

Mathematical pretreatments of spectra—useful tools for reducing instrument differences

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Introduction

Near infrared (NIR) spectra are composed of a mixture of diffuse and specular components that are strongly influenced by the scattering and absorption characteristics of the sample. Diversity of the particle size of the material will affect the scattering and is a major source of variation in NIR spectra. These effects vary from sample to sample and are both additive and multiplicative in nature. Additive effects cause vertical displacement (or shift of the spectra upward or downward) whereas multiplicative effects appear as non-linear slope changes when compared to an “ideal” or reference spectrum. In practice, “ideal” spectra are impossible to obtain, therefore the mean spectrum of a set is assumed to be good representation for the reference base.

Many mathematical manipulations (or treatments) which have been proposed to correct for particle size anomalies are used in NIR spectroscopy. These include calculating first and second derivatives, multiplicative scatter correction (MSC), standard normal variate (SNV) and detrending (DT) transformation. All transformations are applied before entering the calibration process.

The objective of the study reported in this paper was to determine the potential of math pretreatments for reducing differences between two different types of NIR spectrophotometers—a Foss NIRSystems Model 6500 (Silver Spring, MD, USA) and an InfraAlyzer 500 (Braun+Luebbe, Norderstedt, Germany). Only four math treatments (and their combination) were considered: (1) First derivative, (2) second derivative, (3) standard normal variate (SNV) and (4) detrending (DT). It would be advantageous if a calibration developed on one NIR instrument could be successfully transferred to and utilised on another NIR instrument.

Material and methods

Mathematical treatments of all the spectral raw Bran+Luebbe InfraAlyzer 500 data of Polish pig fat samples with chemically determined iodine values¹ were performed using the ISI NIR 3 software ver. 3.11 (Infrasoft International, Port Matilda, PA, USA). Calibration equations for the prediction of four main fatty acids of Iberian pig fat: palmitic acid (C16 : 0), stearic acid (C18 : 0), oleic acid

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(C18 : 1) and linoleic acid (C18 : 2) were also developed using the same software.² The NIR spectra of fatty acids contain bands at 1180, 2143 and 2190 nm which may be attributed to *cis* unsaturation.³

The modified partial least squares (MPLS) regression technique⁴ was used to develop all NIR calibrations. In MPLS regressions, the spectral data are reduced to a few independent factors that retain most of the spectral information—factors correlatable to chemical (reference data) of the samples. The standard normal variate (SNV) and detrend (DT) scatter correction procedure were applied to the spectral data in the following manner.⁵ The SNV pretreatment required that the standard deviation of each spectrum be 1.0. DT removed the linear and quadratic components of each spectrum. Then a first order mathematical calculation was made, using ISI default settings of 1, 4, 4, 1 where the first digit is the number of the derivative, the second is the gap over which the derivative is calculated and the third is the number of data points in the convolution interval (segment or running average).⁶

Figure 1 shows the mean absorbance, SNV, DT, SNV + DT, SNV + DT + 1st derivative and SNV + DT + 2nd derivative spectra of Iberian pig fat recorded on an NIRS 6500 and Polish pig fat (re-recorded on an IA 500). The spectral wavelength range for all analyses in this paper was restricted to 1100–2500 nm.

