Instantaneous quality control of foodstuffs by using NIR technology and a remote reflectance fibre-optic probe

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Introduction

Generically, feeds are naturals materials and elaborated products, of any origin, that alone or suitable mixed are able to provide adequate animal nutrition. The main quality factors of feeds are the energy value, the amount of fibre, proteins and fat, together with the different additives.

Regarding animal feed, traditional analytical methods continue to be used, although there are some studies on feed composition using near infrared (NIR); nevertheless, none of them has used a fibre-optic probe. Such studies have addressed the composition of feed for rabbits,[1] pigs[2,3] poultry,[4] milk cows,[5,6] and in all of them the authors analyse the amount of fibre, protein or the nutritional value of the feed. There are studies, too, that have used NIR to measure how the composition in protein, fibre and energy is affected by freezing, grinding or paste-heating processes,[7,8] and there are works on cereals,[9,10] oats, barley, wheat, and rye and soy in which the content of protein or fibre is predicted. The aim of the present work was to develop an instantaneous quality control protocol for animal feeds and fodder by the determination of protein, fat and fibre without prior sample treatment, using direct application of the probe on the feed.

Experimental

Samples

Seventy-two samples of animal feeds and fodder (for cattle, swine, sheep, poultry, and rabbit) used in different stages of animal rearing (juvenile development, growth, fattening, etc.) with different physical characteristics (meal, tablets, grains, granules). Figure 1 shows the different types of feed studied. Measurement of the NIR spectra was accomplished by direct application of the fibre-optic probe on the feed samples, Figure 2.

Chemical analyses

The reference chemical measurements of fat protein and fibre were obtained using conventional chemical techniques (Table 1).

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Figure 1. Samples.

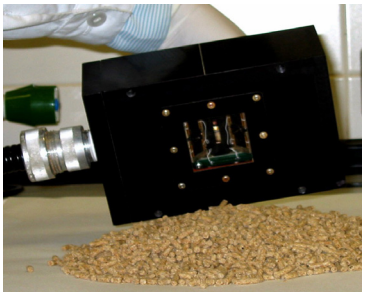

Figure 2. Fibre-optic probe.

Fat contents were determined according to ISO-1443, by extraction of fat from previously hydrolysed and desiccated (petroleum ether) samples, removing the solvents by evaporation, desiccating the residue and later weighing it after cooling. Fibre contents were determined by de-fatting the samples, separating the residue by filtration and measuring the mass loss by calcination. In the calculation of protein, fat and fibre contents, three determination were carried out, taking the mean value and discarding values in which an error value of ±5% was obtained. The chemical compositions are shown in percentage weight.

NIR spectroscopy

A Foss NIRSystems 5000, device fitted with a 1.5 m fibre-optic probe (Ref. Nº R6539-A) of the 210/210 Bundle regular type was used. The window is of quartz, with a 5 cm x 5 cm surface area, and reflectance is measured in the 1100–2000 nm near IR zone. WInISI 1.05 software was used on a Pentium III.

Results and discussion

Chemical analyses and spectral information

The chemical composition of the samples of feed used for calibration is shown in Table 1 . The results obtained in the feeds for protein contents were 11% for feed for weaned pigs and 44% for soy meal; for fat the values lay between 1.2% for soy meal and 11% for adult pigs, while for fibre the values varied between 3.2% for weaned pigs and 16.4% for rabbits. These are the usual ranges for such disparate samples. Mathematical treatment was accomplished using principal component analysis, PCA, together with the percentage of explained spectral variability. For the fat parameter, six principal components were used, accounting for 99.98%; for protein, eight components were used, explaining 99.99% of the spectral variability, while for fibre 11 components were necessary to account for 99.22%. The SNV mathematical treatment corrects the problems associated with particle size[11] while the MSC (multiplicative scatter correction) method prevents the affects of scattering from being imposed on the chemical signals[12] .Figure 3 shows the spectrum of a sample of feed subjected to the optimum treatment for protein, fat and fibre.

Table 1. Statistical overview of chemical analyses (all units in %).

	Minimum	Maximum	Mean	*SD*
Fat	1.2	11.4	4.2	1.3
Protein	11.8	44.5	19.8	9.5
Fibre	3.2	16.4	6.9	1.9

After the number of principal components had been calculated, detection of anomalous spectra was accomplished using the Mahalanobis distance (*H* statistic), establishing $H = 3$ as the limit value. Accordingly, those spectra whose *H* distance was greater than 3 were considered as outliers from the spectral population and were discarded. Four samples were eliminated for the protein parameter; six for the fat parameter and three when the fibre parameter was being determined.

