Determination of Chemical Composition of Maize by Destructive and Non-Destructive Method

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Abstract. Using NIRS technique is an elegant and very precise, this technique become more frequently used for determining physico-chemical properties of feed, especially chemical composition (dry matter, protein, ash, fiber, fat, non-nitrogenous extractive substances. The contents of dry matter, crude protein, crude ash, crude fiber, crude fat, non – nitrogenous extractive substances of maize are determined by destructive methods and then using them was build the calibration curve for NIRS with which we will determine all the chemical properties of the material using prediction.

This paper aims to highlight a way of direct analysis method undestructive of crude protein, crude fat, crude fiber, crude ash, non-nitrogenous extractive substances using near infrared spectrometry technique in conjunction with reflected attenuated total. For each property we have used these method assessment: for the crude protein was used Kjeldahl method, for crude fat was used Soxhlet method, for crude ash performed Hennenberg – Stohmann method, for crude fiber was used Hennenberg - Stohmann method and for non – nitrogenous extractive substances was used mathematical calculation. All these results determined by the classical method, the destructive method, were used to build the calibration curve for device FT-NIR Spectrometer Perkin Elmer Spectrum 100 N with accessory N NIRA. Mathematical model was built using the near infrared spectrofotoscopiei technique in conjunction with multivariable calibration techniques using the Perkin Elmer program Spectrum Quant + 4.60. The method provides a rapid and reliable alternative to traditional quantitative methods for determining which usually requires several hours to complete.

Keywords: NIRS, protein, fat, ash, fiber, non – nitrogenous extractive substances, maize.

INTRODUCTION

Commercial grains are commonly analysed by using NIR spectroscopy. The major constituents of grains are water, protein, oil, fibre, minerals and carbohydrates and it is commercially important to quantities the composition. The NIR spectra of such materials are, thus, dominated by the overtones and combination bands of C-H, N-H, O-H and C=O bonds.

Traditionally, dispersive instruments, available since the 1940s, were used to obtain infrared spectra. In recent decades, a very different method of obtaining an infrared spectrum has superseded the dispersive instrument. Fourier-transform infrared spectrometers are now predominantly used and have improved the acquisition of infrared spectra dramatically. Infrared spectroscopic techniques in combination with chemo-metrics enable the analysis of raw materials without time-consuming sample preparation methods. Fourier transform infrared (FTIR) spectroscopy has been shown to be a promising tool for the analysis of specific sugars, casein and urea. The use of Fourier transform technology in the NIR region

has increased spectral reproducibility and wave number precision in comparison to results from other instruments.

A spectrum may, or may not, contain information related to the sample chemistry measured using any specific reference method. Spectra-structure correlation provides a basis for the establishment of a known cause and effect relationship between instrument response and reference (analyte) data, in order to provide a more scientific basis for multivariate-based near infrared spectroscopy. When performing multivariate calibrations, analytically valid calibration models requires a relationship between X (the instrument response data or spectrum), and Y (the reference data). The use of probability alone tells us only if X and Y 'appear' to be related. If no cause-effect relationship exists between spectra-structure correlation and reference values the model will have no true predictive importance. Thus, knowledge of cause and effect creates a basis for scientific decision-making.

MATERIALS AND METHODS

This study was conducted at the University of Agricultural Sciences and Veterinary Medicine, located on Cluj Napoca in 2008-2009, at ICAR Laboratory and at Grassland and Forage Crops Laboratory for destructive method: the Kjeldhal method for determinating the cude protein, the Henneberg-Stohmann method for determinating the crude ash, Soxhlet method for determinating the crude fat and Henneberg-Stohmann method for determinating the crude fiber and the matematical calculation for non – nitrogenous extractive substances, and then the samples were collected with NIR spectrum at Grassland and Forage Crops Laboratory to build a calibration model for nondestructive method.

Samples of maize cob and maize strains were obtained from The Research – Agricultural Development Turda from during the period from 2005. The samples from 2005 were used solely for calibration. The samples from 2005 were all from The Research – Agricultural Development Turda and were randomly split up into a calibration set and a validation set. NIR measurements were carried out using a FT-NIR spectrometer (PerkinElmer Spectrum One, PerkinElmer) with an NIRA detector. The samples were directly measured, i.e. through the bottom of the intact glass vials by diffuse reflectance without any extra preparation.

