A VERIFICATION OF THE POSSIBILITY OF MYCOTOXIN DETERMINATION IN BARLEY CARYOPSES BY NEAR-INFRARED SPECTROSCOPY

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ABSTRACT

Our objective was to investigate the possibility to use Fourier transform near-infrared (FT-NIR) spectroscopic technique for determination of deoxynivalenol (DON) and nivalenol (NIV) mycotoxins in spring barley caryopses. Both mycotoxins are produced by Fusarium species causing Fusarium head blight. The barley grain samples from inoculated and non-inoculated field trials performed at four locations in the year 2012 were used for analyses. The spectroscopic data were collected using the FT-NIR Nicolet Magna device. All samples were analysed by a reference method of ultra high pressure liquid chromatography coupled to ultra-high resolution mass spectrometry (UHPLC-Orbitrap(®) MS) at the Institute of Chemical Technology in Prague. The statistics of the calibration and of the cross-validation of the FT-NIR data for DON and NIV showed that the best predictive ability was obtained using the first derivation of spectra. The correlation coefficient of calibration and cross-validation models for DON reached 0.875 and 0.513 and for NIV 0.828 and 0.744, respectively. Due to the very low content of mycotoxins in samples the calibrations are applicable only for detection of highly contaminated grain lots.

Key words: barley, mycotoxins, deoxynivalenol, nivalenol, FT NIR

Acknowledgments: This research was supported by the Internal Grant Agency No. TP4/2013 and by the National Agency for Agricultural Research No. QI 111B044.

INTRODUCTION

The occurrence of *Fusarium* mycotoxins in cereal-based foods and feeds is a global issue of high concern due to their potential health risks for human and livestock (Václavíková et al., 2013). Maximum levels for the main representatives of this group are legislatively laid down (EU, 2006, 2007). Apart from analytical methods based on liquid chromatography separation and mass spectrometric detection other alternative approaches are studied. Near-infrared spectroscopy is based on vibrations of the atoms held together by bonds exposed to infrared light and measurement of reflectance or transmittance of this light. NIRS is a rapid non-destructive method suitable for agriculture products working with small amount of a sample. Our objective was to verify the possibility to use NIR spectroscopy for determination of mycotoxins in barley caryopses. We focused on two mycotoxins – DON and NIV from trichothecene B group.

MATERIAL AND METHODS

The samples of barley caryopses were obtained from field trials performed at four localities (Žabčice, Kroměříž, Senice and Libčany) under four regimes of fungicide treatment and with and without *Fusarium* sp. inoculation of plots. Ten barley varieties were included, i.e. Aksamit, Bojos, Malz, Radegast, Gladys, Tocada, Kangoo, Prestige, Xanadu and Sebastian. The content of DON and NIV was analysed in all grain samples by a reference method of ultra high pressure liquid chromatography coupled to ultra-high resolution mass spectrometry (UHPLC-Orbitrap(®) MS) at the Department of Food Analysis and Nutrition, Faculty of Food and Biochemical Technology, Institute of Chemical Technology, Prague.

For FT-NIR analysis the samples were milled in a mill with a 1 mm sieve. The spectra were collected in a compressive cuvette on the integrating sphere of FT-NIR Nicolet Magna device. The number of scans was 64, resolution 8 and the measurement was performed in the reflectance mode of the wavelength of 12 000–4 000 cm^{-1}. The partial least squares (PLS) method was used for calibration based on the first derivation of average spectrum from three measurements of each sample.

RESULT AND DISCUSSION

The calibration models of DON and NIV were created from almost 200 spectra of samples contaminated by mycotoxins by means of the PLS algorithm. The indicator of error of the PLS calibration method is the predicted residual error sum of squares (PRESS). The optimal number of PLS factors is associated with minimal PRESS value. A high number of PLS factors impairs the predictive capability because PRESS also includes the spectral noise. PLS factors are arranged according to the variation quantity they represent. The first factor describes the highest variation of the calibration standards. Each subsequent factor represents the majority of the remaining variation. Nevertheless, the first factor contains the majority of the common information occurred in the data. The remaining factors describe more specific information representing small changes in data, which is often important for the analysis. If the trend of the PRESS function is falling sharply, this gives an evidence of considerable robustness of the calibration model (Šustová et al, 2007).

The calibration model for NIV was created with the value of the correlation coefficient of calibration 0.8284 using 4 factors (Fig. 1, 2). The calibration was tested using the same set of samples by the cross validation method. The value of the correlation coefficient of validation is 0.7435. RMSEC (root mean square error of calibration) is $0.310e^3$, RMSECV (root mean square error of cross-validation) is $0.371e^3$ and RMSEP (root mean square error of prediction) is $0.433e^3$.