The spectral information afforded a series of characteristic absorption bands. The bands were obtained by calculating the β coefficients at the maximum absorbance wavelengths—1264, 1552,1594, 1474, 1754 and 1792—associated with the protein content, while those appearing at 1314,1580,1702,1734,1754 and 1970 nm were related to fat[24] and those at 1150,1176,1248, 1298, 1344, 1484 and 1766 nm were related to fibre.

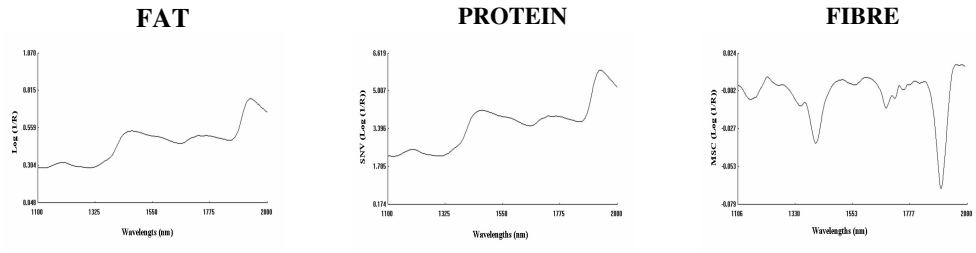

Figure 3. Fat: Corrected spectrum using SNV. Protein: NIR spectrum. Fibre: Corrected spectrum using first derivative and Standard MSC.

Determination of fat, protein and fibre

Calibration equations

For the calibration of fat, protein and fibre, 72 intact samples of feed measured with the fibre-optic probe were employed. Calculation of the statistical parameters of the calibration equations for each component is shown in Table 2. In this step, samples with higher residual values were eliminated, using the $T > 2.5$ criterion. With this criterion, six samples were eliminated for the fat parameter; four for protein and three for fibre.

The results obtained for fat were better than those corresponding to protein. This is reasonable since it is difficult to relate the results obtained by the reference analytical method to the spectroscopic data in the determination of bulk protein considered as %N × 6.25, since nitrogen does not show a vibrational response in NIR. However, it is possible to measure the vibrations of N–H bonds –part of the protein molecule- in the near IR. In sum, NIR and the Kjeldhal reference method do not measure the same type of species and the correlation between both methods may vary as a function of the type of sample involved.[13] The results for fibre are good in view of the tediousness involved in its chemical determination.

Table 2. Calibration statistical descriptors for the NIR determination of fat, protein and fibre in feed samples.

Component	Mathematical treatment	*RSQ*	*SEC*	*SECV*	*SD*	Range
Fat	2/8/6/1 SNV	0.932	0.213	0.278	0.816	1.6–6.6
Protein	2/8/6/1 SNV+DT	0.991	0.948	1.096	9.845	0.5–49.50
Fibre	2/4/4/1 Stand MSC	0.848	0.575	0.917	1.473	2.3–11.1

Validation

Internal validation (prediction)

Evaluation of the calibration model was performed by cross-validation. In this method, the set of calibration samples is divided into groups, using one of them to check the results (prediction) and the others to construct the calibration model. The model is repeated as many times as there are groups, such that all pass through the calibration set and the prediction set. The same process is performed for each of the parameters. The number cross-validation groups were four for protein and fat and six for fibre. The predicted values gave validation errors that were reflected in the *SEP*(*C*) values. Figure 4 shows the correlation of the values obtained at the laboratory with respect to what was predicted by the NIR method for the fat, protein and fibre parameters of the feed samples with measurements made with the fibre-optic probe.

External validation

To check the robustness of the calibration method, an external validation was performed with a set of ten feed samples that did not belong to the calibration set. Of these, the reference or laboratory values were known. The aim was to analyse how each of the equations obtained would predict the fat, protein and fibre parameters by the calibration and then to compare the results obtained with the laboratory results. The results concerning the statistical descriptors for the external validation are shown in Table 4. It may be seen that the parameters are of the same order as those obtained with the calibration set, confirming the goodness of the proposed procedure.

In view of the results, it may be concluded that NIR technique with the aid of a remote reflectance fibre-optic probe offers a good alternative for the determination of fat, protein and fibre by direct application of the probe on the feed sample without any prior sample treatment and with calibration equations contrasted with unknown samples.