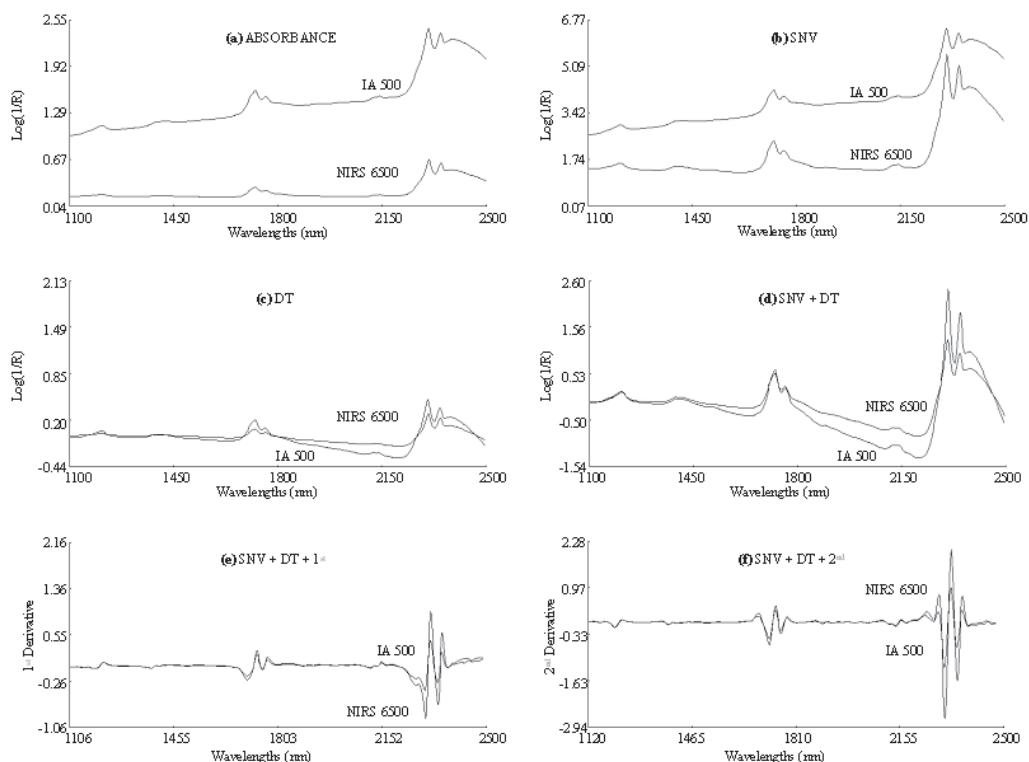


Figure 1. NIR mean spectra of pig fat sample sets: (a) absorbance, (b) SNV, (c) DT, (d) SNV + DT, (e) SNV + DT + 1st derivative and (f) SNV + DT + 2nd derivative.

Results and discussion

Correlations between the main fatty acids content of Iberian-pig-fat samples and iodine value in Polish-pig-fat samples are given in Table 1.

The results of calculation using MPLS regression equations² show (Table 1) that linoleic acid (C 18:2) contents (%) of Iberian pig fat samples were highly correlated ($r = 0.897$) with the iodine values of Polish pig fat samples. The degree of unsaturation of fats is proportional to the iodine value, defined as the number of grams of iodine required to react with 100 g of fat under test conditions.

The graphic presentation of these dependences is illustrated in Figure 2. To validate the calibrations, a new independent sample set of Polish pig fat was collected in from meat grown in an experiment to determine the efficiency of mineral-fat supplement in boars and gilts nutrition.⁷ The vegetable dietary fat supplement had a significant effect on increasing the iodine values of pig back-fat from 57.9 (control group) to 62.3, 63.0 and 67.0 (experimental groups). The effect of vegetable oil on animal fat quality was reflected in the composition of the fatty acids determined by gas chromatography (GC).⁸ The

Table 1. Correlation between main fatty acids contents of Iberian pig fat samples and iodine value in Polish pig fat samples.

Fatty acid	Correlation coefficient (r)
C 16 : 0 (palmitic)	0.272
C 18 : 0 (stearic)	0.071
C 18 : 1 (oleic)	0.158
C 18 : 2 (linoleic)	0.897

