

Detection of Lard Mixed with Body Fats of Chicken, Lamb, and Cow by Fourier Transform Infrared Spectroscopy

Y.B. Che Man* and M.E.S. Mirghani

Department of Food Technology, Faculty of Food Science and Biotechnology, Universiti Putra Malaysia, 43400 UPM, Serdang, Selangor, Malaysia

ABSTRACT: Fourier transform infrared (FTIR) spectroscopy provides a simple and rapid means of detecting lard blended with chicken, lamb, and cow body fats. The spectral bands associated with chicken, lamb, and cow body fats and their lard blends were recorded, interpreted, and identified. Qualitative differences between the spectra are proposed as a basis for differentiating between the pure animal fats and their blends. A semiquantitative approach is proposed to measure the percent of lard in blends with lamb body fat (LBF) on the basis of the frequency shift of the band in the region 3009–3000 cm^{-1} , using the equation $y = 0.1616x + 3002.10$. The coefficient of determination (R^2) was 0.9457 with a standard error (SE) of 1.23. The percentage of lard in lard/LBF blends was also correlated to the absorbance at 1417.89 and 966.39 cm^{-1} by the equations $y = 0.0061x + 0.1404$ ($R^2 = 0.9388$, SE = 0.018) and $y = 0.004x + 0.1117$ ($R^2 = 0.9715$, SE = 0.009), respectively. For the qualitative determination of lard blended with chicken body fat (CF), the FTIR spectral bands in the frequency ranges of 3008–3000, 1418–1417, 1385–1370, and 1126–1085 cm^{-1} were employed. Semiquantitative determination by measurement of the absorbance at 3005.6 cm^{-1} is proposed, using the equation $y = 0.0071x + 0.1301$ ($R^2 = 0.983$, SE = 0.012). The percentage of lard in lard/GF blends was also correlated to the absorbance at 1417.85 cm^{-1} ($y = 0.0053x + 0.0821$, with $R^2 = 0.9233$, SE = 0.019) and at 1377.58 cm^{-1} ($y = 0.0069x + 0.1327$, with $R^2 = 0.9426$, SE = 0.022). For blends of lard with cow body fat (CBF) bands in the range 3008–3006 cm^{-1} and at 1417.8 and 966 cm^{-1} were used for qualitative detection. The equation $y = -0.005x + 0.3188$ with $R^2 = 0.9831$ and SE = 0.0086 was obtained for semiquantitative determination at 966.22 cm^{-1} .

Paper no. J9636 in *JAOCs* 78, 753–761 (July 2001).

KEY WORDS: FTIR spectroscopy, chicken body fat, cow body fat, lamb body fat, lard.

Lipid is a major component of the human diet, contributing up to 40% of the calories in the diet of the developed countries. In the United Kingdom, meat lipids *in situ* or after rendering and processing constitute one-third of the total dietary lipid consumption (1). The Codex Alimentarius (2) specified that all edible animal fats must come from animals fit for human consumption and stated some analytical identity stan-

dards for four products from animal sources. Lard is commonly blended in edible oil/fat products like butter and shortening (3). Although a number of investigations have been carried out by several research groups to develop analytical methods for detection of animal body fats in ghee or butter (4–7), methods are available, and most are either difficult to perform or time-consuming.

DeMan (8) reported that the fatty acid composition of lard differed from that of cow body fat in $\text{C}_{16:1}$, $\text{C}_{18:3}$, $\text{C}_{20:0}$, and $\text{C}_{20:1}$, and from that of chicken fat in $\text{C}_{12:0}$, $\text{C}_{18:3}$, $\text{C}_{20:0}$, and $\text{C}_{20:1}$. Enser (1) reported that lard differed from lamb body fat in $\text{C}_{14:0}$, $\text{C}_{16:1}$, $\text{C}_{18:2}$, and $\text{C}_{18:1r}$. However, the differences are too small to allow fatty acid composition to be used as an indicator. In a few cases, methyl ester analysis by gas-liquid chromatography (GLC) can be employed to detect vegetable oil adulteration with animal fats by measurement of the $\text{C}_{17:0}$ and $\text{C}_{17:1}$ fatty acids. However, the GLC results should be interpreted with care, because a very few vegetable oils, such as Indian sesame seed oil, may contain $\text{C}_{17:0}$ or $\text{C}_{17:1}$ acids (9). DeMan (8) showed that lard and chicken fats were significantly different in the diunsaturated and triunsaturated triacylglycerols (TAGs). Because lard contains a high percentage of saturated fatty acids in the 2-position of the TAGs compared to other animal fats, determination of the fatty acids in the 2-position can be used to detect the presence of lard in other animal fats (10). TAG profiles obtained by high-pressure liquid chromatography (HPLC) may indicate adulteration of lard by beef tallow depending on the fatty acids in the 2-position of the TAGs [according to Kirk and Sawyer (11)]. Foreign fats can be determined in lard by the Boemer number method (12), which is based on the difference between melting points of TAGs and fatty acids, which is large for pure pork fat and small for beef tallow and similar fats. Haryati (13) found that differences in TAG group composition in fats are reflected on the differential scanning calorimetry (DSC) thermograms. Detection of animal body fat in ghee and butter using DSC has been reported by Lambelet (4) and Lambelet *et al.* (5) and by Coni *et al.* (6), respectively. A recent study by Tan and Che Man (14) showed that the DSC heating and cooling curves of edible oils can be used in qualitative and quantitative ways for identification of edible oils.

The use of Fourier transform infrared (FTIR) spectroscopy in the analysis of edible fats and oils has been described by

*To whom correspondence should be addressed. E-mail: yaakub@fsb.upm.edu.my

van de Voort and co-workers (15–18) and Che Man and co-workers (19–24). Guillen and Cabo (25) used FTIR spectroscopy to characterize edible oil and lard. The present study was conducted to investigate the possibility of detection of lard mixed with other animal fats such as lamb, chicken, and cow body fats using FTIR spectroscopic techniques.

MATERIALS AND METHODS

Sample preparation. Adipose tissues from various parts of slaughtered pigs (lard), cows, lambs, and chickens were obtained from a local market. The tissues were cut into small pieces, mixed, melted at 90–100°C (26), and strained through a triply folded muslin cloth. The melted fat was centrifuged at 3000 rpm (Kubota model 2010, Tokyo, Japan) for 20 min. The fat layer was decanted into a test tube containing anhydrous sodium sulfate (Na_2SO_4), shaken well, centrifuged again, and then decanted through Whatman filter paper containing anhydrous Na_2SO_4 . The filtered sample immediately analyzed or kept in a tightly closed container under a nitrogen blanket in the refrigerator.

Lard was mixed with each of the other melted animal fats [lamb body fat (LBF), cow body fat (CBF), and chicken body fat (CF)] by weight to cover the range of 0–35% lard. The pure fats and the blends were analyzed by means of FTIR spectroscopy.

