which can be re-expressed in terms of % triolein and % trielaidin as follows:

$$IV = 3[(C + T)\ (\text{Mw } I_2/\text{Mw TG})] \quad [1]$$

$$IV = 0.8601\ (C + T) \quad [2]$$

where *C*, % *cis* (as triolein); *T*, % *trans* (as trielaidin); Mw, molecular weight; TG, triglyceride (triolein or trielaidin); and I_2, iodine.

Based on this relationship, one would expect the FTIR-predicted $\Sigma cis + trans$ to be linearly related to the IV if the instrumental predictions are on track. Figure 6 presents a plot of the $\Sigma cis + trans$ vs. IV for the data presented in Table 3.

TABLE 3
FTIR-Predicted *cis* and *trans* Contents and Chemical IVs for Samples of Fats and Oils (coded samples were unspecified in terms of makeup)[a]

Fat or oil	% *cis*	%*trans*	Σ*C/T*	Chemical IV
Corn	148.7	0.1	148.8	128.1
Olive	92.6	3.4	96.0	86.9
Safflower	167.4	0.9	168.3	146.6
Lard 1	69.6	2.0	71.6	60.1
Sesame	130.3	2.4	132.7	110.6
Coconut	11.3	−2.0	9.3	8.7
Peanut	109.4	0.2	109.6	95.6
Lard 2	75.4	1.7	77.1	62.0
Cottonseed	123.2	0.5	123.7	105.7
Canola	131.7	0.5	132.2	116.0
Soybean	151.0	0.0	151.0	129.3
Tallow	45.4	7.2	52.6	43.2
Palm kernel oil	22.0	−2.5	19.5	17.3
Sample 870 ST-4	49.8	34.4	84.2	72.7
Sample 869 ST-5	116.5	14.2	130.7	109.6
Sample 469 ST-6	64.6	31.9	96.5	80.7
Sample 460 ST-6	36.0	44.5	80.5	71.0
Sample 866 ST-4	115.5	13.9	129.4	107.3
Sample 448 ST-5	45.8	41.7	87.5	75.3
Sample 473 ST-8	39.5	43.0	82.5	69.5
Sample 865 ST-5	98.9	21.0	119.9	102.2
Sample 864 ST-9	42.0	40.7	82.7	71.2
Majestic ST-8	49.7	36.5	86.2	74.0
RH 26039	52.0	37.3	89.3	76.8
ST-9	46.0	6.4	52.4	48.3
ST-7	59.2	32.0	91.2	77.7
ST-6	50.6	40.1	90.7	77.3
K	42.2	44.5	86.7	70.6
PBY 18844	38.4	46.0	84.4	73.0
EFFEM	36.5	46.2	82.7	74.3
JP	40.1	39.9	80.0	70.3

[a]All samples supplied by CanAmera Foods Ltd. (Toronto, Canada); FTIR, Fourier transform infrared; *C, cis; T, trans.* Other abbreviations in Table 1.

The plot illustrates a good linear correspondence between S FTIR $cis + trans$ and IV, with linear regression of these data producing the following relationship:

$$\Sigma cis + trans = -0.523 + (1.172)IV \quad r^2 = 0.997 \quad SD = \pm 2.60\% \quad [3]$$

The overall error of ~2.5% in terms of accuracy is reasonable because it includes the experimental error associated with the IV analysis itself plus that associated with the *cis* and *trans* predictions. The inverse of the slope of the plot (0.853) is in line with the theoretical conversion factor of 0.8601 (Eq. 2).