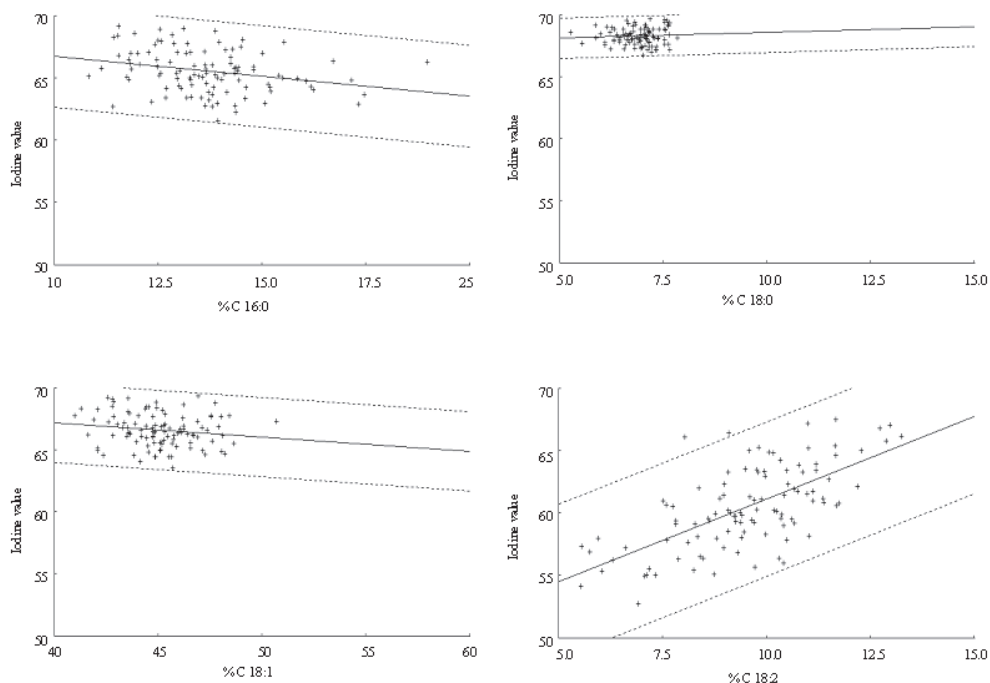


Figure 2. Graphic relationship of correlation between Iberian pig fat samples fatty acids contents and iodine values of Polish pig fat samples.

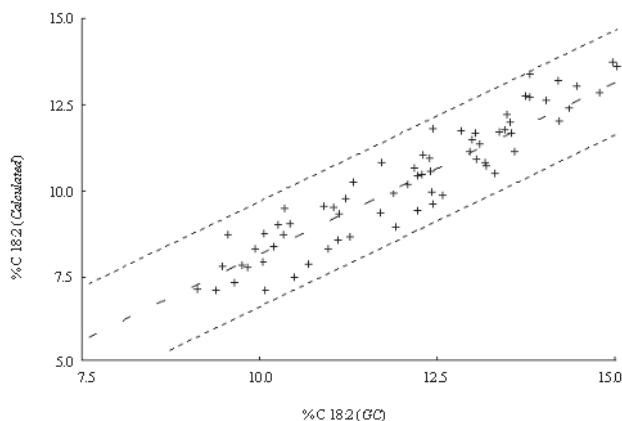


Figure 3. Relationship between GC-determined and calculated lineolic acid contents for independent prediction set of Polish pig fat samples.

contents of unsaturated (oleic, linoleic) fatty acids in the experimental groups were found to increase significantly (linoleic acid—from 9.6 to 12.8%).

The prediction ability of the calibrations for the determination of linoleic acid content is plotted in Figure 3, comparing the calculated contents to their corresponding linoleic acid concentration determined by GC method. Although the regression statistics are good (Figure 3), the deviations of predicted contents of linoleic acid are high. The relatively high correlation coefficient ($r = 0.878$) with the worse *SEP* ($SEP = 0.37\%$) is probably due to fact that r only measures the degree to which the calibrations fits the data. It does not give an indication of how reliable these predicted values are.

Data presented in Figure 3 suggest the need for a bias correction. The different spectral response of spectrophotometers and/or the different GC methods^{8,9} of the fatty acids determination can be the sources of bias (bias = 2.4%).

These results demonstrate that the linoleic acid concentration in pig fat can be estimated from the “calibration” curve calculated from the NIR data transferred from other type of NIR monochromator spectrophotometer. However, the high error and low precision, at this stage of our investigations, are less than satisfactory.

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