

# Measurement of metabolic parameters in lactating dairy cows by near infrared reflectance spectroscopy analysis using cattle faecal samples

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## Introduction

Nutritional information about animal production is difficult to assess due to the difficulty in determining the amount and quality of food intake and, in particular, if forages are the most important compound in the diet for milk production. One of the most common procedures for the evaluation of nutritional value in foods is *in vivo* measurement. These metabolic studies with cows requires at least a knowledge of the amount of all food consumed and the excretion of milk faeces and urine. Taking these criteria as a basis, forage and total intake, dietary digestibility and balances of nitrogen and energy can be calculated. However, these food evaluation experiments with animals involve a lot of time and are very expensive.

The quality of individual forage varies widely. This fact makes it necessary to find alternative evaluation methods which are more rapid and cheaper to operate. An effective method for predicting grass and silages intake is an essential prerequisite to the accurate rationing of dairy cows. However, most authors have used simple or multiple regression analysis to examine the relationships between chemical composition and individual animal parameters.<sup>1</sup> In these situations near infrared (NIR) reflectance spectroscopy has produced promising results for predicting intake and biological parameters.<sup>2-4</sup>

The faeces excreted by animals contain undigested residues of the diet consumed. For this reason, their analysis can be a successful tool to determine the amount and quality of food intake and other important biological parameters.

The aim of this work has been to find out if faecal analysis by NIR could be used to determine, with enough accuracy, some attributes of the diets of different lactating dairy cows.

## Materials and methods

### Diet

The basic diets were eight grass, three grass silages and two maize silages, *ad libitum*, twice a day, supplemented with 0–5 kg concentrate cow<sup>-1</sup> day<sup>-1</sup>.

### Experimental design and animals

Six Friesian dairy cows, in the 29th week of lactation, were housed and milked in individual stalls. The animals were divided into three groups of two and allocated, at random, to three treatments ac-

cording to design,<sup>5</sup> were used in a latin square design experiment with three periods each of twenty one days duration, fifteen days for adaptation period and seven days for collection period.

During the collection period of the trial, daily samples of food offered and refused, milk, urine and faeces were collected. All subsequently were analysed. A total of 79 faecal samples were used for the initial analysis with the aim of obtaining information on a wide range of forage and total intake, organic matter digestibility, gross energy digestibility and digestible and metabolisable energy.

### NIR scanning and calibration procedures

The samples of faeces collected were freeze-dried and ground at 0.75 mm. The samples were then scanned using a Foss-NIRSystem 6500 scanning monochromator over a wavelength from 1100 to 2500 nm in steps of 2 nm. Due of the heterogeneous nature of the material, each sample was measured in two replicates and the mean of the replicate spectra obtained ( $\log 1/R$ ) was used in the calibration.

Population boundaries were established with a maximum standardised  $H$  distance from the average spectrum of 3.0.<sup>6</sup> Calibration equations were obtained by WINISI II v. 1.5 software (Infrasoft International, Port Matilda, PA, USA), using a full wavelength range and modified partial least square as the regression method. Standard normal variate<sup>7,8</sup> (SNV) was used for scatter correction and second derivative as the mathematical treatment. Cross-validation, to minimise overfitting of the equations, was used to test the calibration equations. Those were selected according to the lowest standard error of cross-validation ( $SECV$ ), the highest coefficient of determination ( $r^2$ ) and the  $RER^9$  (ratio range  $SECV^{-1}$ ).

## Results and discussion

The mean, range and standard deviation for forage and total intake (FI and TI, respectively), organic matter digestibility (OMD), gross energy digestibility (GED) and digestible and metabolisable energy (DE and ME, respectively) values for the 79 faecal samples are given in Table 1. These values are typical of metabolic trials on lactating dairy cows with diets based on green and preserved forages and confirm that the samples had a wide range for all parameters. The amount of total and forage intake decreased with progressing maturity. As a result OMD, GED, DE and ME obviously also decreased.

The untreated NIR average spectrum ( $\log 1/R$ ) of global population and symmetry plot of PCA scores for faecal samples in the wavelength range from 1100 to 2500 nm, are shown in Figure 1. The three-dimensional display of the sample scores provides a good sample distribution in the population.

