Determination of Slip Melting Point in Palm Oil Blends by Partial Least Squares and Principal Component Regression Modeling of FTIR Spectroscopic Data

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ABSTRACT: The determination of slip melting point (SMP) in refined, bleached, and deodorized palm oil is a common and regular requirement in the manufacture of this oil. An FTIR spectrometer in the mid-IR wavelength region (4000–750 cm^{-1}) can provide the basis for a suitable on-line and off-line quantitative analytical methodology if used with a suitable calibration model. In this study, by using data from selected regions, the calibration models, including principal component regression and partial least squares regression, have been examined and evaluated for efficacy in determining SMP directly in palm oil blends. Results indicate that orthogonal models using selected wavelength data offer superior predictive performance.

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KEY WORDS: FTIR, palm oil, PCR, PLS, slip melting point.

In palm oil trading, properties such as moisture content, impurities, color, slip melting point (SMP), and FFA are used for quality assessment. Among the important physical properties of palm oil parameters applied in end products is SMP. A number of standard procedures are available for the determination of the SMP of fats and oils, e.g., AOCS Cc 3-25 (1) and Cc 1-25 (2); British Standard 684 (3); Indian Standard 548 (4); and that described by Cocks and van Rede (5). These methods are cheap and robust; unfortunately, they are usually tedious and strongly dependent on visual observation by the operator.

Our research group has been engaged in the development of rapid and more technologically advanced methods of palm oil analysis through the use of FTIR spectroscopy to replace traditional chemical methods, including the determination of iodine value (6,7), FFA (8), PV (9), anisidine value (10), saponification number (Setiowaty, G., and Y.B. Che Man, unpublished data), and moisture content (11) with reliable and satisfactory results. These methods are designed to produce analytical results within 5 min per sample and are based on spectral information obtained from palm oil in the mid-IR portion of the electromagnetic spectrum.

The application of quantitative chemometrics, such as partial least squares regression (PLS) and principal component regression (PCR), to chemical data is becoming more widespread owing to the availability of digitized spectroscopic data and commercial software for laboratory computers. These approaches may constitute a rapid, feasible, and attractive alternative for satisfying the demands of control and routine analyses.

In this study, an FTIR spectroscopic method was developed for the determination of SMP of palm oil using PLS and PCR calibration methods. The predictive abilities of both methods were investigated.

MATERIALS AND METHODS

Materials. Raw materials used in this study were palm oil products such as refined, bleached, deodorized (RBD) palm olein and RBD palm stearin, which were obtained from a local refinery [Ngo Chiew Hong (M) Sdn. Bhd., Kuala Lumpur, Malaysia]. All chemicals used were of analytical grade.

Analytical methods. A set of 37 calibration standards was prepared by blending two oils (w/w) to obtain oil blends with a wide range of SMP. The SMP of each oil was analyzed in duplicate using AOCS standard method Cc 1-25 (2). Completely melted sample was placed in three capillary tubes to a height of 10 mm in each tube. The tube was placed in a beaker and held in a refrigerator at 4–10°C for 16 h. The tube was then attached to the thermometer with a rubber band. The thermometer was suspended in a 600-mL beaker half-full of distilled water. The initial bath temperature was adjusted to 8–10°C below the SMP of the sample. The water bath was agitated with a magnetic stirrer and heat was applied to increase the bath temperature at the rate of 0.5°C/min. The temperature at which the sample in each tube started to melt and become clear was observed, and the average temperature of all tubes was calculated. The start and end of SMP were marked as SMPs and SMPe, respectively. The average of the start and end of SMP was marked as SMPa.

Instrumentation. A PerkinElmer 1725X series FTIR spectrometer (PerkinElmer Corp., Norwalk, CT) was interfaced with a PerkinElmer model 7300 professional computer and run under Infrared Data Management System software. This instrument was also connected to a room-temperature deuterated triglycine sulfate detector and a horizontal attenuated

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total reflectance (ATR) accessory with a 45° zinc selenide element. Two automatic dehumidifiers were used to protect the instrument from water vapor interference.

