Detection of ergot bodies in cereals by near infrared spectroscopy and hyperspectral near infrared imaging

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Introduction

Contamination of cereals with ergot, formed by the fungus *Claviceps purpurea,* is well known. For the farmer, the damage caused by ergot is a yield reduction: the ergot replaces the kernels in the grain ears. For the feed/food sector, the presence of ergot in feeding-stuffs and agro-food products involves high toxicity risk for animals and humans, in relation to the alkaloid composition and content in the ergot. The neuro-toxic signs comprise feed/food refusal, dizziness, and also convulsions. A survey on the presence of undesirable botanic substances in feed, carried out in 2006 inside official control labs from all member states of the European Union, showed a resurgence of the ergot presence in cereal samples.[1] To reduce the risk of poisoning, the European directive 2002/32/EC on undesirable substances in animal feed fixed a limit in the EU of 0.1% for ergot in all feeding-stuffs containing unground cereals.[2] The Commission is preparing regulations for ergot alkaloids in food and feed, but methods of analysis are still lacking.[3] The existing microscopy method provides an elegant early warning tool for ergot contamination but is time-consuming. The current work, performed partially in the framework of the CON*ff*IDENCE project (http://www.conffidence.eu), aims to assess, by NIR spectroscopy and hyperspectral NIR imaging, the presence of ergot bodies in cereals.

Materials and methods

For this experiment, ergot bodies issued from different sources (Belgium, The Netherlands, Germany and Denmark) and wheat kernels issued from several varieties and Belgian locations have been collected and analysed with two NIR instruments: the Bruker MPA and the MatrixNIR hyperspectral NIR imaging system. The Bruker MPA is a NIR spectrometer active in the 1100–2400 nm range, and allows the collection of a mean spectrum of a kernel bulk in less than 1 min. Each sample was measured 5 times. In total, 70 spectra were acquired with the Bruker MPA,

30 wheat spectra and 40 ergot spectra. The MatrixNIR (Malvern instruments Ltd) is a hyperspectral NIR imaging system recording sequential images with an InGaAs array detector (240×320 pixels), and is active in the 900–1700 nm range.[4] A total of 76,800 spectra were acquired in 5 minutes over the sampling area measured. Each image included 10 wheat kernels or 4 to 12 ergot bodies, depending on the sample availability. A total of around 3,000 spectra from each kernel was obtained. In total, 21 images were acquired with the hyperspectral NIR imaging system, and 120 mean spectra were calculated, corresponding to 60 wheat kernels and 60 ergot bodies. The mean spectrum of each kernel was calculated by the application of a morphological mask obtained through a process of erosion, to determine the contour of each kernel. Isys software was used for the image processing. The data treatment was carried out with MS-Excel 2007 and with the PLS toolbox under Matlab 7.5.0 (R2007b).

Results

For the study using the Bruker MPA, a calibration set (25 ergot bodies spectra and 20 wheat kernel spectra) and a validation set (15 ergot bodies spectra and 10 wheat kernel spectra) were

