Near infrared analysis of liquid and dried ewe milk

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Introduction

The routine analysis of chemical components of milk is of major importance both for the management of animals in dairy farms and for quality control in dairy industries. A recent review presented by Laporte and Paquin[1] points out that, despite widespread use of near infrared (NIR) spectroscopy in the dairy industry and the general enthusiasm surrounding this technology, there are still problems regarding NIR analysis routines which must be solved before its final implantation in dairy industries and laboratories.

Sample preparation has often been considered as one of the most critical aspects in NIR analysis of liquid products. The main problem associated with their sample preparation is water content because it absorbs most of the infrared radiation and this disturbs the calibration for other constituents. To solve this problem, the dry extract spectroscopy by infrared reflectance (DESIR)[2] system was developed. This method consists of drying a glass fibre filter previously impregnated with the liquid under test. It has been proposed, for goat's milk analysis, to modify the typical drying conditions of this method (70°C, 15 min) to 40°C, 24 h to avoid protein denaturation.[3,4] However, this makes NIR analysis lose one of its main advantages: to provide instantaneous results with little or no sample preparation.

There are other measurement methods appropriate for liquids or semi-liquids, such as transmittance[5] and folded transmission or transflectance.[6–8] In this sense, the prediction results of DESIR and transmittance measurements of prepared liquid foods have been compared by Isaksson *et al*.[9]

The aim of the present study was to compare the accuracy of folded transmission (liquid milk) and reflectance (dried milk) NIR calibration equations to predict quality parameters in ewe's milk.

Materials and methods

Milk samples

A set of 101 ewe's milk samples was used to develop the calibration equations. All of the samples came from individual controls in different lactation in order to obtain maximum seasonal variation. The samples were preserved by adding potassium dichromate, stored at 2–4°C and analysed within 72 hours of collection.

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Chemical analysis

Prior to chemical and NIR analyses, milk samples were heated at 40°C, mixed gently in order to achieve uniform dispersion of fatty matter and other components and then left to cool at room temperature.

Milk samples were analysed in duplicate in order to determine the following chemical parameters: total protein (using the colorimetric method described by Bradford),[10] total casein (precipitation of milk casein and subsequent determination of whey protein using the method indicated above), fat (Gerber), total solids (oven drying at 103ºC ± 2ºC) and somatic cell count-SCC (Fossomatic).

NIR analysis and chemometric treatments

A Foss NIRSystems 6500 SY-I scanning monochromator (400–2500 nm), equipped with a spinning module, was used. All samples were analysed using the two spectroscopic methods to be compared in this study:

Reflectance (R): Small ring cups for solid product analyses were used. One filter per sample was prepared and oven dried at 40°C for 24 h. After one hour in a desiccator, the filters were placed in the cup with the readable side against the quartz window to perform NIR analysis.

Folded transmission (FT): An aluminium reflector 0.1 mm pathlength cam-lock cell for liquid product analyses was used. A sample of 0.85 mL was placed in the cell and the sample was scanned through the quartz window. Two cells per sample were filled and the average spectrum was used in the data analyses.

Folded transmission and reflectance spectra of a milk sample are shown in Figure 1.

Both spectral data collection and chemometric treatment of the data were performed using ISI software (NIRS 3 ver 3.11; Infrasoft International, Port Matilda, PA, USA). MPLS (modified partial least squares) was used for regression purposes; 400–2500 nm and 1100–2500 nm regions (in 2 nm steps) were tested; SNV and Detrending treatments were applied for scatter correction. Several first and second derivative treatments were also evaluated. The methodology followed for the development and evaluation of NIR calibrations is described in different publications.[11–13] The following statistics were used to select the most accurate calibration equations: the standard error of the residuals for the calibration (*SEC*) and for the cross-validation (*SECV*), the coefficient of determination for the calibration