Spectral acquisition. All standards were equilibrated to 65°C in a conventional oven and applied with a soft-tipped disposable pipette (1 mL) to ensure complete coverage to the top ATR element. All spectra were recorded from 4000 to 750 cm^{-1} at a resolution of 4 cm^{-1}. For each spectrum, 32 interferograms were co-added before Fourier transformation. The Norton–Beer normal apodization and a gain of 2 were employed. Each sample single-beam spectrum was ratioed to a single-beam spectrum of the clean ATR element, collected under identical conditions, before conversion into absorbance units. The ATR element was cleaned between each sample by removing the previous sample with tissue and cleaning with acetone. A new single-beam reference was collected before each sample was measured. The time required from completion of a measurement on a given sample to completion of the corresponding measurement on the next sample was about 7–10 min.

Data analysis. The full-spectrum multivariate calibration methods such PLS and PCR are known to reduce the calibration spectral-intensity data at many frequencies to a relatively small number of intensities in a transformed full-spectrum co-ordinate system (12,13). PLS is related to PCR in that spectral decomposition is also performed, but this decomposition step is performed differently. In PCR, the spectra are decomposed on the basis of the maximal variance between spectral data without using information about the concentrations. PLS differs from PCR in that it uses both the spectral and the concentration data in modeling (14,15). Hence, PLS sacrifices some fit of the spectral data relative to PCR in order to achieve better correlations to concentrations during prediction. The PLS and PCR computations were performed by the Spectrum Quant+ (Version 4.1, Perkin-Elmer) software package.

To select the number of factors in the PLS and PCR methods, in order to model the system without overfitting the concentration data, a cross-validation method leaving out one sample at a time was used (16). Given the set of 37 calibration spectra, the PCR and PLS calibration on 36 calibration spectra was performed, and using this calibration, the SMP of the standard left out during calibration was predicted. This process was repeated 37 times until each calibration standard had been left out once. The predicted SMP in the standard was compared with the known SMP in this standard and the prediction error sum of squares (PRESS) was calculated. The maximum number of factors used to calculate the optimum PRESS was selected as 10.

The root mean square difference (RMSD), an indication of the average error in the analysis; the square of the correlation coefficient (R^2), an indication of the quality of fit of all the data to a straight line; and the relative error of prediction (REP), the square root of the mean square of the error in the prediction for each component expressed as a percentage of the mean of the true concentrations, were also calculated and compared between the PLS and PCR methods.

RESULTS AND DISCUSSION

Analytical methods. The standard analysis showed that the SMP of the samples ranged from 22 to 55°C, with a mean of 34.28 and SD of 8.52°C. This range covered almost all of the palm oil products as specified by the Palm Oil Refiners Association of Malaysia, i.e., RBD palm olein, maximum 24°C; RBD palm stearin, minimum 44°C (17).

Selection of the spectral region for the analysis. Figure 1 illustrates the overlaid FTIR absorbance spectra of samples having SMP values of 22.9, 26.3, 30, 35, 40.9, 46.6, 50.8, and 54.7°C. The oils appear to exhibit only minor spectral differences. On the scale shown, the only noticeable spectral differences are in the *cis*-CH stretching and CH$_2$/CH$_3$ stretching regions (3100–2800 cm^{-1}) and the ester linkage carbonyl band (1750–1740 cm^{-1}) (10). Although many combinations and permutations of wavelength regions can provide fairly reasonable results, the regions determined to be optimal for the samples were the CH$_2$/CH$_3$ stretching region (2992–2832 cm^{-1}) and the ester linkage carbonyl absorption band (1750–1724 cm^{-1}). Furthermore, most of the spectral information is contained in those regions, so the use of data points extracted from only those regions in the PCR and PLS analyses did not reject important data.

Multivariate calibration methods. To improve the analyses for SMP in the palm oil blends, we evaluated two different chemometric approaches. Multivariate calibrations are useful in spectral analyses because the simultaneous inclusion of multiple spectral intensities can greatly improve the precision and predictive ability. Haaland and Thomas (13) compared the different multivariate calibration methods for quantitative spectral analysis and concluded that generalizing about the superiority of one method over another is very difficult, because the relative performances of the methods are often dependent on the particular data set being analyzed. They also recommended the use of PLS in the absence of specific information about the data set.
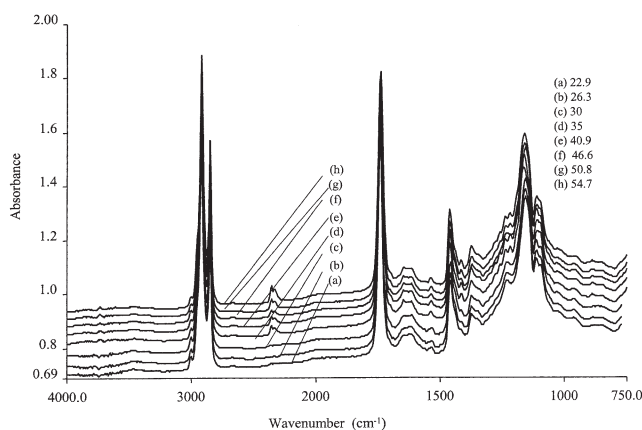