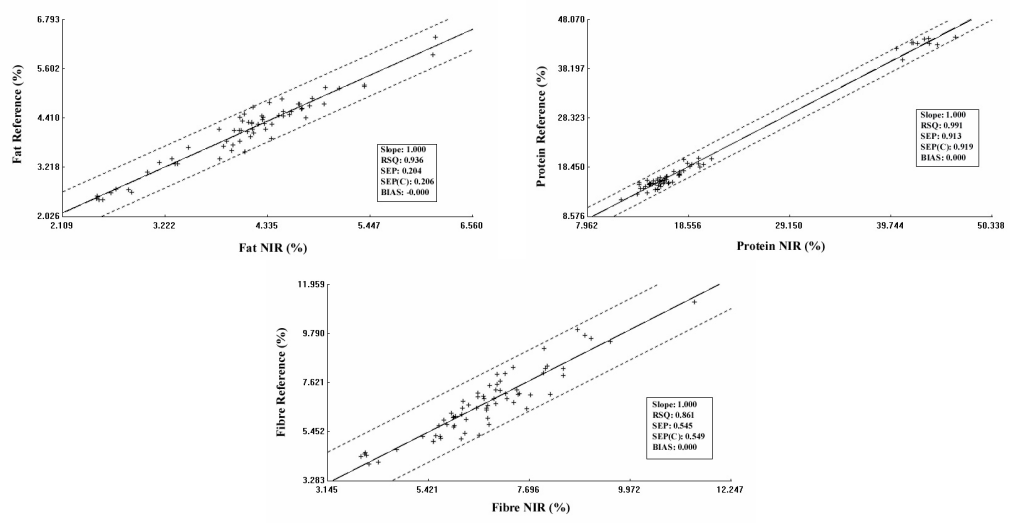

Figure 4. Comparison of reference values with the values predicted by the calibration equations for fat, protein and fibre.

Table 4. Statistical descriptors of the external validation for the determination of fat, protein and fibre in feed using NIR (%).

Components	Range	*SD*	*SEP*	*SEP*(*C*)	Bias	Differences,%
Fat	2.07–4.8	0.994	0.218	0.209	0.092	4.1
Protein	14.5–45.6	12.5	1.190	1.251	–0.079	3.0
Fibre	4.4–7.8	1.2	0.624	0.605	–0.244	5.4

References

1. C. Xiccato, A. Trocino, A.Carazzolo, M. Meurens, L. Maertens and R. Carabaño, *Anim. Feed Sci. Tech*. **77,** 201(1999).
2. E.V. Valdes and S. Leesson, *Poultry Sci.* **71,** 1179 (1992).
3. S.L.Y. Chen, A. Hsu and M.L. Lee, *J. Assoc Off. Anal. Chem.* **70,** 420 (1987).
4. J. Aufrère, D. Gravion, C. Demarquilly, J.M. Pérez and J. Andrieu,. *Anim. Feed Sci. Tech.* **62,** 77 (1996).
5. A. Purnomoadi, K. Batajoo, K. Ueda and F. Terada, *Int. Dairy J.* **9,** 447 (1999).
6. R. Biston, N. Bartiaux-Thrill, A. Thewis, E. François, P. Limbourg, M. Giels and P. Dardenne, *Reprod. Nutr. Dev.* **28,** 61 (1988).
7. E.R. Deaville and D.I. Givew, *Anim. Feed Sci. Tech*. **72,** 41 (1998).
8. D. Alomar, R. Montero and R. Fuchslocher, *Anim. Feed Sci. Tech.* **78,** 57 (1999).
9. D. Alomar, R. Fuchslocher and J. Stockebrands, *Anim. Feed Sci. Tech*. **80,** 309 (1999).
10. A.M. Bruno-Soares, I. Murray, R.M. Paterson and J.M.F. Abreu, *Anim. Feed Sci. Tech*. **75,** 15 (1998).
11. M.S. Dhanoa, S.J. Lister and R.J. Barnes, *Appl. Spectrosc*. **49,** 765 (1995).
12. P. Geladi, D. Mac Dougall and H. Martens, *Appl. Spectrosc.* **39,** 491 (1985).
13. A. Garrido-Varo, R.Carrete and J. Fernández-Cabanas, *J. Near Infrared Spectrosc.* **6,** 89 (1998).