In general, equation statistics were acceptable for all parameters of interest. The best NIR calibrations were obtained when the spectral data were converted into second derivative (Table 2).  $SECV$

**Table 1. Mean, range and standard deviation of reference values for calibration sets of faeces samples.**

Parameters	Range	Mean	SD
Total intake (kg DM cow <sup>-1</sup> day <sup>-1</sup> )	15.4	10.3–19.8	2.216
Forage intake (kg DM cow <sup>-1</sup> day <sup>-1</sup> )	10.9	5.3–15.8	2.654
Organic matter digestibility (%)	72.0	54.2–80.8	5.271
Gross energy digestibility (%)	66.6	52.2–74.4	4.974
Digestible energy (MJ kg DM <sup>-1</sup> )	12.2	10.1–13.8	0.864
Metabolisable energy (MJ kg DM <sup>-1</sup> )	10.1	8.2–11.7	1.012

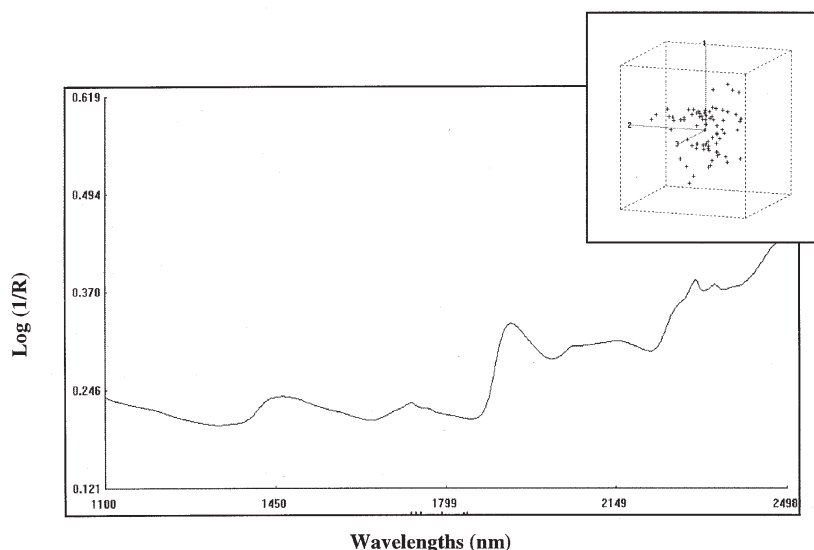


Figure 1. Untreated NIR average spectrum of global population and symmetry plot of PCA scores for faeces samples ( $n = 79$ ).

ranged from 0.434 for ME to 2.626 for OMD. Coefficients of determination to cross-validation ranged from 0.560 for TI to 0.819 for EM. Values for the *RER* ranged from 6.50 for TI to 10.13 for OMD.

The equation statistics for a direct relationships between faecal NIR spectra and intake, OMD, GED, DE and ME clearly indicate the superiority of the relationship between the composition of green and preserved forages determined by conventional laboratory methods and these parameters. Steen *et al.*<sup>1</sup> cited lower coefficients when intakes were predicted using relationships based on regressions involving a range of chemical parameters.

In this study, OMD can be predicted with a high degree of accuracy. Although several reports<sup>2</sup> have noted the limitation in the usefulness of faecal samples for predicting OMD over a wide range of for-

Table 2. Coefficients of determination for calibration ( $R^2$ ) and cross validation ( $r^2$ ), standard error of calibration (*SEC*) and cross-validation (*SECV*), with different sample sets in NIR equations.

Parameters	<i>SEC</i>	$R^2$	<i>SECV</i>	$r^2$	<i>RER</i>
Total intake (kg DM cow <sup>-1</sup> day <sup>-1</sup> )	1.349	0.629	1.467	0.560	6.50
Forage intake (kg DM cow <sup>-1</sup> day <sup>-1</sup> )	0.967	0.867	1.309	0.757	8.02
Organic matter digestibility (%)	1.994	0.857	2.626	0.750	10.13
Gross energy digestibility (%)	1.874	0.856	2.622	0.715	8.49
Digestible energy (MJ kg DM <sup>-1</sup> )	0.344	0.841	0.475	0.695	7.81
Metabolisable energy (MJ kg DM <sup>-1</sup> )	0.262	0.933	0.434	0.819	8.16

ages, these results agree with those reported by other authors<sup>10–12</sup> based on direct OMD prediction results using regression equations developed directly on forage samples.

The accuracy for predicting GED, DE and ME are acceptable. The data reported here contains an inherent variability on the reference values.

In order to know if a calibration equation is acceptable for prediction, the *RER* were calculated for each parameter. Values for the ratio range *SECV*<sup>-1</sup> ideally should be at least ten.<sup>9</sup> Only for OMD has the *RER* value been higher than ten. Nevertheless, *SECV* have not been too different from the required parameters, which are associated with a relatively high standard error.

The variation coefficients in the present study were 9.5, 12.0, 3.6, 3.9 and 4.3 % for TI, FI, OMD, GE, DE and ME, respectively, in relation to the population mean.

## Conclusion

In conclusion, faecal analysis by NIR can be used to determine diet quality. The results indicate that the forage and total intake, organic matter digestibility, gross energy digestibility and digestible and metabolisable energy can be directly predicted with enough accuracy from the NIR spectra of freeze-dried faecal samples.

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