FIG. 1. Overlaid spectra of eight standards of palm oil blends with slip melting point (SMP) values of (a) 22.9, (b) 26.3, (c) 30, (d) 35, (e) 40.9, (f) 46.6, (g) 50.8, and (h) 54.7°C as measured by AOCS method Cc 1-25 (2).
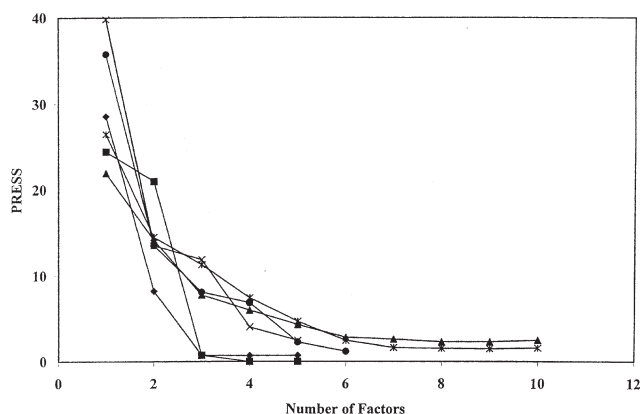
FIG. 2. Representation of PRESS values generated from the prediction of SMP by the PLS and PCR methods, as the function of the number of factors used in the calibration, for (◆) PCR and (■) PLS in the 1750–1724 cm^{-1} region; (▲) PCR and (×) PLS in the 2992–2832 cm^{-1} region; and (*) PCR and (●) PLS for both regions. PRESS, prediction error sum of squares; PLS, partial least squares; PCR, principal component regression; for other abbreviation see Figure 1.

Selection of the optimal number of factors. One reasonable choice for the optimal number of factors would be that number that yielded the minimum PRESS. However, using the number of factors ($k*$) that yields a minimum PRESS usually leads to some overfitting. A better criterion for selecting the optimal number of factors involves the comparison of PRESS from models with fewer than $k*$ factors. The model selected is the one with the fewest number of factors, such that the PRESS for that model is not significantly greater than the PRESS from the model with $k*$ factors. The F statistic was used to make the significance determination. Haaland and Thomas (13) determined that an F-ratio probability of 0.75 is a good choice. We selected, as the optimum, the number of factors for the first PRESS value where the F-ratio probability dropped below 0.75.

Figure 2 shows the PRESS value obtained by optimizing the calibration model of the absorption spectra in the regions of 2992–2832, 1750–1724 cm^{-1}, and both regions with the PLS and PCR method applied. When using the 1750–1724 cm^{-1} region, the optimal number of factors was found to be 3 for the

TABLE 1
Statistical Parameters of the PCR and PLS Methods in the Selected Spectral Regions in the Quantification of SMP Using All Palm Oil Blend Standards[a]

Region (cm^{-1})	PCR		PLS	
	RMSD	R^2	RMSD	R^2
1750–1724	0.46 (4)	0.99	0.56 (3)	0.99
2992–2832	1.29 (7)	0.98	1.29 (5)	0.98
1750–1724 and 2992–2832	0.94 (7)	0.99	0.83 (6)	0.99

[a]PCR, principal component regression; PLS, partial least squares; SMP, slip melting point; RMSD, root mean squares difference; R^2, square of correlation coefficient.
[b]Values in parentheses correspond to the number of factors used for prediction.

TABLE 2
Relative Error of Prediction Values (%) for Slip Melting Point by PCR and PLS Methods[a]

Region (cm^{-1})	PCR	PLS
1750–1724	1.35	1.63
2992–2832	3.76	3.78
1750–1724 and 2992–2832	2.75	2.43

[a]See Table 1 for abbreviations.

PLS method and 4 for the PCR method. For the spectral region 2992–2832 cm^{-1}, the optimal number of factors was found to be 5 for the PLS method and 7 for the PCR method. If both spectral regions mentioned above are used, the optimal number of factors was found to be 6 for the PLS method and 8 for the PCR method.