All spectra were recorded on a PerkinElmer FT-NIR Spectrometer Spectrum 100N fitted with a „plug-and-play" sampling system accessory for reflectance measurement (NIRA). In the same time each sample was measured using the Kjeldhal method for determinating the cude protein, the Henneberg-Stohmann method for determinating the crude ash, Soxhlet method for determinating the crude fat and Henneberg-Stohmann method for determinating the crude fiber and the matematical calculation for non – nitrogenous extractive substances. Then using these values for spectra we build a mathematical model for direct determination of these two chemical properties of the samples. For this calculation Spectrum Quant+ v4.60 is used.

RESULTS AND DISCUSSION

Forty-six different samples of maize cob and maize strains were supplied and measured with no additional milling or grinding (no processing of spectra). Spectra were recorded by filling a standard sample cup with samples and scanning in interleaved mode. This mode of operation alternately takes a background spectrum as well as the rationed spectrum, which minimizes changes in atmospheric effects.

Eight replicate measurements of each of the calibration samples were collected, and the mean spectrum used for the generation of the calibration equations. Data was collected over the range 10000 to 4000 cm^{-1} at 8 cm^{-1} resolution with 2 cm^{-1} step, and then data was collected over the whole range of the NIR spectrum since this data set may be used to determine a number of other properties in maize cob and maize strains from these spectra.

A partial least squares analysis (PLS) was performed on the data (46 spectra). It is possible to predict values for protein and nitrogen content in sample in the independent validation set. Various mathematical pretreatments were tested and a second derivative function chosen to provide Standard Error of Prediction (SEP) value of 0.29 for protein and 0.71 for crude fat, 0.34 for crude fiber, 0.22 for crude ash, 0.17 for non – nitrogenous extractive substances, using 18 PLS factors and full cross validation. Full cross validation excludes each standard in turn from the calibration set, performs the calibration and then predicts the excluded standard using that calibration. Smaller prediction errors may be obtained using a larger number of PLS factors.

However, it was decided to optimize the calibration for robustness which is better achieved by performing independent validation over time.

Fig. 1, 2, 3 and 4 are the illustrated plots of Estimated versus Specified values, first for crude protein, second for crude fiber third crude fat and fourth for crude ash. This provides an adequate starting point for the calibration model.
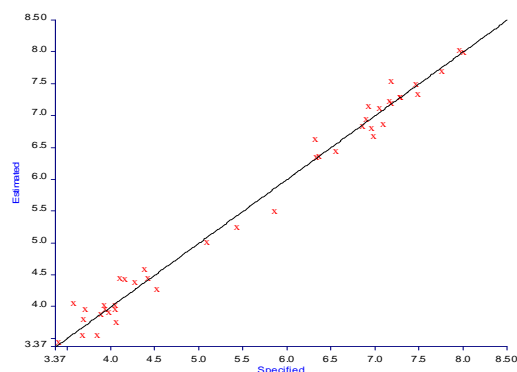

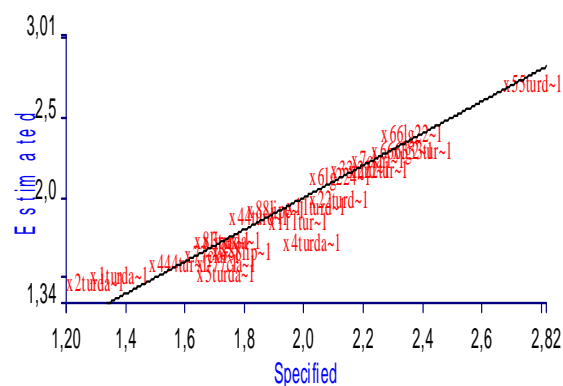Fig. 1. Estimated vs. Specified
plot for crude protein