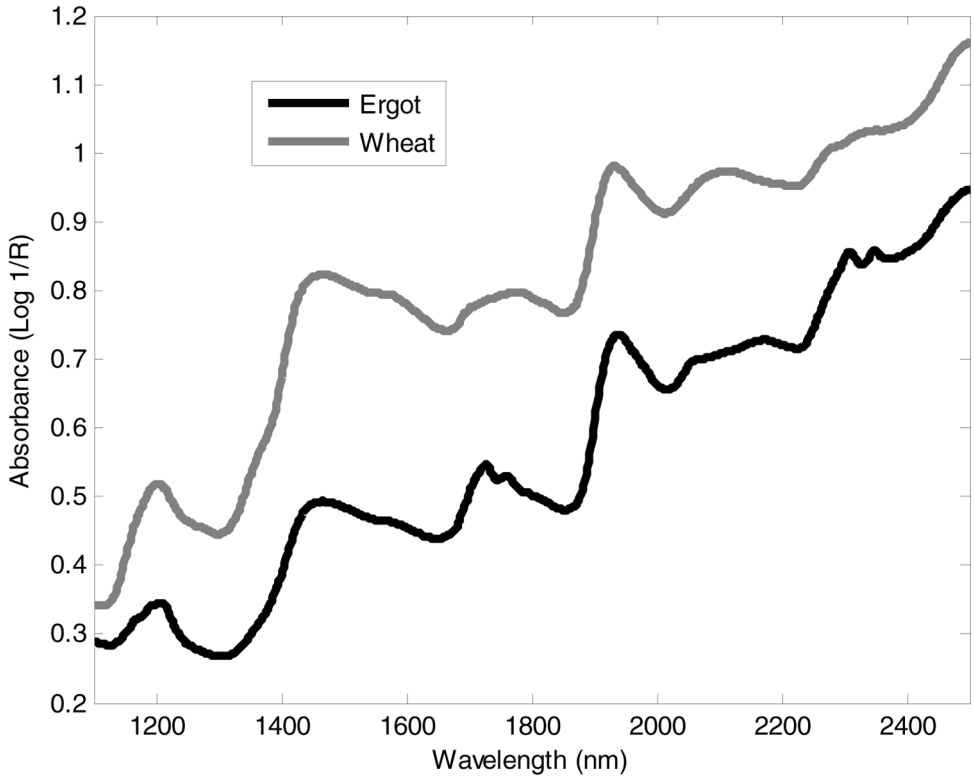

Figure 1. Bruker MPA mean spectra for wheat kernel (grey) and ergot body (black).

built from the database by selecting, for the validation set, samples from different sources than the calibration set. Figure 1 shows the mean raw spectra for ergot and wheat. From this figure it can be observed that the raw spectra for ergot are different than the raw spectra for wheat in several spectral ranges, around 1200 nm, 1400 nm, 1800 nm, 2100 nm and 2300 nm.

The data were pre-processed by the Standard Normal Variate transform followed by 1st derivative Savitzky-Golay treatment (15, 2, 1). Figure 2 shows the pre-processed spectra. From this figure it can be confirmed that the pre-processed spectra of ergot are different from the wheat spectra in the same spectral ranges.

In order to discriminate between ergot bodies and wheat kernels, the Fisher coefficient was used to select the wavelengths where the between-classes variation, i.e between ergot and wheat, was higher than the within-classes variation. Two wavelengths, 1748 nm and 2126 nm were selected, based on the specific spectral region of the ergot, and on the Fisher coefficient value. Figure 3 shows the discrimination between ergot bodies and wheat kernels using in the X axis the preprocessed data near 1748 nm, and in the Y axis the preprocessed data near 2126 nm. The validation samples (empty circles for wheat kernels and squares for ergot bodies) are included or very close to the ellipse corresponding to the 95% confidence limit. From this figure it can be