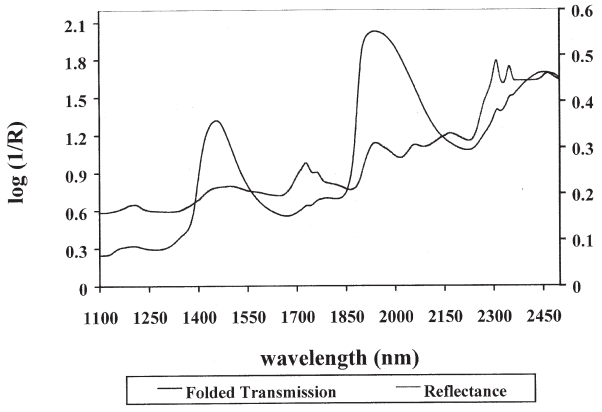

Figure 1. Folded transmission (liquid) and reflectance (dry extract) spectra of one milk sample.

Table 1. Calibration statistics obtained for quantitative analysis of ewe milk in both the Folded Transmission (FT) and Reflectance (R) modes.

Parameter	Mode	Mean	*SD*	*SEC*	*SECV*	r^2	*RPD*	*CV*
Casein	FT	5.49	0.60	0.17	0.21	0.88	2.86	3.77
	R	5.52	0.60	0.13	0.19	0.88	2.95	3.50
Protein	FT	5.95	0.68	0.18	0.19	0.92	3.58	3.21
	R	5.95	0.65	0.11	0.16	0.94	4.06	2.75
Fat	FT	8.06	2.02	0.11	0.14	0.99	14.43	1.71
	R	7.85	1.82	0.21	0.43	0.94	4.23	5.52
Total Solids	FT	17.97	2.23	0.19	0.25	0.99	8.92	1.41
	R	17.78	2.20	0.24	0.34	0.98	6.47	1.91
SCC 10^{-3}	FT	260.63	156.55	25.40	53.11	0.88	20.38	2.95
	R	278.60	169.12	25.39	55.57	0.89	19.95	3.04

(R^2) and for the cross-validation (r^2), the coefficient of variation (*CV*), calculated as (*SECV* * MEAN^{-1}) × 100 and the *RPD*, calculated as SD * *SECV*$^{-1}$.

Results and discussion

The results obtained for chemical analysis, calibration and cross-validation of selected calibration equations for each constituent in both NIR analysis modes under study are shown in Table 1.

Protein, fat and total solids calibrations have excellent capacity for quantitative analysis,[12] as their r^2 values are higher than 0.9. Casein r^2 values are slightly lower, but still are high. The equation obtained for somatic cell count (SCC) has adequate accuracy, similar to that obtained for goat's milk by Pérez *et al*.[14] (r^2 = 0.81) and much higher than the model reported by Tsenkova *et al*.[15] in cow's milk (r^2 = 0.35).

In general, both analysis modes present low prediction errors, estimated by the *SECV*, *RPD* and *CV* values. The RPD statistic values were always higher than three, recommended by Williams and Sobering[13] to consider a calibration equation as suitable to use in real conditions of process control.

The accuracy of casein, protein and SCC equations is not affected by sample analysis methods, as they present similar *SECV* values in reflectance and folded transmission modes. Nevertheless, the accuracy of folded transmission fat and total solids equations is significantly higher than the corresponding reflectance equations. Isaksson *et al*.,[9] likewise, reported lower prediction errors with the use of direct measurements (transmittance, 1 mm cuvettes) on liquid foods compared to the DESIR method. *CV* value for folded transmission protein calibration is very similar to those obtained by Albanell *et al*.[7] (4.09%) and by Pascual *et al*.[8] (2.21%) who used an analysis system (transflectance) very similar to the one employed in this study. However, the *CV* value obtained for folded transmission fat and total solids are lower than those obtained by Albanell *et al*.[7] in goat's milk (4.55% vs 3.03%, respectively).

Conclusion

In terms of accuracy and speed of analytical response, NIR analysis of liquid milk (folded transmission) is recommended instead of NIR analysis of dry extract of milk (reflectance) although both analysis modes offer satisfactory results.

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