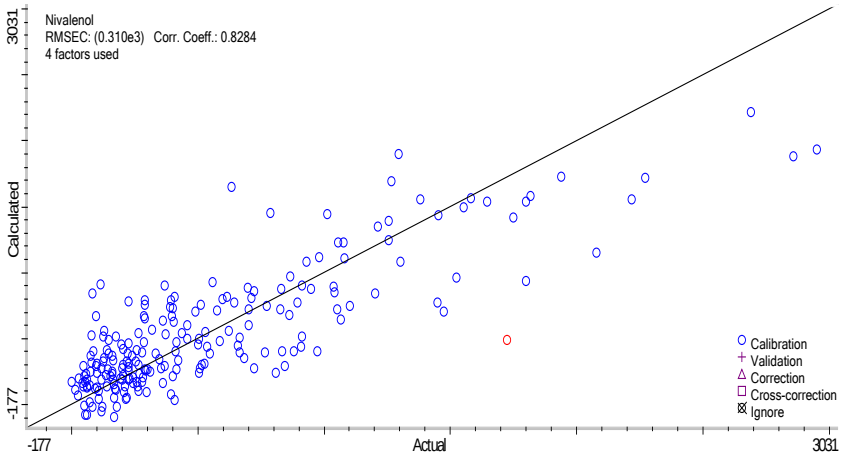

Fig. 1: Predictive PLS model for NIV – linear regression plot of measured (actual) and estimated (calculated) concentrations (μg.kg^{-1})

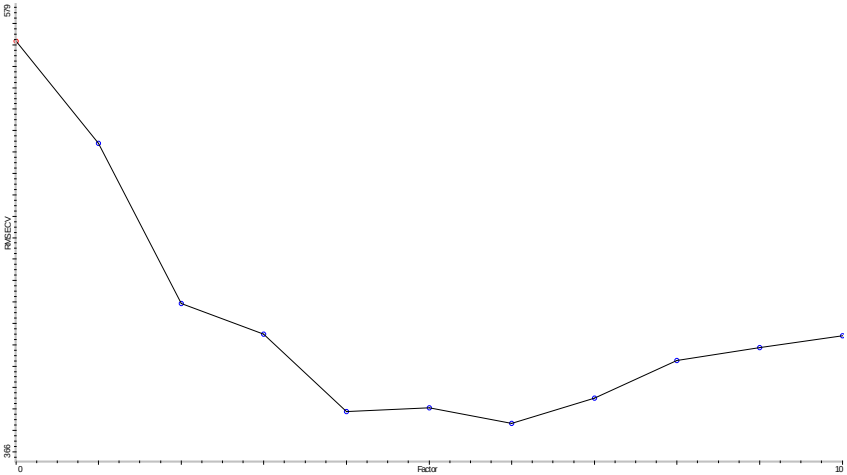

Fig. 2: The PRESS function for NIV (4 factors used)

The calibration model for DON was created with the value of the correlation coefficient of calibration 0.8751 using 6 factors (Fig. 3, 4). The calibration was tested using the same set of samples by the cross validation method. The value of the correlation coefficient of validation is 0.5131. RMSEC is 0.147e^4, RMSECV is 0,268e^4 and RMSEP 0,399e^4.

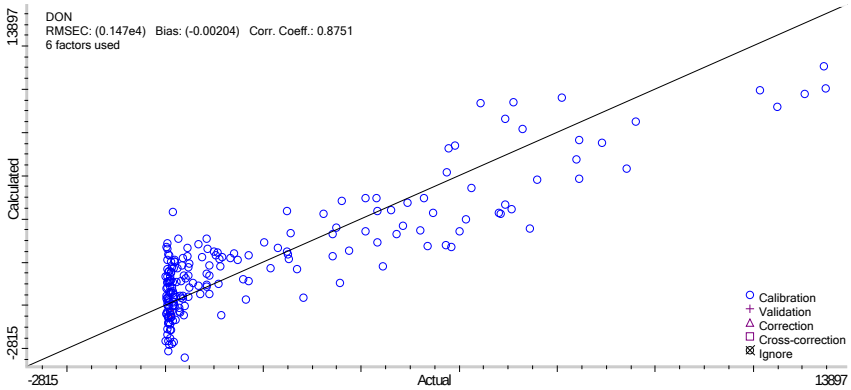

Fig. 3: Predictive PLS model for DON – linear regression plot of measured (actual) and estimated (calculated) concentrations (μg.kg^{-1})