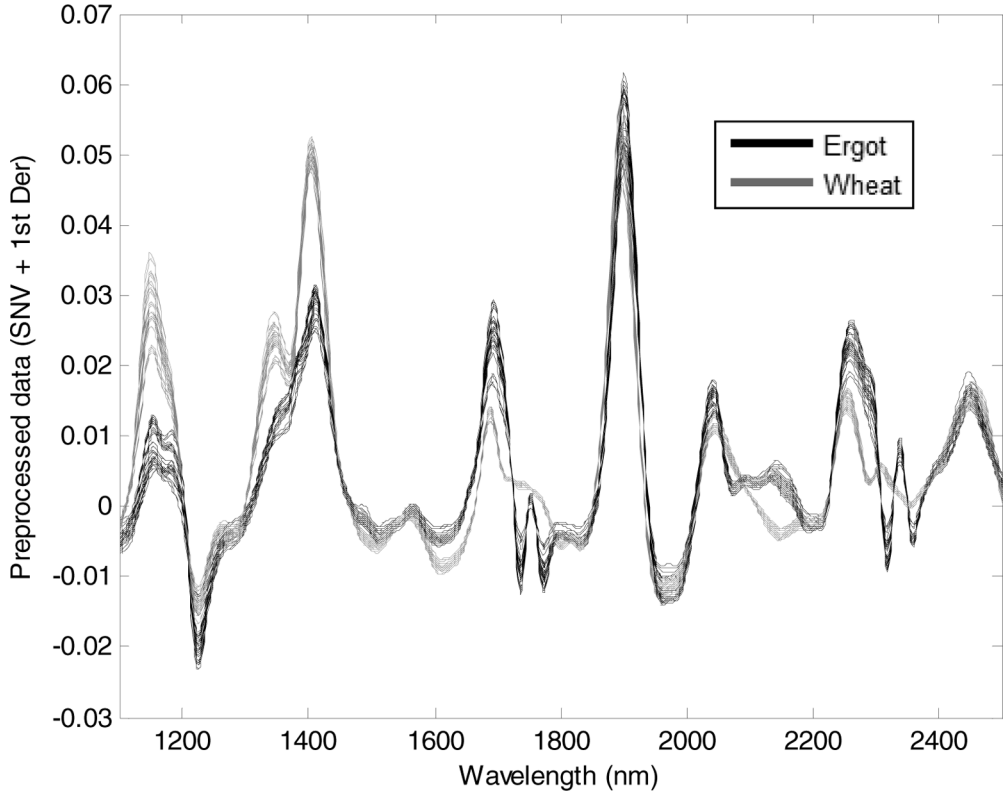

Figure 2. Preprocessed spectra (SNV + Derivative (15.2.1)) for wheat kernel (grey) and ergot body (black).

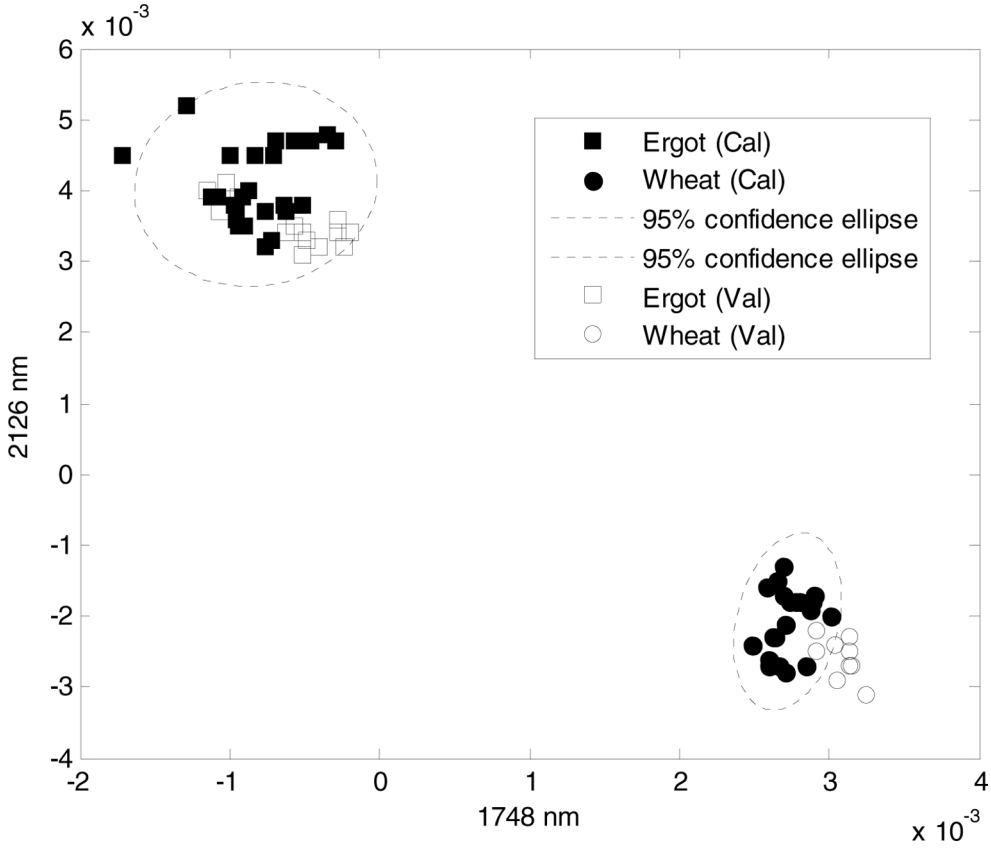

Figure 3. Bruker MPA: Discrimination Ergot/Wheat based on 2 wavelengths (1748 nm/2126 nm).

concluded that ergot bodies can be easily discriminated from the wheat kernels using 2 wavelengths of the spectra acquired with the Bruker MPA.

