

The contribution of near infrared spectroscopy to the fight against the mad cow epidemic

Dr Vincent Baeten^a and Dr Pierre Dardenne^b

^aFirst Research Assistant and ^bDirector at the Quality of Agricultural Products Department of the Agricultural Research Centre of Gembloux (CRAGx, Belgium). E-mail: dardenne@cragx.fgov.be

In June 2001, the European Commission decided to extend its ban on meat and bone meal (MBM) in feedstuffs. This decision has confirmed and extended the Commission Decision of 29 December 2000 specifying the ban on certain materials in animal feeds after the BSE (Bovine Spongiform Encephalopathy) crisis has suddenly re-appeared. The prohibition on the use of protein derived from all mammalian tissues in feedstuffs was initially stated by the Commission Decision 94/381/EC after the outbreak of the "mad cow" epidemic and its probable relationship with new variant Creutzfeldt-Jacob disease in humans. In fact, all processed animal proteins are forbidden from use in the feeding of all animals. Exceptions are made for some processed animal proteins (fish meal and hydrolysed animal proteins) and dicalcium phosphate which may be used to feed animals other than ruminants. To be implemented and policed, the ban requires analytical methods for control laboratories throughout Europe. To propose analytical solutions to this problem, a consortium of 10 European partners (see Figure 1), led by the Agricultural Research Centre of Gembloux, submitted a research proposal to Brussels. The STRATFEED project (www.stratfeed.cragx.be) has been accepted and financed by the European Commission. The project is entitled "Strategies and methods to detect and quantify mammalian tissues in feedstuffs" and regroups teams involved in the analysis of feed using different techniques. The STRATFEED project is centred around three techniques: classical microscopy, PCR methodology using the last developments in molecular biology and near infrared (NIR) spectroscopy.

The classical microscopic method is currently the only official method for the detection of MBM in feedstuffs accepted throughout Europe.¹ This method is based on the microscopic identification and estimation of sieved and decanted fractions of particles (mainly bones) of animal origin in feedstuffs. Since the examination is visual, the results depend on the analyst's knowledge of various features of the ingredients and



Figure 1. The STRATFEED consortium.

the application of the microscopic technique. The speed of the method is around four to six samples a day. The DNA technology (PCR) method is based on the detection of taxon-specific DNA sequences from a number of heterogeneous matrices. By using the PCR-based procedures and appropriate primer pairs, the technology allows a rapid and sensitive detection of taxon specific DNA-sequences from MBM. Two additional methods based on infrared spectroscopy were added: NIR spectroscopy and NIR microscopy.

NIR spectroscopy meets all the criteria of speed of response, reliability, cost-effectiveness and fitness for a radically new approach to qualifying raw materials (ingredients) and finished feed products. It can analyse a sample in a few seconds even for multiple constituents and, importantly, it is non-destructive. This feature is important since it permits further analysis or a second expert assessment. When integrated in any official quality control system in trade or industry, NIR spectroscopy will allow the number of samples analysed to increase and provides an instantaneous method to detect and flag suspected materials. The application of NIR spectroscopy to the detection and quantification of MBM in feed is mainly based on two preliminary and pioneering works done by Professor Ana Garrido from the University of Córdoba and Dr Ian Murray from the Scottish Agriculture College, Aberdeen, both members of

the STRATFEED consortium. In their feasibility study on the use of the quantitative prediction of MBM added to a feed compound and presented in 1998 at the Workshop "Identification of animal ingredients in compound feed focusing on the microscopic method for identification" of the CEMA group held in Lingby,² Professor Ana Garrido and Victor Fernández-Cabanas have shown the possibility to calibrate a NIR spectrometer for the quantification of MBM in feedstuffs. At the same time, Dr Ian Murray has prepared an internal report entitled "Authentication of fish meals by the near infrared reflectance spectroscopy with particular reference to the absence of land animal tissues". This work will be followed by a publication in *Journal of Near Infrared Spectroscopy*. These studies have demonstrated the potential of NIR spectroscopy in the detection of MBM in fish meals. In the framework of the STRATFEED project, the NIR approach will be developed and validated as a screening method to classify unknown samples and also to detect and quantify the addition of mammalian tissues in feedstuffs. In addition to the teams of Professor Ana Garrido and Dr Ian Murray, the teams of Rob Frankhuizen from the State Institute for Quality Control of Agricultural Products in Wageningen (RIKILT, The Netherlands), Alba Puigdomenech from the Laboratory of the Autonomous Government of Catalonia in Barcelona (LAGC, Spain), Jos Zegers from Nutreco (The