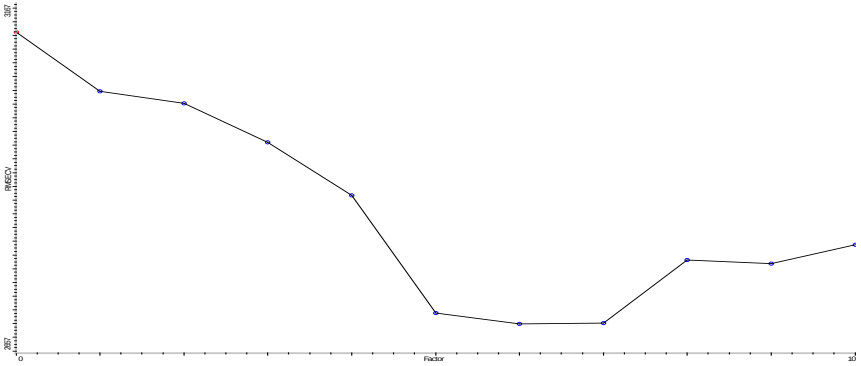

Fig. 4: The PRESS function for DON (6 factors used)

The contents of both mycotoxins in samples were very low from undetectable concentration to about 10 000 μg.kg^{-1}. The NIR spectroscopy method is applicable mainly for measurement of basic components in samples, as protein, moisture, starch and oil. Nevertheless the possibility to employ NIR spectroscopy for detection of low concentration substances is also targeted. Mlček et al. (2013) created calibrations for amino acids in cheese with a concentration about 300 nmol.g^{-1}. Gaspardo et al. (2012) determined fumonisins B1 and B2 in corn meal with a concentration about 4 mg.kg^{-1}. Tripathi and Mishra (2009) developed a method for quantification of aflatoxin B1 in red chilli powder in the range of 15–500 μg.kg^{-1}, which could be used for bulk sorting of the chilli samples from the infected ones before going for any other chemical procedures.

CONCLUSIONS

The applicability of FT-NIR spectroscopy for determination of DON and NIV mycotoxins in spring barley caryopses was tested. The calibration models for both mycotoxins were created and can be used for a prediction of highly contaminated samples. Although the accuracy of the FT-NIR technique is lower than that of the reference method, the results suggested that it can be applied for monitoring of mycotoxins in barley grain, particularly for a rapid screening of contaminated grain.

REFERENCES

EU. (2006). *Commission Regulation (EC) No 1881/2006 of 19 December, 2006* setting maximum levels for certain contaminants in foodstuffs. Official Journal of European Union, L364, 5–24.

EU. (2007). *Commission Regulation (EC) No 1126/2007 of 28 September 2007* amending Regulation (EC) No 1881/2006 setting maximum levels for certain contaminants in foodstuffs as regards Fusarium toxins in maize and maize products. Official Journal of European Union, L255, 14–17.

GASPARDO, B., DEL ZOTTO, S., TORELLI, E., CIVIDINO, S.R., FIRRAO, G., DELLA RICCIA, G., STEFANON, B., 2012: *A rapid method for detection of fumonisins B1 and B2 in corn meal using Fourier transform near infrared (FT-NIR) spectroscopy implemented with integrating sphere*, Food Chemistry 135(2012), 1608-1612

MLČEK, J., ŠUSTOVÁ,K., ROP,O., JURÍKOVÁ, T., HUMPOLÍČEK, P., BALLA, Š., 2013: *Rapid assessment of selected free amino acids during Edam cheese ripening by near infrared spectroscopy*, Acta Vet. Brno 2013, 82: 191-196 doi:10.2754/avb201382020191

ŠUSTOVÁ, K., RŮŽIČKOVÁ, J., KUCHTÍK, J., 2007*: Application of FT near spectroscopy for determination of true protein and casein in milk* , Czech J. Anim. Sci., 52, 2007 (9): 284–291

TRIPATHI, S., MISHRA, H.N., 2009: *A rapid FT-NIR method for estimation of aflatoxin B1 in red chili powder,* Food Control 20 (2009) 840–846

VÁCLAVÍKOVÁ, M., MALACHOVÁ, A., VEPRIKOVÁ, Z., DŽUMAN, Z., ZACHARIÁŠOVÁ, M., 2013: *'Emerging' mycotoxins in cereals processing chains: Changes of enniatins during beer and bread making.* Food Chemistry 136 (2013) 750–757