Instrumentation/spectral acquisition. After vigorous shaking of each sample using an Autovortex Mixture SAI (Stuart Scientific, Redhill, United Kingdom), a few drops were sandwiched between clean and dry sodium chloride (NaCl) windows. A polytetrafluoroethylene (PTFE) spacer was placed between the windows to give a fixed pathlength of 0.1 mm. After the spectrum of the sample was scanned, the NaCl windows were rinsed at least three times with pure acetone and then dried. Spectral acquisition was carried out using a Perkin-Elmer 1650 series FTIR spectrophotometer (Perkin-Elmer Corporation, Norwalk, CT) equipped with a deuterated triglycine sulfate (DTGS) detector and connected to a Perkin-Elmer model 7300 professional computer operating under Infrared Data System (IRDM) software. FTIR data were collected by co-adding 64 scans at a resolution of 4 cm^{-1} with strong apodization over the region 4000–600 cm^{-1} . All spectra were ratioed against a background air spectrum and stored as absorbance values at each data point. The Windows™-based software program Spectrum Lite version 1.4 (Perkin-Elmer) was used to obtain the frequency of each band using the “label peaks” command of the software or using the vertical cursor by moving it to find the frequency at the maximum absorbance for the selected band. A simple Beer’s law approach was developed for mathematical treatment of the FTIR data. The assessment of accuracy was based on the standard error (SE) and the coefficient of determination (R^2) (27).

RESULTS AND DISCUSSION

Lamb body fat. Figure 1 shows the FTIR spectra of LBF and lard. The spectra illustrate the dominant spectral features as-

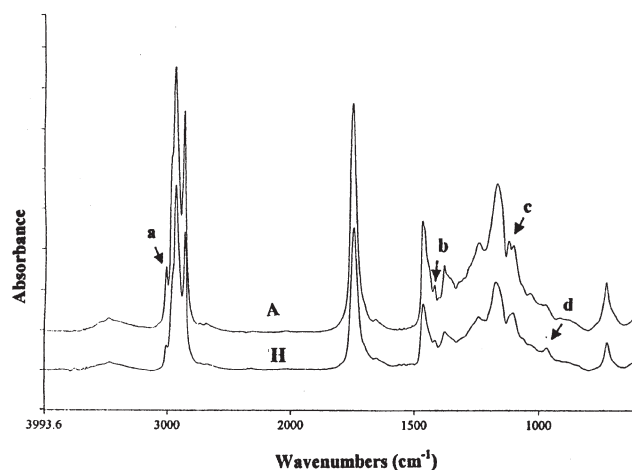


FIG. 1. Fourier transform infrared (FTIR) spectra of (A) pure lard and (H) pure lamb body fat (LBF). The labeled peaks are absorption bands that are significant in differentiating between lard and LBF.

sociated with fats and oils (28–29): CH stretching absorptions in the frequency range of 3050–2800 cm^{-1} , the carbonyl absorption of the triacylglycerol ester linkage at 1746–1744 cm^{-1} , and the bands associated with the fingerprint region (1500–1000 cm^{-1}). Differences between the raw spectra of LBF and lard are observed in four frequency ranges: 3009–3000, 1418–1417, 1119–1096, and 968–966 cm^{-1} . These four regions are illustrated in Figure 1 as a, b, c, and d, respectively.

(i) **Frequency range 3009–3000 cm^{-1} .** Figure 2 illustrates the difference between the spectra of LBF and lard in the frequency range 3009–3000 cm^{-1} (a in Fig. 1). The lard spectrum (A) has a sharp band at 3008.69 cm^{-1} , whereas the LBF spectrum (H) has a shoulder peak at lower frequency (3001.75 cm^{-1}). Spectra B–G in Figure 2 represent lard/LBF blends containing 5–30% lard. Bands in region a are due to

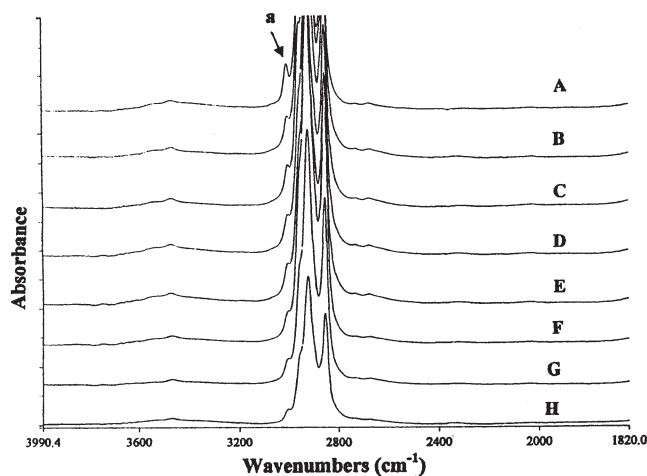


FIG. 2. FTIR spectra of (A) pure lard, (B)–(G) lard/LBF blends, and (H) pure LBF, illustrating changes in the frequency value of the band in the region 3009–3000 cm^{-1} (a, Fig. 1). The percentages of lard in the blends are (B) 30, (C) 25, (D) 20, (E) 15, (F) 10, and (G) 5%. See Figure 1 for abbreviations.

TABLE 1
Band Frequencies in the Fourier Transform Infrared (FTIR) Spectra of Lamb Body Fat (LBF), Lard and Their Blends in Region a (3009–3000 cm^{-1})

Spectrum ^a	% Lard	Frequency (cm^{-1}) ^b
A	100	3008.69 \pm 0.09
B	30	3006.38 \pm 0.08
C	25	3006.22 \pm 0.04
D	20	3005.58 \pm 0.04
E	15	3005.22 \pm 0.08
F	10	3003.48 \pm 0.08
G	5	3002.90 \pm 0.07
H	0	3001.75 \pm 0.05

^aLetters refer to spectra in Figure 2.

^bMean \pm standard deviation of two replicates.

the CH stretching vibration of *cis* double bonds (25, 30 and 31); accordingly, fats with a high proportion of linolenic or linoleic acyl groups show a higher frequency for this band than fats with a high proportion of oleic acyl groups. Thus, the frequency shifts in Figure 2 are consistent with the fatty acid compositions of lard and LBF (1), since LBF is high in oleic and lower in linoleic and linolenic compared to lard. A semiquantitative assessment is proposed based on the variation in frequency as a function of the percentage of lard in the blends with LBF. The data shown in Table 1 are plotted in Figure 3 and fitted by the equation $y = 0.1616x + 3002.1$ with an R^2 of 0.9457 and an SE of 1.23, where y is the frequency of the absorption maximum within the range 3009–3000 cm^{-1} , and x is percent lard in the blends with LBF (covering the range of 0–30%, w/w).

(ii) *Frequency range 1418–1417 cm^{-1} .* The FTIR spectrum of LBF has no peak in the frequency range 1418–1417 cm^{-1} whereas that of lard has a clear one as illustrated in Figures 1 and 4 (peak b). In the spectra of lard/LBF blends, this peak showed increasing in absorbance with increasing percent lard in the blend (Table 2). However, the arithmetic band (+) (the band of spectrum that appears as a result of adding spectrum of pure lard to that of LBF) has higher absorbance (0.39) than the pure lard band (A) (0.30), and the difference band (–) (the

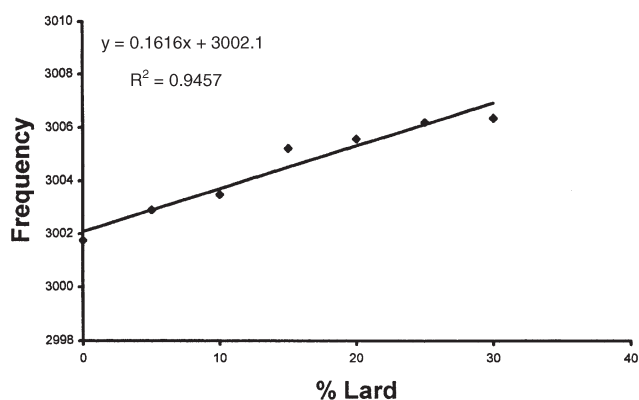


FIG. 3. Frequency value of band a (Figs. 1 and 2) in the FTIR spectra of LBF and lard/LBF blends containing up to 30% lard (w/w) vs. % lard. See Figure 1 for abbreviations.

