

# The NIR camera: a new perspective for meat and bone meal detection in feedingstuffs

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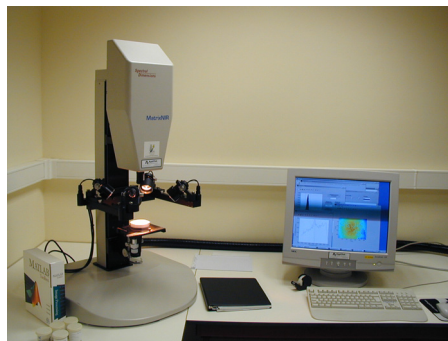
## Introduction

With the emergence of the BSE crisis in the beginning of the nineties in Europe, authorities have taken lots of legal decisions.<sup>1</sup> The respect of these laws requires analytical tools to fight against fraud and accidental contaminations. Originally, classical microscopy was the only method available for the detection of meat and bone meal (MBM) in feedingstuffs. The necessity of fast and reliable methods led to the development of alternative methods like molecular biology methods (PCR and ELISA), chromatographic methods (HPLC) and spectroscopic methods (NIR).

In 1998, the Quality of Agricultural Products Department proposed the use of near infrared microscopy (NIRM). This method showed good results for the discrimination of the different ingredients making up feedingstuffs. In 2001, the limitation of the sequential collection of spectra (particle by particle) led us to use a recent and high-performance technology: the near infrared imaging.<sup>2</sup> This instrument as the NIRM combines the advantages of spectroscopic and microscopic methods.<sup>3</sup>

## Material and method

The principle of the instrument is simple. Four lamps light the surface where the sample is spread. The reflected light goes through two liquid crystal tuneable filters (LCTF) and finally hits the camera made of 76 800 pixels (240 × 320). LCTF are set in such a way that they select light with steps of 10 nm in the spectral range from 900 nm to 1700 nm.



**Figure 1. Camera Matrix NIR**

For each pixel, the compilation of the images taken at each wavelength gives a spectral cube. For absorbances at each wavelength gives a spectrum.

The spectral bank contains 5521 spectra collected on 111 different samples. Plant particles came from 21 different vegetal species covering three major classes of ingredients; i.e. rich in fat, starch and animal particles came from large range of meat and bone meals (mammalian meals, poultry by-products and fish meals). Each particle covered a surface equivalent to approximately 30 pixels and the corresponding spectra were averaged.

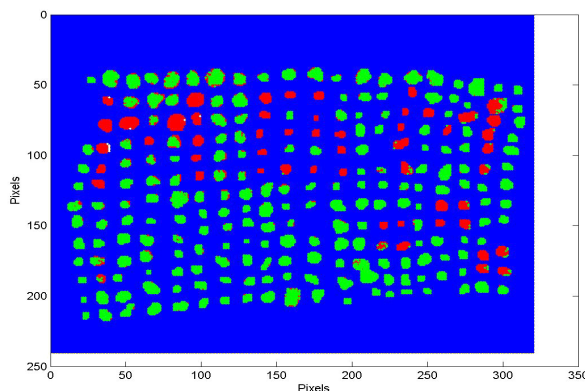
## Results and discussion

Chemometric methods such as PLS or ANN can be applied in order to build reference equations for the discrimination of animal particles from plants ones. For this, two groups of spectra have been created for the development of the equations. Spectra coming from animal particles were merged together in the first group while the second group contains the plant spectra. For the discrimination, a value of 1 and  $-1$  was attributed respectively to each group.

PLS was performed on the spectral bank collection. The *RMSE* (root mean square error) is determined by leave-one-out cross-validation. The optimal model complexity is selected as the one with the minimum *RMSE*. For this data set, the model requires 15 factors with a *RMSE* of 0.397.

ANN was also applied using the back-propagation procedure. The optimal topology of the network has been determined on the calibration set and it is found to be [15:7:1], i.e. 15 input nodes, 7 nodes in the hidden layers (hyperbolic function) and one output layer (linear function) with a *RMSE* of 0.184.

These models can now be applied on new data test sets. The first one is a sample made of animal particles displayed on the surface in such a way that it forms the letters MBM and three dots. Figure 2 shows the results after applying the PLS equation. Red pixels correspond to the spectra classified by the model as animal particles, green pixels correspond to plant particles and the white pixels are samples that can not be attributed to any group. These samples are outside of the confidence limits of the group (95% level).