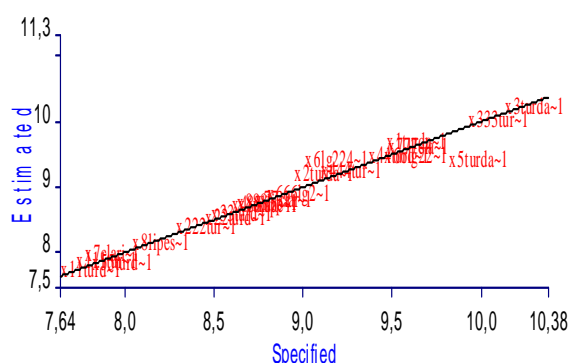Fig. 2. Estimated vs. Specified
plot for crude fiber

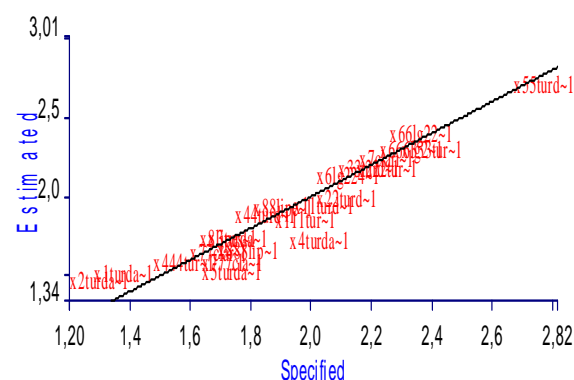Fig.3. Estimated vs. Specified
plot for crude fat

Fig. 4. Estimated vs. Specified
plot for crude ash

The regression model summaries for the full cross validation model are shown in Tab. 1.

Tab.1

Regression model summaries

```
Method Name:      MAIZE
Ident:      Spectrum QUANT+ v4.60

     No. of properties:   4
     No. of standards:   46
     Calibrated:           Yes

     Calculation Parameters:
     Algorithm:          PCR+
Range:            10000 to 4000 cm-1
     Interval:           2 cm-1

Analysis Type:      Absorbance
     Scaling (Spectra):   None
     Scaling (Property):  Mean
     Smooth:             Yes
     Baseline correction:  None
     Order:              2
     Normalization:     None
     Ordinate threshold:
     Upper threshold:   1.5 A
     Lower threshold:    None
     Number of factors:
     Minimum:           1
     Maximum:           100
     Blank regions:     None
```

To support validation, a series of samples were run a week later and both the protein and total nitrogen content predicted using the calibrated model. Table 2 shows the results along with the reference values supplied. Additional statistics in terms of the total M-distance and residual ratio give an indication of how well the model covers these samples.

Tab. 2

Spectrum Quant+ v4.60 PREDICTION RESULTS

Sample	T41	LIP1	CL1
Normalization	None	None	None
RMS Error	2.152 e-004 A	1.690 e-004 A	2.077 e-004 A
Peak to Peak Error	1.267 e-003 A	1.274 e-003 A	1.479 e-003 A
Total M-Distance	0.2603	0.4817	0.3813
Residual Ratio	1.3450	0.8301	1.2540
CRUDE PROTEIN			
Calculated Value	4.9680 %	6.3160 %	7.9520 %
Reference Value	5.0546 %	6.3293 %	7.9673 %
R-error	0.5188	0.4822	0.5775
M-Distance	0.5191	0.2861	0.9291
CRUDE FAT			
Calculated Value	9.5900 %	8.0520 %	7.8560 %
Reference Value	9.4700 %	8.0400 %	7.7800 %
R-error	0.2105	0.2602	0.2637
M-Distance	0.5004	0.5381	0.7088
CRUDE FIBER			

Calculated Value	1.4540 %	1.0100 %	2.1850 %
Reference Value	1.2800 %	1.6300 %	2.1600 %
R-error	0.2609	0.2039	0.2054
M-Distance	0.5744	0.2897	0.3390
CRUDE ASH			
Calculated Value	9.9100 %	9.1330 %	10.4300 %
Reference Value	11.1730 %	8.9394 %	9.9448 %
R-error	7.246e-007	7.152e-007	7.801e-007
M-Distance	0.2344	0.2023	0.4306
NON-NITROGENOUS EXTRACTIVES SUBSTANCES			
Calculated Value	74.0780 %	75.4890 %	71.5770 %
Reference Value	73.0224 %	75.0613 %	72.1479 %
R-error	0.9192	0.9371	0.9271
M-Distance	0.0877	0.1371	0.1095

CONCLUSIONS

The results obtained in this paper work demonstrate that several compositional fractions of forage from different types maize cob and maize strains can be accurately predicted by NIRS on fresh plant material.