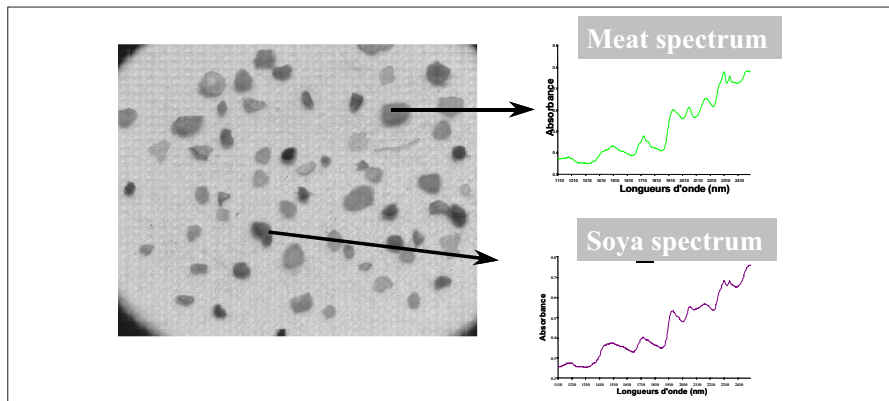


Figure 2. Sample particles spread on the sample holder and NIR spectra of a particle of meat and a particle of soya meal.

Netherlands) and Pierre Dardenne from the Agricultural Research Centre in Gembloux (CRAgX, Belgium) are collaborating in the STRATFEED project to achieve this objective and establish a European network of NIR instruments for the detection and quantification of MBM.

The NIR microscopy method consists of the analyses of several hundred particles produced by grinding of a compound feedstuff. The sample particles are spread on a sample holder and a spectrum of each particle is successively collected. A microscope connected to a Fourier transform near infrared (FT-NIR) instrument is used to collect the spectra of each particle. Figure 2 shows the sample holder and the spectra of two ingredients allowed and prohibited in the formulation of feedstuffs. Since 1998, the Agricultural Research Centre (CRAgX) has developed a method based on NIR microscopy to detect and

quantify meat and bone meals in feedstuffs.^{3,4} Actually, a database of about 10,000 spectra of single particles obtained from about 300 different animal and vegetable ingredients supplied mainly by the Belgian Ministry of Small Enterprises, Traders and Agriculture as well as local Belgian feed producers has been constructed. This reference library is used to construct different discriminant equations using the PLS and ANN approaches and allows the detection and the quantification of meat and bone meal in feedstuffs. The great advantage of this technique is that the recognition is not dependent on the expertise of the analyst. The spectral signature of each particle can be stored and used in further investigation and it is possible to automate all the procedures. In the framework of the STRATFEED project, a NIR microscopic method for the rapid and reliable detection and quantification of animal meals

in feedstuffs will be developed and validated. In addition to the CRAgX team, the team of Christoph von Holst from the Joint Research Centre (JRC, Ispra, Italy) is involved in these researches. The transfer between instruments and the standardisation procedure will be established between NIRM instruments.

References

1. Commission Directive 98/88/EC establishing guidelines for the microscopic identification and estimation of constituents of animal origin for the official control of feed-ingstuffs. *Official Journal*, L 318, 27/11/1998 p. 0045 (1998).
2. Workshop on "identification of animal ingredients in compound feed focusing on the microscopic method for identification" organised by the CEMA group, 25–26 May 1998. DG XII. Contract SMT4-CT97-6524, pp. 1–8 (1998).
3. F. Piraux and P. Dardenne, "Feed authentication by near-infrared microscopy", in *Near Infrared Spectroscopy: Proceedings of the 9th International Conference*, Ed by R. Giangiacomo and A.M.C. Davies. NIR Publications, Chichester, UK, p. 535 (2000).
4. V. Baeten, A. Michotte and P. Dardenne, *Analysis of feedingstuffs by FT-NIR-Microscopy*. Poster presented at 10th International Diffuse Reflectance Conference (IDRC-2000), Chambersburg, PA, USA, 13–18 August 2000.