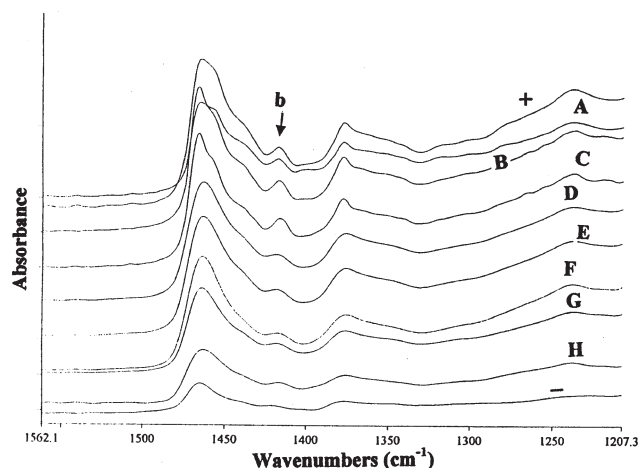


FIG. 4. FTIR spectra of (A) pure lard, (B)–(G) lard/LBF blends, and (H) pure LBF, illustrating changes in the absorbance value of the band in the region 1418–1417 cm^{-1} (b, Fig. 1). +, Arithmetic spectrum; –, difference spectrum. See Figure 1 for abbreviations; see Figure 2 for compositions of the blends.

band of spectrum that appears as a result of subtracting LBF spectrum from the pure lard) has lower absorbance (0.03) than the LBF band (H) (0.15), which may indicate the presence of fewer long-chain fatty acids in the composition of LBF than in lard. This result is consistent with the fatty acid compositions of these fats given in a review by Enser (1). Using the data in Table 2, the equation $y = 0.0061x + 0.1404$ was obtained for semiquantitative determination of percent lard in lard/LBF blends with an R^2 of 0.9388 and SE of 0.018 (Fig. 5).

(iii) *Frequency range 1119–1096 cm^{-1} .* In the frequency range of 1119–1096 cm^{-1} , lard showed two overlapping peaks having maxima at 1118.69 and 1100.03 cm^{-1} (c in Fig. 1). These peaks have been found to be inversely related to the proportion of saturated acyl groups and oleic acyl groups, respectively (28). Figure 6 shows that this region could be used for qualitative determination, whereby the spectra of lard/LBF blends containing more than 10% lard show two overlapping peaks, but those of blends containing 10% or less

TABLE 2
Absorbance Values in the FTIR Spectra of LBF, Lard, and Their Blends in Region b (1418–1417 cm^{-1})^a

Spectrum ^b	% Lard	Absorbance ^c
+	—	0.39
A	100	0.30 \pm 0.08
B	30	0.31 \pm 0.05
C	25	0.29 \pm 0.04
D	20	0.29 \pm 0.06
E	15	0.24 \pm 0.04
F	10	0.18 \pm 0.05
G	5	0.16 \pm 0.03
H	0	0.15 \pm 0.03
–	—	0.03

^aSee Table 1 for abbreviations.

^b+, arithmetic spectrum; –, difference spectrum. Letters refer to spectra in Figure 4.

^cMean \pm standard deviation of two replicates.

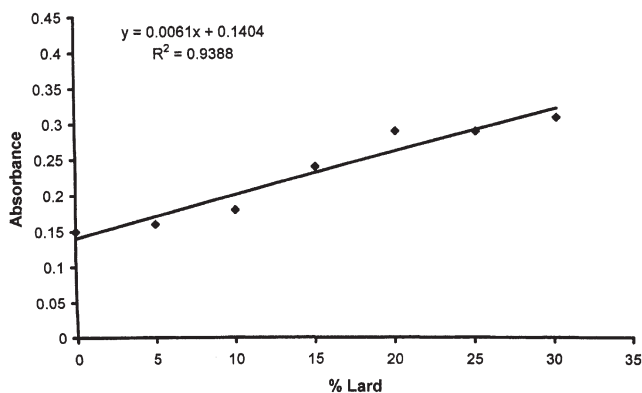


FIG. 5. Absorbance value of band b (Figs. 1 and 4) in the FTIR spectra of LBF and lard/LBF blends containing up to 30% lard (w/w) vs. % lard. See Figure 1 for abbreviations.

show one broad band as in the spectrum of pure LBF (H). However, as shown in Figures 6 and 7, the arithmetic spectrum (+) exhibits higher absorbance (0.72) than the spectrum of pure lard (A) (0.45), and the difference spectrum (-) shows lower absorbance (0.09) than the spectrum of pure LBF (H) (0.27). From the above results, it can be concluded that both lard and LBF contain saturated and oleic acyl groups in their structures, but the different relative proportions of these groups in the two fats allow for the qualitative determination of $10 \pm 3.0\%$ lard in LBF.

(iv) *Frequency range 968–966 cm^{-1}* . The LBF spectrum (H) showed a clear band at 966.39 cm^{-1} (peak labeled in Figs. 1 and 8), which is known to be due to the C=C-H bending vibration of *trans* double bonds (32,33), whereas the lard spectrum (A) has no clear band in this region. However, the absorbance in the lard spectrum is equal to or slightly higher than that in the pure LBF spectrum. The band in the same region $975\text{--}965 \text{ cm}^{-1}$ in the IR spectrum is the basis of the AOCS official method for determination of *trans* groups (34).

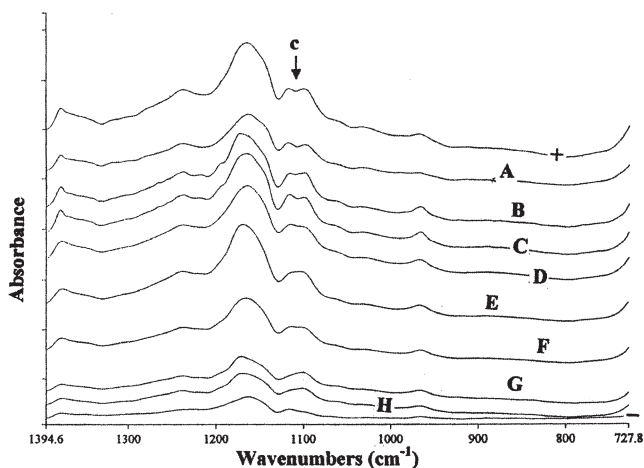


FIG. 6. FTIR spectra of (A) pure lard, (B)–(G) lard/LBF blends, and (H) pure LBF, illustrating changes in the absorbance value of the band in the region $1119\text{--}1096 \text{ cm}^{-1}$ (c, Fig. 1). +, Arithmetic spectrum; -, difference spectrum. See Figure 1 for abbreviations; see Figure 2 for compositions of the blends.