**Figure 2. Result of PLS on « MBM model »**

The second data set was a group of samples with an increasing amount of MBM from 0.1% until 8.0%. Most of these samples were provided by the STRATFEED European project. Applying PLS,

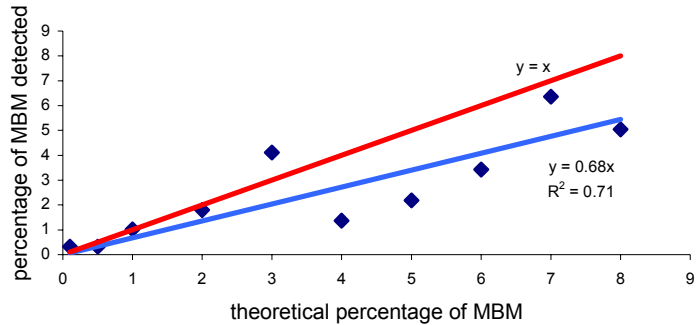
the results of animal versus plant spectra are displayed in Table 1. It shows that the technique allows the detection of animal particles at a level of contamination as low as 0.1%. Three sub-samples (repetitions a, b and c) were analysed for each sample. The number of particles analysed was adjusted for the sample theoretically containing 0.1% in order to reach the limit of detection. One can see that there is a relative coherence for the number of particles detected between sub-samples. From the sample containing 1% to 8% of MBM, taking only one sub-sample would have been enough.

**Table 1. Analytical results of the NIR imaging analysis of ten samples with a concentration included in 0.1% to 8% range.**

Figure 3 is a graphical representation of the results of Table 1. It shows that the detection of MBM

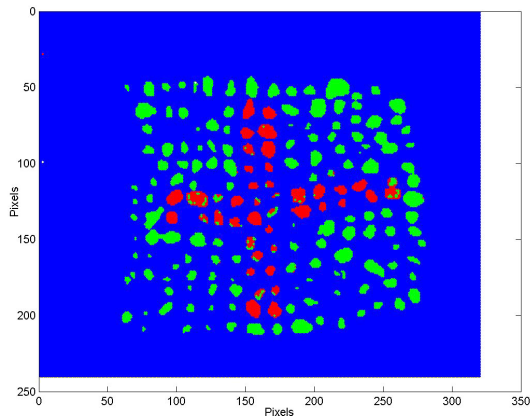
Theoretical pourcentage	Repetitions	Number of animal particles	Number of particles measured	Total		Detected pourcentage
				Number of animal particles	Number of particles measured	
0.1%	a	1	240	9	2793	0.3%
	b	1	263			
	c	0	254			
	d	0	270			
	e	2	291			
	f	2	302			
	g	0	262			
	h	1	297			
	i	1	286			
	j	1	328			
0.5%	a	0	332	3	930	0.3%
	b	1	286			
	c	2	312			
1%	a	3	308	9	883	1.0%
	b	3	300			
	c	3	275			
2%	a	4	200	12	668	1.8%
	b	4	240			
	c	4	228			
3%	a	7	254	32	779	4.1%
	b	13	254			
	c	12	271			
4%	a	5	272	12	876	1.4%
	b	3	304			
	c	4	300			
5%	a	8	340	21	960	2.2%
	b	4	287			
	c	9	333			
6%	a	14	367	37	1079	3.4%
	b	14	392			
	c	9	320			
7%	a	13	204	38	597	6.4%
	b	11	170			
	c	14	223			
8%	a	7	269	41	814	5.0%
	b	17	271			
	c	17	274			

is semi-quantitative. One can see that there is a trend to under-estimate the MBM percentage. This can be explained by the difference of density between plant and MBM particles.



**Figure 3. Graphical results from Table 1**

In Figure 4, the ANN equation was applied on a positive sample consisting of several animal and vegetal particles displayed in a « cross » shape. From the figure, one can see that the animal particles can easily be detected by ANN model.



**Figure 4. Result of ANN on « Cross model ».**

## Conclusion

This study has demonstrated that the combination of the NIR camera and the most advanced chemometric techniques is a powerful tool for the detection of animal particles in feedingsuffs.

Depending on the number of analysed particles the technique can reach a limit of detection of 0.1%. There is a balance to find between the time needed to measure a given number of particles and the limit of detection desired.

This work has shown that PLS and ANN give promising results for the discrimination of animal and vegetal particles.

## Acknowledgements

The authors wish to gratefully thank the Public Federal Service (RCS-S6112, P01/03(376)-C03/07, P01/03(376)-C03/08) and the Agricultural Research Centre of Gembloux for their logistical and financial support.

Thanks are also due to the collaborators to the UE STRATFEED project G6RD-2000-00414 for their useful contribution to the elaboration of the sample bank.

This work wouldn't have been possible without the enthusiastic and organised implication of Isabelle Fissiaux.

## Reference

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