In Table 3, the range of *trans* content runs from negligible to ~46%. The only apparent problem with the data is some negative predictions associated with samples of high saponification numbers (palm and coconut). A similar, but more muted, tendency was seen in predictions of spectral mixtures of the calibrations standards that have *trans* values of zero, specifically mixtures corresponding to SN >250 (average chainlengths of ~C_{12}–C_{14}). To determine how the AOCS method would perform in this regard, the 70 ideal spectral mixtures were analyzed by the modified AOCS method, with trielaidin as a standard, and the results were plotted vs. the theoretical values for the spectral mixtures. Figure 7 shows only the lower portion of this plot (0–30% *trans*) and illus-

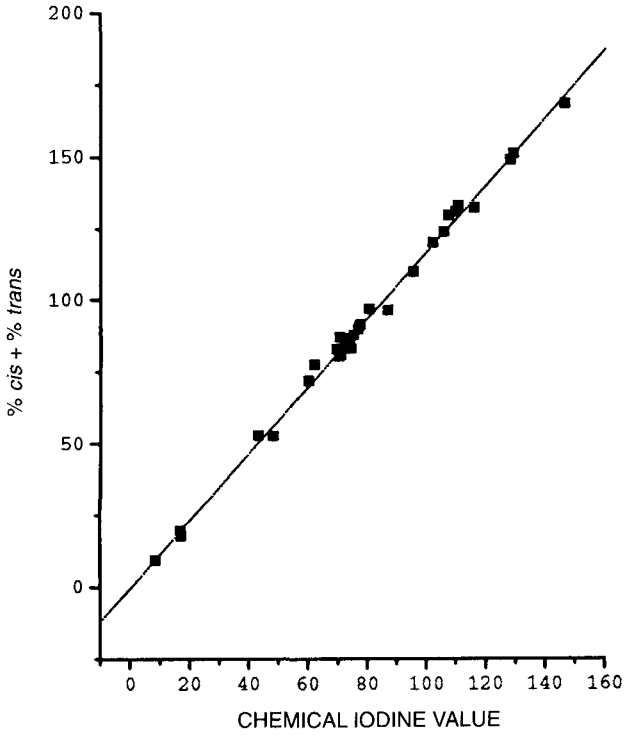

FIG. 6. Plot of the total degree of unsaturation (*cis* + *trans*) for the 29 CanAmera Foods Ltd. (Toronto, Canada) samples as determined by Fourier transform infrared analysis vs. their chemical iodine value.

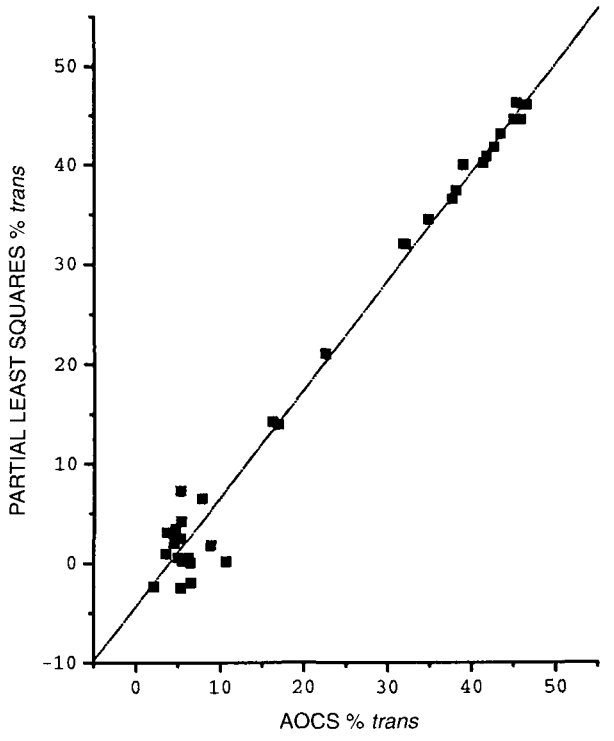

FIG. 8. Plot of predicted % *trans* obtained for the 29 CanAmera samples by the partial least squares (PLS)–Fourier transform infrared method vs. the predictions obtained from the modified American Oil Chemists' Society (AOCS) method with trielaidin as the standard. Company location as in Figure 6.