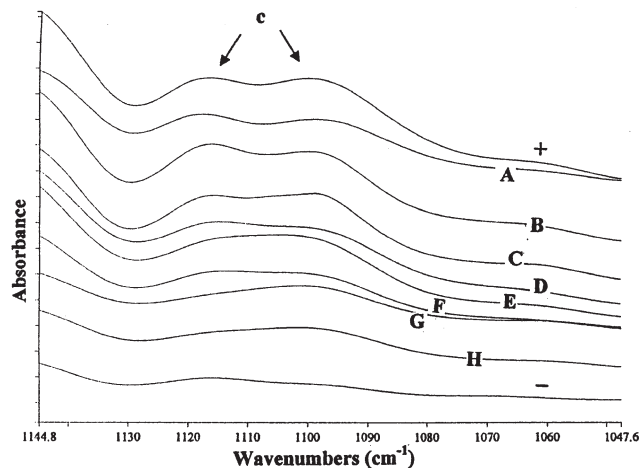


FIG. 7. Expanded view of region c in Figure 6.

As illustrated in Figure 8, the spectra of the lard/LBF blends showed higher absorbance than the pure LBF spectrum, which indicates that the lard contains some *trans* double bonds in its composition. The absorbance in this region could be used for semiquantitative determination of the percent lard in the lard/LBF blends using the following equation obtained from Figure 9 with an R^2 of 0.9715 and an SE of 0.009: $y = 0.004x + 0.1117$, where y is the absorbance at 966.39 cm^{-1} under the conditions of the test, and x is percent lard in the blend (range $\approx 0.0\text{--}35\%$ w/w).

The four frequency ranges mentioned above (a, b, c, and d) are illustrated in Figure 10, which shows the full spectra for all blends.

Chicken body fat (CF). Figure 11 shows the spectra of lard (A) and CF (P). They exhibit differences in four frequency ranges: a, $3008\text{--}3000 \text{ cm}^{-1}$; b, $1418\text{--}1417 \text{ cm}^{-1}$; e, $1385\text{--}1370 \text{ cm}^{-1}$; and c, $1126\text{--}1085 \text{ cm}^{-1}$.

(i) *Frequency range 3008–3000 cm^{-1}* . As illustrated in Figure 12, in the frequency range $3008\text{--}3005 \text{ cm}^{-1}$, the sharp

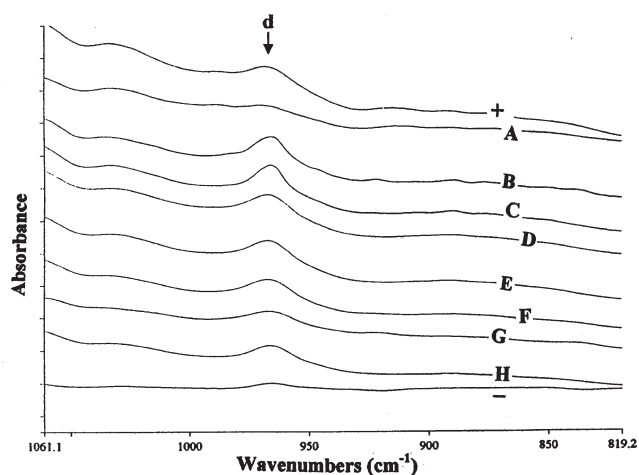


FIG. 8. FTIR spectra of (A) pure lard, (B)–(G) lard/LBF blends, and (H) pure LBF, illustrating changes in the absorbance value of the band in the region $968\text{--}966 \text{ cm}^{-1}$ (d, Fig. 1). +, Arithmetic spectrum; -, difference spectrum. See Figure 1 for abbreviations; see Figure 2 for compositions of the blends.

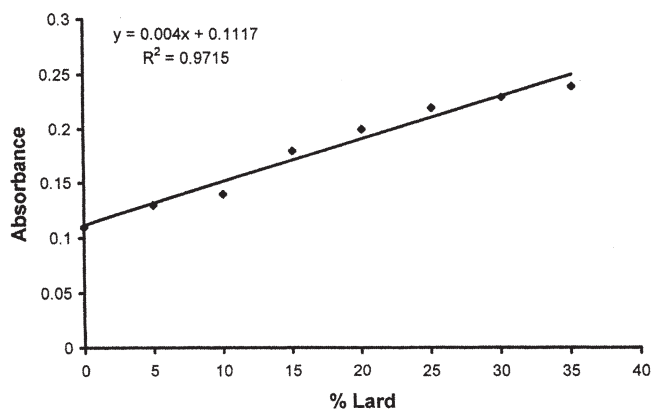


FIG. 9. Absorbance value of band d (Figs. 1 and 8) in the FTIR spectra of LBF and lard/LBF blends containing up to 30% lard (w/w) vs. % lard. See Figure 1 for abbreviations.

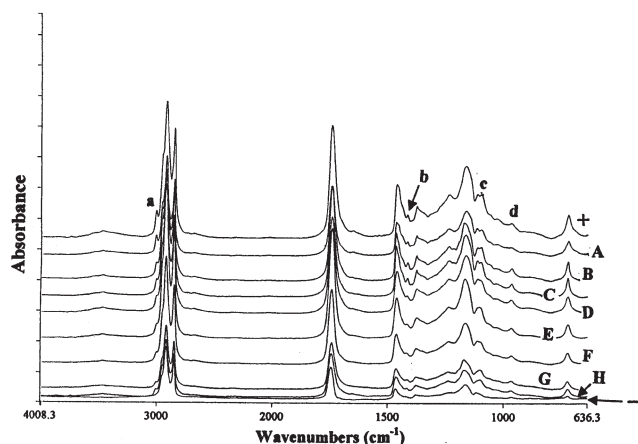


FIG. 10. FTIR spectra of (A) pure lard, (B)–(G) lard/LBF blends, and (H) pure LBF. +, Arithmetic spectrum; –, difference spectrum. The labeled peaks (a–d) are absorption bands that were used in the determination of lard in lard/LBF blends. See Figure 1 for abbreviations; see Figure 2 for compositions of the blends.

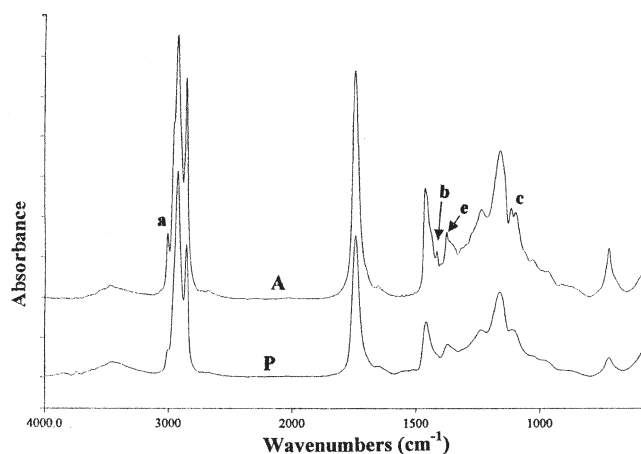


FIG. 11. FTIR spectra of (A) pure lard and (P) pure chicken body fat (CF). The labeled peaks are absorption bands that are significant in differentiating between lard and CF. See Figure 1 for abbreviation.