Statistical parameters. In Table 1, the values of RMSD and R^2 found for the absorbance spectral data are summarized. In Table 2 it can be seen that the values of the RMSD are similar for PCR and PLS in the 2992–2832 cm^{-1} region. The lowest RMSD was given by PCR applied to the region of 1750–1724 cm^{-1}, followed by the PLS method for the same region. Utilization of both spectral regions chosen increased RMSD in either the PLS or PCR methods. The values found for the RMSD are, in all instances, less than 1.3, and there are no significant differences when using the PLS or PCR methods in the applied regions in the spectral data. The values of R^2 are similar in the two methods used for the spectral regions examined. In all instances, the values of R^2 obtained are satisfactory; particularly in the region 1750–1724 cm^{-1}, both methods gave superior results.

The RMSD values are an estimate of the absolute error prediction for a property. The predictive ability of each method for a property can also be described in terms of the REP. In Table 2, the REP for the SMP of the palm oil blends using absorbance spectral data are reported. The values found for the REP are less than 3.8%, and there is no significant difference in the precision of prediction in either 2992–2832 cm^{-1}, 1750–1724 cm^{-1}, or both regions for the PLS and PCR methods.
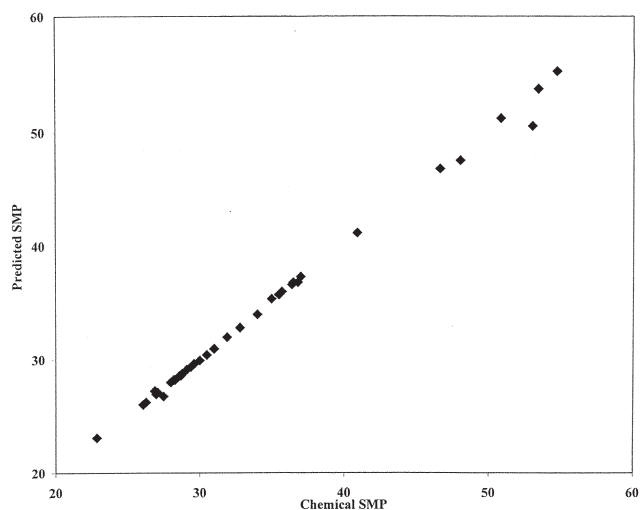

FIG. 3. Predicted vs. chemical values plot for SMP in palm oil using the PCR method with four factors. See Figures 1 and 2 for abbreviations.
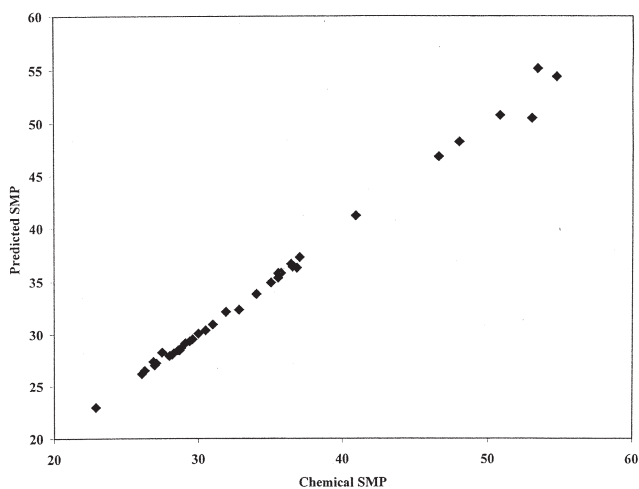
FIG. 4. Predicted vs. chemical values plot for SMP in palm oil using the PLS method with three factors. See Figures 1 and 2 for abbreviations.

In spite of the statistical analysis performed on the predicted ability of the methods, which showed no significant differences in the REP values when using the spectral regions chosen, slightly better results were obtained for the prediction of SMP of palm oil blends when using the region 1750–1724 cm^{-1} for the PLS and PCR methods. Figures 3 and 4 show the results of the determination of SMP in palm oil compared with their chemical values for the PLS and PCR methods, respectively. It can be seen that there was good agreement between the real and predicted values.

The results presented here indicate that good prediction ability is obtained using PLS and PCR models applied to the spectral range of 1750–1724 cm^{-1} in the FTIR data set. The unique advantage of the FTIR approach combined with appropriate calibration models is that it can provide a powerful tool for a rapid quality control method for the determination of the SMP of palm oil and its products.

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