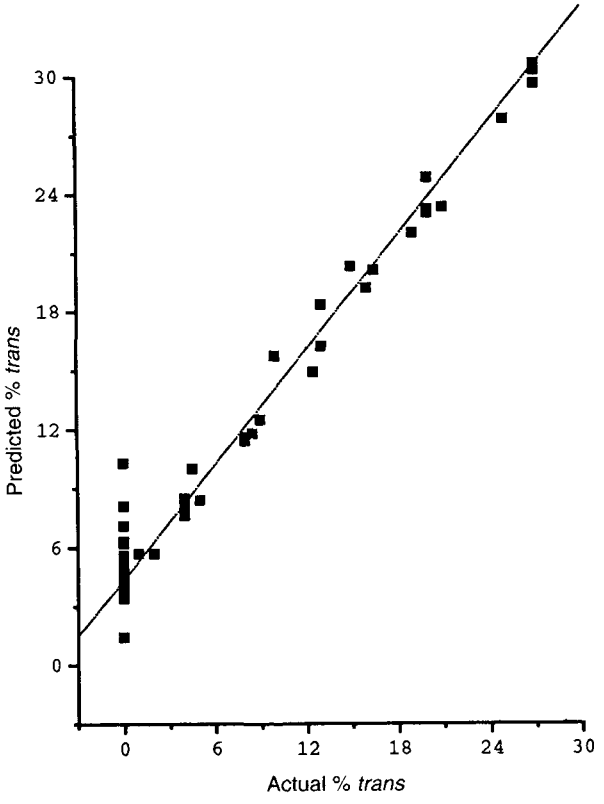

FIG. 7. Plot of predicted % *trans* obtained for co-added spectral mixtures of the base triglycerides by the modified American Oil Chemists' Society method using trielaidin as the standard vs. the theoretical % *trans*.

trates the effect that the SN of saturated triglycerides (zero *trans*) has on their predicted *trans* values. As such, the AOCS method cannot differentiate between fully saturated lipids and lipids with low *trans* contents, requiring the use of the methylation procedure for samples with *trans* contents of <15%. Methylation is not required in the PLS approach, which does well down to ~1% *trans*. Figure 8 shows a plot of the PLS-predicted values for the CanAmera samples vs. the values obtained by analyzing the same spectral information from the modified AOCS method with trielaidin as a standard. A good linear relationship holds between the FTIR–PLS and FTIR–AOCS methods above values of ~12%, but the relationship breaks down at lower *trans* values. Below 12% *trans*, the AOCS method again becomes limiting because it cannot account for underlying saturated triglyceride absorptions. The PLS calibration, on the other hand, holds because the Σcis + *trans* continues to correlate with the chemical IV values close to 0% *trans*. Hence, there is ample evidence that the PLS calibration is more robust and has fewer sources of variability and error relative to the AOCS method. In addition, the FTIR–PLS method is advantageous in terms of speed and accuracy and is able to determine both *cis* and *trans* content, as well as IV and SN, in a single analysis.

As structured, the FTIR method makes use of a generalized sample-handling accessory designed specifically for fats and oils. Sample handling is straightforward, and because the calibration is based on 13 commercially available triglycerides, the calibration is applicable to triglyceride-based oils in gen-

eral, barring any extensive degree of oxidation. Not discussed here are calibration transfer concepts, which have been developed to allow calibrations to be transferred between instruments, nor the programs written to automate and simultaneously allow for the analysis of *cis*, *trans*, IV, and SN. These additional elements of the analytical system effectively eliminate the need for calibration or for an operator to have any knowledge of FTIR spectroscopy. As such, the system is designed for industrial "on line" use in an oil-processing environment, where the operator is prompted to present the sample to the instrument by the computer controlling the spectrometer, and is able to obtain process results within 1–2 min. This system is the first step in the development of an integrated sample-handling platform for FTIR edible oil analysis, with transferable calibrations, computerized automation, and the appropriate use of chemometrics. Such a system provides an opportunity for replacing a host of tedious AOCS wet chemical methods, many of which are starting to be affected by environmental concerns.

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