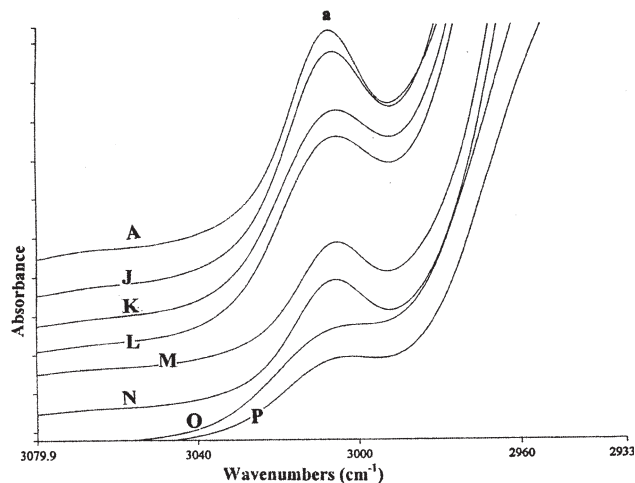


FIG. 12. FTIR spectra of (A) pure lard, (J)–(O) lard/CF blends, and (P) pure CF, illustrating changes in the absorbance value of the band in the region 3008–3000 cm^{-1} (a, Fig. 11). The percentages of lard in the blends are (J) 30, (K) 25, (L) 20, (M) 15, (N) 10, and (O) 5%. See Figures 1 and 11 for abbreviations.

peak observed for pure lard (A) decreases in intensity as the proportion of lard in lard/CF blends decreases (from J to O), becoming a shoulder peak in the spectrum of pure CF (P). The equation $y = 0.0071x + 0.1301$ (where y is the absorbance and x is percent lard), obtained from the plot in Figure 13, could be used for semiquantitative determination of the percentage of lard in lard/CF blends with, R^2 and SE of 0.983 and 0.012, respectively.

(ii) *Frequency range 1418–1417 cm^{-1} .* In this frequency range, the pure lard spectrum (A) has a sharp peak 1417.85 cm^{-1} (peak b) with an absorbance of 0.25 whereas the pure CF spectrum (P) has no peak (Fig. 14). For the blends, the peak starts to appear at a lard content of 10% (Table 3). Figure 15 shows a plot of the absorbance of this peak vs. percent lard with $R^2 = 0.9233$. This relationship could be used for semiquantitative analysis of lard in blends with CF up to 35% (w/w) with lard, an SE of 0.019. The equation of the line is $y = 0.0053x + 0.0821$, where y is the absorbance at 1417.85 cm^{-1} , and x is percent lard.

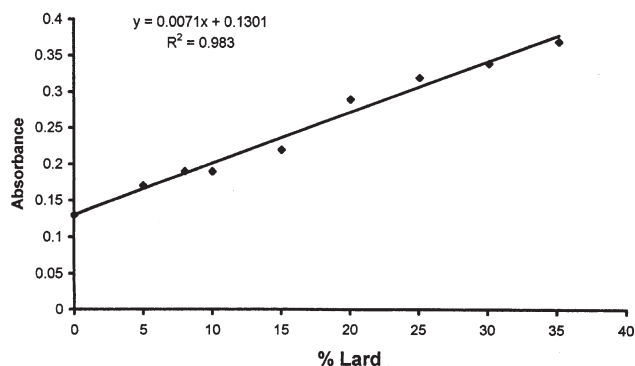


FIG. 13. Absorbance value of band a (Figs. 11 and 12) in the FTIR spectra of CF and lard/CF blends containing up to 35% lard (w/w) vs. % lard. See Figures 1 and 11 for abbreviations.

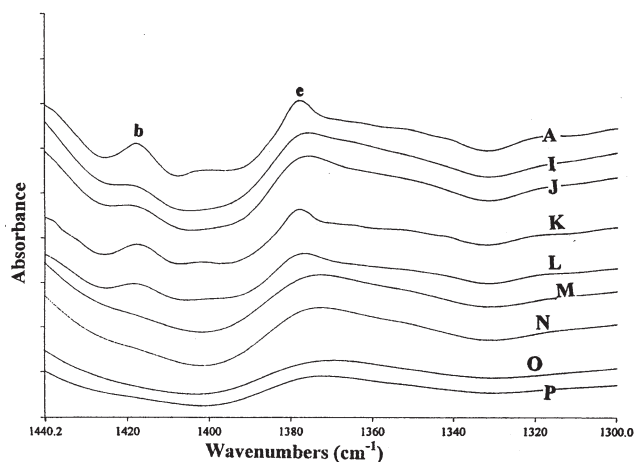


FIG. 14. FTIR spectra of (A) pure lard, (J)–(O) lard/CF blends, and (P) pure CF, illustrating changes in the absorbance values of the bands at 1417.85 cm^{-1} (b, Fig. 11) and 1377.58 cm^{-1} (e, Fig. 11). See Figures 1 and 11 for abbreviations; see Figure 12 for compositions of the blends.

(iii) *Frequency range 1385–1370 cm^{-1}* . Both lard and CF spectra have peaks in this region, but the peak in the pure lard spectrum (A), is high in absorbance and sharp at 1377.58 cm^{-1} , whereas the peak in the pure CF spectrum (P) is flat (broad) and has no shoulder (Fig. 14). The absorbance values in this region in the spectra of lard, CF, and their blends are shown in Table 3. A semiquantitative determination of lard in lard/CF blends is suggested in Figure 16, from which the equation $y = 0.0069x + 0.1327$, with R^2 and SE of 0.9426 and 0.022, respectively, was obtained, where y is the absorbance at 1377.58 cm^{-1} , and x is percent lard in blends with CF covering the range 0–35% (w/w).

Frequency range 1126–1085 cm^{-1} . As shown in Figure 17, in this frequency range the spectra of lard/CF blends look very similar to those of blends of lard with LBF. The pure lard spectrum (A) shows two overlapping peaks at $1100\text{--}1099$ and 1116.64 cm^{-1} with a maximum absorbance of 0.45, whereas the pure CF spectrum (P) has one broad band in the region

TABLE 3
Absorbance Values in the FTIR Spectra of Chicken Body Fat, Lard, and Their Blends at 1417.85 cm^{-1} and 1377.58 cm^{-1} ^a

Spectrum ^b	% Lard	Absorbance ^c :	
		1417.85 cm^{-1}	1377.58 cm^{-1}
A	100	0.30 ± 0.08	0.37 ± 0.07
I	35	0.29 ± 0.05	0.40 ± 0.05
J	30	0.24 ± 0.03	0.35 ± 0.03
K	25	0.22 ± 0.04	0.30 ± 0.04
L	20	0.16 ± 0.05	0.24 ± 0.02
M	15	0.14 ± 0.02	0.21 ± 0.03
N	10	0.13 ± 0.03	0.20 ± 0.02
O	8	0.12 ± 0.02	0.18 ± 0.04
—	5	0.11 ± 0.02	0.19 ± 0.01
P	0	0.11 ± 0.02	0.15 ± 0.05

^aSee Table 1 for abbreviation.

^bLetters refer to spectra in Figure 14.

^cMean \pm standard deviation of two replicates.

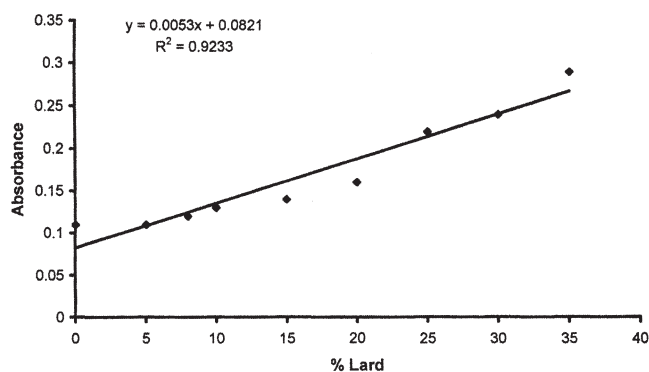


FIG. 15. Absorbance value of band b (Figs. 11 and 14) in the FTIR spectra of CF and lard/CF blends containing up to 35% lard (w/w) vs. % lard. See Figures 1 and 11 for abbreviations.

between 1117 and 1099 cm^{-1} with a maximum absorbance of 0.23. The latter band appears in the spectra of the blends containing up to 10% lard (N), but the spectrum of the 15% lard sample (M) shows the two overlapping peaks, which become much more clearly discerned at higher lard levels (up to 35%). However, changes in absorbance did not correlate well enough with the percent lard or CF in the blends to be used for quantitative analysis.