The example detailed here illustrates that it is possible to determine a number of properties present in maize cob and maize strains samples with accuracy that is of a similar order to that of the reference method using FT-NIR spectroscopy.

The crude protein content determinate both destructive method and non-destructive method was between 4.23 – 7.91%, the crude fat was between 0.22 – 5.32%, the crude ash was between 8.67 – 11.95%, crude fiber (celluloses) was between 6.01 – 17.92% and the crude non-nitrogenous extractive substances was between 30.72 – 45.67%.

Based on the samples supplied, it has been shown that FT-NIR and partial least squares can be used to determine protein of maize cob and maize strains with very good standard error of prediction (SEP). This proves that FT-NIR spectroscopy is an extremely reliable, non-destructive and rapid technique for the quantity of many chemical and physical properties.

REFERENCES

1. Büning-Pfaue, H. (2003). Analysis of water in food by near infrared spectroscopy. Food Chem. 82, 107–115.

2. Carlier, L. (1994). Breeding, forage quality, feeding value and animal performance. Proc. 19th Fodder crop section meeting Eucarpia. Ed. D. Reheul and A. Ghesquière - RvP Merelbeke.

3. Carlier, L., I. Puia and I. Rotar. (1998). For a better grass production. Ed. Risoprint. Cluj Napoca. p90:149.

4. Carlier, L., I. Rotar, Mariana Vlahova and Roxana Vidican. (2009). Importance and Functions of Grasslands. Notulae Botanicae Horti Agrobotanici. Cluj-Napoca. 37(1): 25-30.

5. Eskandari, H., A. Ghanbari-Bonjar, M. Galavi and M. Salari. (2009). Forage Quality of Cow Pea (*Vigna sinensis*) Intercropped with Corn (*Zea mays*) as Affested by Nutrient Uptake and Light Interception. Notulae Botanicae Horti Agrobotanici. Cluj-Napoca. 37(1): 171-174.

6. Haaland, D. M. and E. V. Thomas. (1988). Partial least-squares methods for spectral analyses: Relation to other quantitative calibration methods and the extraction of qualitative information. J. Anal. Chem. 60: 1193–1202.

7. Hansen, P. W. (1998). Urea determination in milk using Fourier transform infrared spectroscopy and multivariate calibration. Milk Sci. Intr. 53:251–255.

8. Hansen, P. W., A. S. van Brakel, J. Garman, and L. Norgaard. (1999). Detection of specific sugars in dairy process samples using multivariate curve resolution. J. Dairy Sci. 82:1351–1360.

9. Hewavitharana, K. A. and B. van Brakel. (1997). Fourier transform infrared spectrometric method for the rapid determination of casein in raw milk. Analyst 122:701–704.

10. Sørensen, L. K. (2004). Prediction of Fermentation Parameters in Grass and Corn Silage by Near Infrared Spectroscopy. 87:3826-3835.

11. Rotar, I. and L. Carlier. (2005). Cultura Pajiştilor. Editura Risoprint. Cluj-Napoca. p.321.

12. Rotar, I., Roxana Vidican and N. Sima. (2007). Cultura Pajiştilor şi a Plantelor Furajere – ghid practic. Editura Risoprint. Cluj-Napoca. 209-250.

13. Vidican, Roxana, I. Rotar and N. Sima. (2000). Tehnica NIRS (Near Infrared Reflectance Spectroscopy) şi aplicaţiile sale în analiza calităţii furajelor, Simpozion „Agriultura şi alimentaţia". Cluj-Napoca. cod ISSN: 1454-2382, 271-275.

14. Williams, P. C., and K. H. Norris. (1987). Qualitative applications of near-infrared reflectance spectroscopy in Near Infrared Technology in Agricultural and Food Industries. Ed. American Association of Cereal Chemists: St. Paul. USA.

15. ***Spectrum QUANT+ Tutorial. Perkin-Elmer Co. (2006).