A similar work was carried out on the hyperspectral NIR imaging system, using the same set of samples. For this study, a calibration set (40 ergot bodies spectra and 40 wheat kernels spectra) and a validation set (20 ergot bodies spectra and 20 wheat kernels spectra) were built from the database by selecting, for the validation set, samples from different sources than the calibration set. The data were pre-processed by the Standard Normal Variate transform, followed by 1st derivative Savitzky-Golay treatment (7, 2, 1). The Fisher coefficient was calculated on preprocessed data for the wavelength range of the NIR camera. Two wavelengths, 1220 nm and 1440 nm were selected. Figure 4 shows the discrimination between ergot bodies and wheat kernels, using in the X axis the preprocessed data near 1220 nm and in the Y axis the preprocessed data near 1440 nm. The validation samples (empty circles for wheat kernels and squares for ergot bodies) are clearly included inside the ellipse corresponding to the 95% confidence limit. From this figure it can be concluded that ergot bodies can be easily discriminated from the wheat kernels using 2 wavelengths of the spectra acquired with the hyperspectral NIR imaging system.

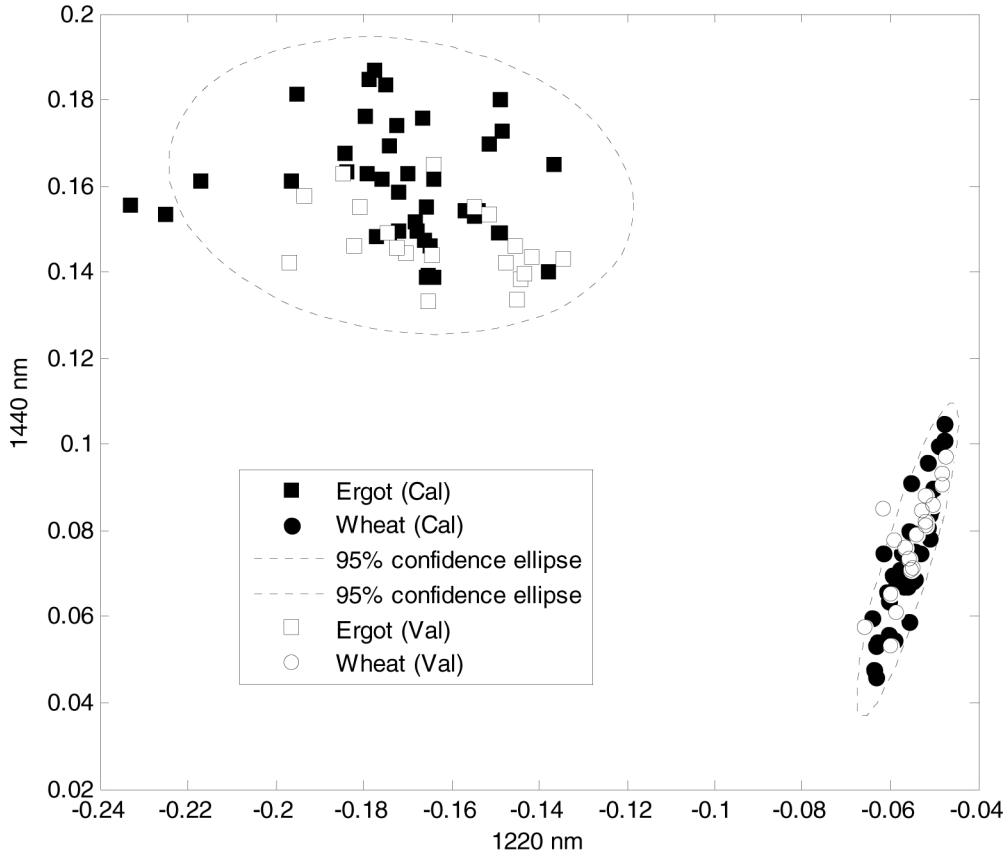

Figure 4. MatrixNIR: Discrimination Ergot/Wheat based on 2 wavelengths (1220 nm/1440 nm).

Conclusion

This study showed the potential of NIRS and hyperspectral NIR imaging to discriminate ergot bodies from wheat kernels, based on 2 wavelengths selected from the specific spectral region of the ergot, and the Fisher coefficient value. Additional developments on hyperspectral NIR imaging will be undertaken for the quantification of ergot bodies in the samples.

Acknowledgements

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