The four frequency regions mentioned above (a, b, c, and e) are illustrated in Figure 18, which shows the full spectra for all blends.

Cow body fat. Figure 19 shows the spectra of lard (A) and CBF (Q). In the frequency range $3008\text{--}3005\text{ cm}^{-1}$, the CBF spectrum has a small or shoulder peak with an absorbance of 0.33 ± 0.04 whereas the lard spectrum has a clear, sharp peak with high absorbance (0.35 ± 0.02). The second difference between the two spectra is band b at 1417.8 cm^{-1} for the lard spectrum whereas CBF has no clear band as illustrated in Figure 21, which can be attributed to rocking vibrations of CH bonds of *cis*-disubstituted olefins (25,31). The third difference is the presence of a sharp peak at 966 cm^{-1} , due to the *trans* C=CH (peak d) in the CBF spectrum (Q) whereas no clear band is observed in the lard spectrum (A).

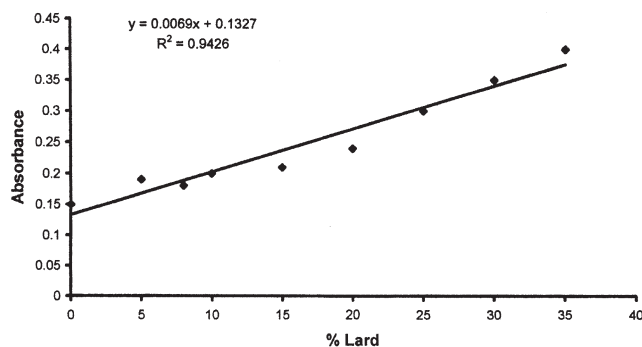


FIG. 16. Absorbance value of band e (Figs. 11 and 14) in the FTIR spectra of CF and lard/CF blends containing up to 35% lard (w/w) vs. % lard. See Figures 1 and 11 for abbreviations.

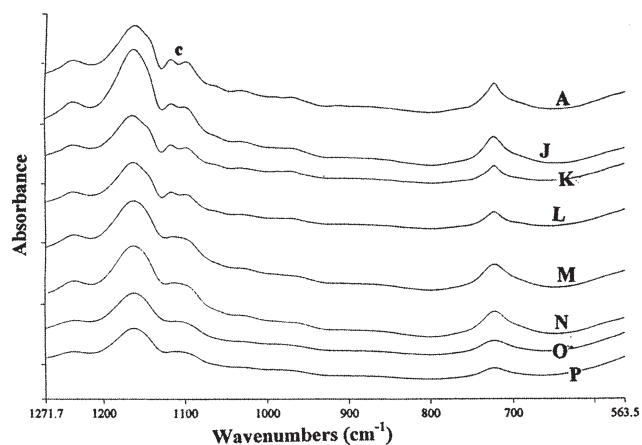


FIG. 17. FTIR spectra of (A) pure lard, (J)–(O) lard/CF blends, and (P) pure CF, illustrating changes in the absorbance values of the band in the region 1126–1085 cm^{-1} (c, Fig. 11). See Figures 1 and 11 for abbreviations; see Figure 12 for compositions of the blends.

(i) *Frequency range 3008–3001 cm^{-1} .* Band a at 3008–3005 cm^{-1} could be used for qualitative analysis to differentiate between pure lard and pure CBF on the basis of the spectral differences seen in Figure 19, as described above. However, despite the variations in the spectra of lard/CBF blends in this region (a in Fig. 20), these changes are not correlated to the blend ratio and, therefore this region is not suitable for analysis of mixtures.

Frequency range 1418–1417 cm^{-1} . As can be seen in Figure 21, this region (denoted b in Fig. 20) can be used for qualitative determination of lard in lard/CBF blends covering the range of 0–35% (w/w) lard. However, the absorbance values or peak frequencies in this region do not correlate well enough to blending ratio to be used for quantitative determination.

(iii) *Frequency range 968–965 cm^{-1} .* Qualitatively it is easy to differentiate between the FTIR spectra of lard and

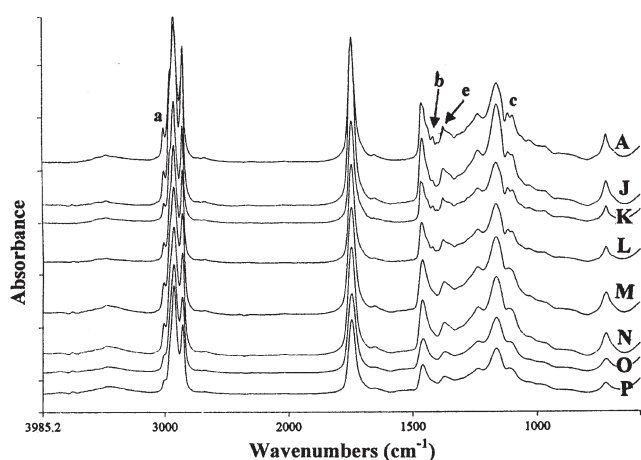


FIG. 18. FTIR spectra of (A) pure lard, (J)–(O) lard/CF blends, and (P) pure CF. The labeled peaks (a, b, c, and e) are absorption bands that were used in the determination of lard in lard/CF blends. See Figures 1 and 11 for abbreviations; see Figure 12 for compositions of the blends.

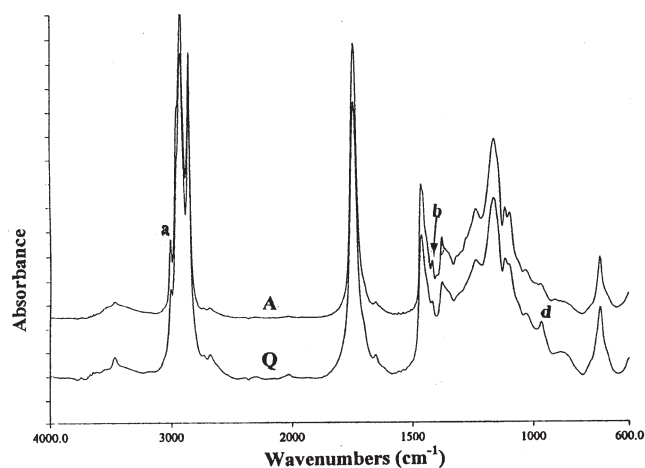


FIG. 19. FTIR spectra of (A) pure lard and (Q) pure cow body fat (CBF). The labeled peaks are absorption bands that are significant in differentiating between lard and CBF. See Figure 1 for abbreviation.

