

## USE OF NIR TO ESTIMATE NITROGEN DEGRADABILITY OF FORAGE.

R. Agneessens, P. Dardenne and Ph. Lecomte.

Station de Haute Belgique (Centre de Recherches Agronomiques de Gembloux), rue du Serpont 100, B-6800 Libramont, Belgique.

### ABSTRACT

Near infra red spectrometry (NIRS) is commonly used to characterise animal feeds at the level of their intrinsic composition (dry matter, protein, fat, fiber) as well of their digestibility (Dardenne et al., 1991). The use of new systems in the evaluation of the protein value has induced the necessity to determine parameters such as the theoretical ruminal degradability of nitrogen (DT).

The definition of this criterion necessitates the implementation of the nylon bag technique which is time consuming and costly and is reserved to reference laboratories.

An alternative to this technique, based on an enzymatic hydrolysis of the proteinic content, has been developed by Aufrère et al. (1988) whose work has lead to define a relationship between the theoretical degradability measured in sacco (DT) and the enzymatic degradability of proteins (DE) for compound feeds.

Resuming their work, Kamoun (1995) has adapted the methodology to forage substrates and determined for the former a same relationship.

The study presents for forage substrates a predictive model of the enzymatic degradability of nitrogen by Near Infra Red Spectrometry, based on 69 samples of wilted grass silage analysed by the reference method.

Characteristics of this model are SE 2.24, R<sup>2</sup> 0.74.

### INTRODUCTION.

In most of the estimation systems of the protein value of ruminant feeds e.i. the French systems (PDI) or Dutch system (DVE/OEB), knowing the theoretical ruminal degradability of nitrogen (DT) is essential (Aufrère et al., 1990).

The reference method to study this parameter is based on the nylon bag technique which is long, costly and has a poor reproducibility, proves with difficulty applicable as a routine method and is reserved to a limited number of laboratories. For these reasons it was interesting to have an easier replacement method.

Works realised by Aufrère et al. (1988) have defined for compound feeds the relationship existing between the DT and the enzymatic degradability (DE) of nitrogen (hydrolysis of the substrate by a protease extracted of *Streptomyces Griseus* in a borate -phosphate buffer at pH8 during 1 hour).

Subsequently works undergone by Kamoun (1995) have allowed, not only, to widen the field of application of this enzymatic method to various forage substrates and define for these the relationship between DE and DT, but also to put forward the potential of the Near Infra Red Spectrometry (NIRS) technique to predict that DE parameter.

The aim of this study is to test the real adaptability of NIRS to predict the criterion DE on a range of grass wilted silage.

### MATERIAL AND METHODS.

Studied samples are prewilted silages harvested in 1993 and 1994 in

various farms of southern Belgium. The samples collected with a probe have been dried during 48 hours at 60°C and milled cyclotec (1mm). The chemical composition and quality parameters have been determined by reference methods particularly for this study: the analytical dry matter (MSA), the crude protein content (CP) and the enzymatic degradability of protein (DE).

The dry matter has been determined according to AFNOR norm, the crude protein content according to the method Kjeldahl and the enzymatic degradability of nitrogen according to the method described by Kamoun (1995) (hydrolysis of the substrate by a protease extracted of *Streptomyces Griseus* in a borate - phosphate buffer at pH8 during 24 hours).

The NIRS analysis has been performed on ground dried samples with a NIRSYSTEMS 5000 monochromator collecting reflection data's between wavelengths of 1100 to 2500 nm.

Mathematical treatment of the optical data was optimised using the ISI-NIRS-3 software (Infrasoft International, Port Matilda, PA, USA). Calibration equations were obtained using a modified Partial Least Squares (MPLS) regression technique Shenk J.S. and Westerhaus M.O. (1993). This regression technique requires cross-validation to prevent overfitting. Cross-validation obtains validation errors by partitioning the calibration set into several groups. A calibration is performed for each group, reserving that group for validation and calibrating on the remaining groups, until every sample has been predicted once. The validation errors are combined into a standard error of cross-validation (SECV).

Calibration equations are optimised by selecting the mathematical treatment of the log 1/R optical reflection signals. The first and second derivatives were obtained by the segment-gap method and optimised by changing segments in gaps.

### RESULTS AND DISCUSSION.

The best equation to predict DE has been obtained with a PLS regression and a mathematical treatment on the reflexion spectra's in first derivative, a gap of 5, and a smoothing of 5 and 3. The main parameters of the model are presented in the table 1 where SE<sub>c</sub> and R<sub>c</sub><sup>2</sup> are the residual standart error and determination coefficient when calibrating on the whole set of data's; SE<sub>cv</sub> and R<sub>cv</sub><sup>2</sup> the same parameters according to cross validation technique.

Figure 1 shows the relation existing between values observed in laboratory and values predicted by NIRS.

With a determination coefficient of 0.74 and a residual error of 2.2, slightly higher than the reference enzymatic method (1.8) the equation appears useful to estimate the nitrogen degradability of forages.

These results strengthen the potential offered by the technique to predict the enzymatic degradability of the nitrogen of forage substrates and this with all the advantages that the technique presents that is to say: rapidity, non destructive and non pollutant character as well as reduced analytical cost.

## REFERENCES

**Aufrère J. et Cartailier D.** (1988) Mise au point d'une méthode de laboratoire de prévision de la dégradabilité des protéines alimentaires des aliments concentrés dans le rumen. *Ann. Zootech.*, **37** (4): 255-270.

**Aufrère J. et Michalet-Doreau B.** (1990) Nouvelles méthodes d'estimation de la valeur alimentaire des fourrages II. Méthodes enzymatiques. *Fourrages*, **122**: 203-217.

**Aufrère J., Graviou D., Demarquilly C., Vérité R., Michalet-Doreau B. et Chapoutot P.** (1989) Prévision de la valeur azotée PDI à partir d'une méthode enzymatique standardisée INRA Prod. Anim., **2** (4): 249-254.

**Biston R. et Dardenne P.** (1985) Application de la spectrométrie dans le proche infrarouge. Prévision de la qualité des fourrages en vue de leur exploitation rationnelle *Bull. Rech. Agron. Gembloux*, **20** (1/2): 23-41.

**Dardenne P., Sinnaeve G., Biston R. and Lecomte P.** (1991) *Making Light Work: Advances in Near Infrared Spectroscopy* Ed. by Murray I. and Cowe I. 4th ICNIRS, Aberdeen, Scotland. VCH, pp. 277-283.

**Kamoun M.** (1995) Utilisation de l'azote-15 et de la spectrométrie dans le proche infrarouge pour la détermination de la dégradabilité réelle in situ et in vitro des matières azotées des fourrages. Dissertation originale en vue de l'obtention du grade de docteur en sciences agronomiques (faculté des Sciences Agronomiques de Gembloux).

**Shenk J.S. and Westerhaus M.O.** (1993) *Analysis of Agriculture and Food Products by Near Infrared Reflectance Spectroscopy*. Monograph, Infrasoft International, Port Matilda, PA, USA, p. 103.

**Table 1**

Parameters of the model predicting enzymatic degradability of forages according to NIRS technique.

N	Range	Mean	SE <sub>C</sub>	R <sup>2</sup> <sub>C</sub>	SE <sub>CV</sub>	R <sup>2</sup> <sub>CV</sub>
69	56.66-78.44	68.76	1.87	0.82	2.24	0.74

**Figure 1**

Relation between observed and NIRS predicted values of DE.