CBF in this region (Fig. 22), because the latter spectrum has a clear band (d) at 966.22 cm^{-1} , due to the *trans* fatty acid content in CBF (1), whereas lard shows no peak at this frequency. Measurement of the absorbance at this frequency in the spectra of lard/CBF blends (Table 4) could be used for quantitative determination of lard. The equation obtained from the plot shown in Figure 23 is $y = -0.005x + 0.3188$ with R^2 and SE of 0.9831 and 0.0086, respectively. As seen in Figures 20 and 23, when the ratio of lard to CBF increases, the absorbance decreases, showing the lower content of *trans* fatty acids in lard than in CBF. In the study by Guillen and Cabo (25), there is no mention of any peak at 966 cm^{-1} in the spectrum of lard.

The use of FTIR spectroscopy in this study offers a rapid, consistent, reproducible, and cost-effective analytical technique that could be used as a quality control measure for ani-

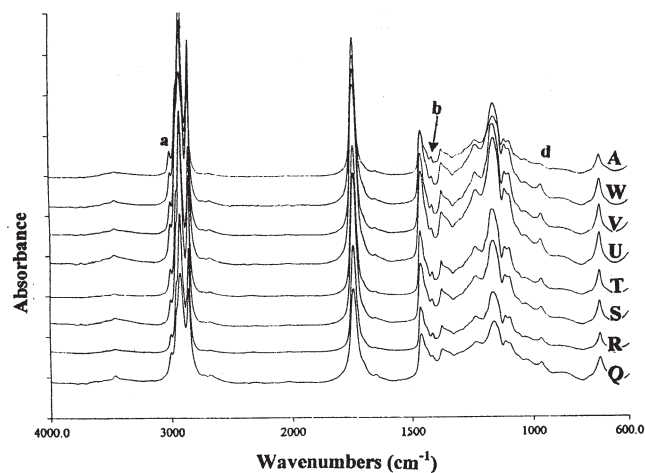


FIG. 20. FTIR spectra of (A) pure lard, (R)–(W) lard/CF blends, and (Q) pure CBF. The percentages of lard in the blends are (R) 10, (S) 15, (T) 20, (U) 25, (V) 30, and (W) 35%. The labeled peaks (a, b, and d) are absorption bands that were used in the determination of lard in lard/CF blends. See Figures 1 and 19 for abbreviations.

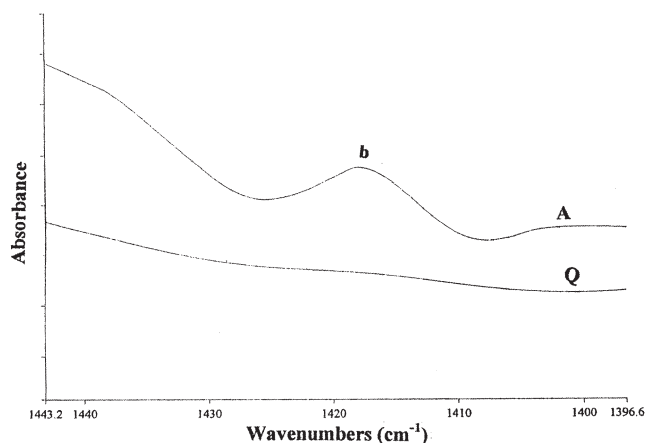


FIG. 21. Expanded view of region b in Figure 19.

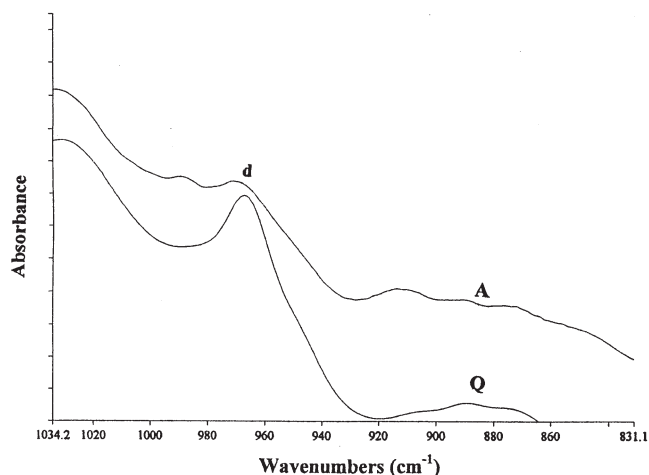


FIG. 22. Expanded view of region d in Figure 19.

mal fats in their neat form. An additional benefit of using this FTIR spectroscopic method is that the tedious time- and chemical-consuming standard chemical methods can be avoided. The study could be extended by using specific sampling regarding, for example, the age of the animal, sex, diet,

TABLE 4
Absorbance Values in the FTIR Spectra of Cow Body Fat, Lard, and Their Blends at 966.22 cm⁻¹^a

Spectrum ^b	% Lard	Absorbance ^c
Q	0	0.32 ± 0.06
R	10	0.26 ± 0.04
S	15	0.25 ± 0.02
T	20	0.23 ± 0.01
U	25	0.19 ± 0.02
V	30	0.16 ± 0.02
W	35	0.15 ± 0.03
A	100	0.13 ± 0.01

^aSee Table 1 for abbreviation.

^bLetters refer to spectra in Figure 20.

^cMean ± standard deviation of two replicates.

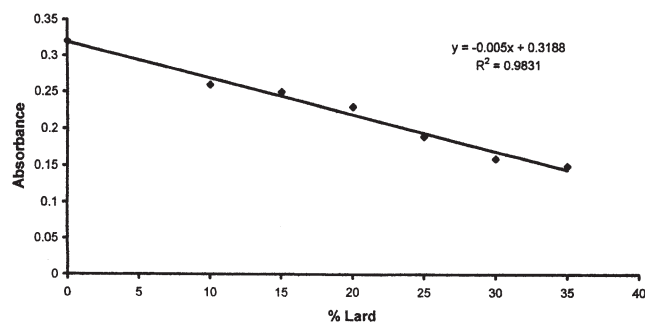


FIG. 23. Absorbance value of band d (Figs. 20 and 22) in the FTIR spectra of CBF and lard/CBF blends containing up to 35% lard (w/w) vs. % lard. See Figures 1 and 19 for abbreviations.

and different sites (e.g., rump, belly, perirenal, intermuscular) and also by including other types of fats and oils, such as milk fats and vegetable oils. Once the database is established and the model developed, each individual analysis can be achieved in about 2 minutes. Thus, FTIR spectroscopy has potential as a rapid method for the authentication of animal fats and/or detection of adulteration of fats.

ACKNOWLEDGMENTS

The authors thank the Universiti Putra Malaysia (UPM) for providing the funding (IRPA No. 03-02-04-048) for this study. The authors are grateful to the Department of Chemistry, Faculty of Science and Environmental Studies (UPM), for allowing the use of the 1650 Series FTIR spectrometer.

REFERENCES

- Enser, M., Meat Lipids, in *Developments in Oils and Fats*, edited by R.J. Hamilton, Blackie Academic and Professional, London and New York, 1995, pp. 1–31.
- Codex Alimentarius*, Vol. 8, Part 3, *Fats, Oils and Related Products*, Food and Agriculture Organization of the United Nations, World Health Organization, Rome, 1993.
- Sonntag, N.O.V., Fat Splitting, Esterification and Interesterification, in *Bailey's Industrial Oil and Fat Products*, 4th edn., edited by D. Swern, Wiley-Interscience, New York, 1982, Vol. 2, pp. 159–161.
- Lambelet, P., Detection of Pig and Buffalo Body Fat in Cow and Buffalo Ghee by Differential Scanning Calorimetry, *J. Am. Oil Chem. Soc.* 60:1005–1008 (1983).
- Lambelet, P., O.P. Singhal, and N.C. Ganguli, Detection of Goat Body Fat in Ghee by Differential Thermal Analysis, *Ibid.* 57:364–366 (1980).
- Coni, E., M.D. Pasquale, P. Coppolelli, and A. Bocca, Detection of Animal Fats in Butter by Differential Scanning Calorimetry: A Pilot Study, *Ibid.* 71:807–810 (1994).
- Hammond, E.W., and J.W. Irwin, The Analysis of Lipids by HPLC, in *HPLC in Food Analysis*, 2nd edn., edited by R. Macrae, Academic Press, London, 1988, pp. 95–132.
- DeMan, J.M., Functionality Requirements of Fats and Oils for Food Applications, presented at *MOSTA Tech-In, Recent Advances in the Sciences of Oils and Fats*, Malaysian Oil Scientists' and Technologists' Association, Petaling Jaya, Selangor D.E. Malaysia, 1999.
- Kaufmann, H.P., and G. Manked, *Fette, Seifen Amstrichsm.* 65, 179, (1963), cited in *Bailey's Industrial Oil and Fat Products*, 4th edn., edited by D. Swern, Wiley-Interscience, New York, 1982, Vol. 2, p. 482.

10. IUPAC Commission on Oils, Fats and Derivatives, *Standard Methods for the Analysis of Oils, Fats and Derivatives*, 6th edn., Pergamon, New York, 1979, Method 2.210.
11. Kirk, R.S., and R. Sawyer, *Pearson's Composition and Analysis of Foods*, 9th edn., Longman Scientific and Technical, Essex, 1991, pp. 611–612.
12. *Official and Tentative Methods and Recommended Practices of the American Oil Chemists' Society*, 4th edn., edited by D. Firestone, American Oil Chemists' Society, Champaign, 1993, Method Cb 5-40.
13. Haryati, T., Development and Application of Differential Scanning Calorimetric Technique for Physical and Chemical Analysis of Palm Oil, Ph.D. Thesis, Faculty of Food Science and Biotechnology, Universiti Putra Malaysia, Serdang Selangor DE, Malaysia, 1999.
14. Tan, C.P., and Y.B. Che Man, Differential Scanning Calorimetric Analysis of Edible Oils: Comparison of Thermal Properties and Chemical Composition, *J. Am. Oil Chem. Soc.* 77:143–155 (2000).
15. Ismail, A.A., F.R. van de Voort, G. Emo, and J. Sedman, Rapid and Quantitative Determination of Free Fatty Acids by FTIR Spectroscopy, *Ibid.* 70:335–341 (1993).
16. van de Voort, F.R., A.A. Ismail, J. Sedman, J. Dubois, and T. Nicodemo, The Determination of Peroxide Value by Fourier Transform Infrared Spectroscopy, *Ibid.* 71:921–926 (1994).
17. van de Voort, F.R., A.A. Ismail, and J. Sedman, A Rapid Automated Method for the Determination of *cis* and *trans* Content of Fats and Oils by Fourier Transform Infrared Spectroscopy, *Ibid.* 72:873–880 (1995).
18. van de Voort, F.R., K.P. Memon, J. Sedman, and A.A. Ismail, Determination of Solid Fat Index by Fourier Transform Infrared Spectroscopy, *Ibid.* 73:411–416 (1996).
19. Che Man, Y.B., and G. Setiowaty, Determination of Anisidine Value in Thermally Oxidized Palm Olein by Fourier Transform Infrared Spectroscopy, *Ibid.* 76:1–5 (1999).
20. Setiowaty, G., Y.B. Che Man, S. Jinap, and M.H. Moh, Quantitative Determination of Peroxide Value in Thermally Oxidized Palm Olein by Fourier Transform Infrared Spectroscopy, *Phytochem. Anal.* 11:74–78 (2000).
21. Che Man, Y.B., and M.E.S. Mirghani, Rapid Method for Determining Moisture Content in Crude Palm Oil by Fourier Transform Infrared Spectroscopy, *J. Am. Oil Chem. Soc.* 77:631–637 (2000).
22. Che Man, Y.B., G. Setiowaty, and F.R. van de Voort, Determination of Iodine Value of Palm Oil by Fourier Transform Infrared Spectroscopy, *Ibid.* 76:693–700 (1999).
23. Che Man, Y.B., and G. Setiowaty, Multivariate Calibration of Fourier Transform Infrared Spectra in Determining Iodine Value of Palm Oil Products, *Food Chem.* 67:193–198 (1999).
24. Mirghani, M.E.S., Y.B. Che Man, S. Jinap, B.S. Baharin, and J. Bakar, Rapid Method for Determining Malonaldehyde as Thiobarbituric Acid Reactive Substance (TBARS) in Palm Olein System by Fourier Transform Infrared Spectroscopy, *Phytochem. Anal.*, PCA BC 8889, in press.
25. Guillen, M.D., and N. Cabo, Characterization of Edible Oils and Lard by Fourier Transform Infrared Spectroscopy. Relationships Between Composition and Frequency of Concrete Bands in the Fingerprint Region, *J. Am. Oil Chem. Soc.* 74:1281–1286 (1997).
26. Hoffmann, G., *The Chemistry and Technology of Edible Oils and Fats and Their High Fat Products*, Academic Press, London, 1989, p. 127.
27. Dubois, J., F.R. van de Voort, J. Sedman, A.A. Ismail, and H.R. Ramaswamy, Quantitative Fourier Transform Infrared Analysis for Anisidine Value and Aldehyde in Thermally Stressed Oils, *J. Am. Oil Chem. Soc.* 73:787–794 (1996).
28. Firestone, D., and Sheppard, A., Determination of *trans* Fatty Acids, in *Advances in Lipid Methodology—One*, edited by W.W. Christie, The Oily Press, Alloway, Scotland, 1992, pp. 273–322.
29. van de Voort, F.R., A.A. Ismail, J. Sedman, and G. Emo, Monitoring the Oxidation of Edible Oils by Fourier Transform Infrared Spectroscopy, *J. Am. Oil Chem. Soc.* 71:243–253 (1994).
30. Safar, M., D. Bertrand, P. Robert, M.F. Devaux, and C. Genot, Characterization of Edible Oils, Butters and Margarines by Fourier Transform Infrared Spectroscopy with Attenuated Total Reflectance, *Ibid.* 71:371–377 (1994).
31. Guillen, M.D., and N. Cabo, Usefulness of the Frequency Data of the Fourier Transform Infrared Spectra to Evaluate the Degree of Oxidation of Edible Oils, *J. Agric. Food Chem.* 47:709–719 (1999).
32. Firestone, D., and P. LaBouliere, Determination of Isolated *trans* Isomers by Infrared Spectrophotometry, *J. Assoc. Off. Anal. Chem.* 48:437–443 (1965).
33. *Official Methods and Recommended Practices of the American Oil Chemists' Society*, 4th edn., American Oil Chemists' Society, Champaign, 1995, Method Cd 14-95.
34. Sedman, J., F.R. van de Voort, and A.A. Ismail, Upgrading the AOCS Infrared *trans* Method for Analysis of Neat Fats and Oils by Fourier Transform Infrared Spectroscopy, *J. Am. Oil Chem. Soc.* 74:907–913 (1997).

[Received May 24, 2000; accepted April 2, 2001]