

Comparison of commercial near-infrared reflectance spectroscopy (NIRS) calibrations and standard chemical assay procedures for prediction of crude protein levels in poultry feed ingredients

A. KRYEZIU^{1*}, R.I. BAKALLI^{1,2}, M.A. KAMBERI¹, R. KASTRATI¹, N. MESTANI¹

¹Faculty of Agriculture/ Department of Livestock and Veterinary, University of Prishtina, 10000 Kosovë; ²University of Georgia/ Department of Poultry Science, Athens, Georgia 30602-2772, USA.

*Corresponding author: ajkryeziu@yahoo.com

The objective of this research was to determine whether manufacturer supplied near-infrared (NIR) calibrations could produce analyses of crude protein in poultry feed ingredients as accurate as those obtained by Kjeldahl procedure. Twenty samples of each feed ingredient commonly used to prepare poultry feed (wheat, corn, rye, barley, oat, soybean, and sunflower meals) were analyzed by Kjeldahl procedure and by NIRS (NIRS 6500 Feed-Forage Analyzer, using WinISI-III).

Commercial calibration equations were for wheat: mean=14.92, Standard error of calibration (SEC)=0.39, $r^2=0.98$, Standard error of cross validation (SECV)=0.42; corn: mean=9.49, SEC=0.33, $r^2=0.93$, SECV=0.34; rye: mean=15.27, SEC=0.47, $r^2=0.98$, SECV=0.58; barley: mean=12.53, SEC=0.31, $r^2=0.96$, SECV=0.39; oat: mean=13.59, SEC=0.54, $r^2=0.91$, SECV=0.62; soybean meal mean=54.71, SEC=0.59, $r^2=0.92$, SECV=0.61; sunflower meal: mean=36.76, SEC=0.87, $r^2=0.97$, SECV=0.91, respectively.

Protein contents by NIRS and Kjeldahl methods were: mean=11.66 vs. 11.63 ($r^2=0.97$) for wheat, mean=7.29 vs. 6.95 ($r^2=0.78$) for corn, mean=9.34 vs. 9.28 ($r^2=0.83$) for rye, mean=10.80 vs. 10.97 ($r^2=0.79$) for barley, mean=10.54 vs. 10.60 ($r^2=0.92$) for oat, mean=44.00 vs. 43.33 ($r^2=0.98$) for soybean meal, mean=31.80 vs. 31.32 ($r^2=0.91$) for sunflower meal, respectively.

These results indicate that the accuracy of commercial calibration of NIRS could be very acceptable for prediction of crude protein levels to wet chemical methods.

Keywords: protein prediction; NIRS; Kjeldahl; feed ingredients

Introduction

Nutrition is a complex of processes involved to utilize nutrients necessary for proper maintenance, growth, production and reproduction of animals (Pesti et al., 2005). Proteins are of key nutrients required in specific amount and quality to fully support all these activities in poultry production (Pond et al., 1995).

Ensuring qualitative feed and right requirements of poultry, still remains one of main concern for nutritionists (Pesti et al., 2005), and since poultry production is quite much dependent on the level of proteins in a diet, continuous and accurate determination of their content in feedstuffs is an imperative.

Crude protein content of the feed ingredients can be determined by Kjeldahl an old and well known laboratory procedure (Weende procedure adapted by Kirchgeßner, 1987) and by Near Infrared Reflectance Spectroscopy (NIRS) as a new nondestructive procedure. Ben-Gera and Norris (1968) first described the potential on NIRS as a technique for routine quantitative analysis of agricultural products. NIRS has since been developed as a rapid nondestructive method for predicting the chemical

composition of feed and feed ingredients, including moisture, protein, carbohydrates, fats, etc. Conventional laboratory procedures are both time-consuming, costly and environmentally unfriendly. For NIRS procedure, no reagents are involved and no fumes or waste products are generated (Bakalli et al., 2000).

The initial objective of the work reported herein was to compare the results from our Kjeldahl standard laboratory techniques with those from NIRS analyses. We wanted to determine whether NIRS could be used with the standard calibration curves supplied by the manufacturer instead of the more costly and laborious wet chemical procedures without further calibration of the instrument.

Materials and methods

Seven test ingredients with 20 samples each were chosen: wheat, corn, rye, oat, barley, soybean meal and sunflower meal because of its importance as nutrients and protein sources for poultry production. Crude protein (CP) content was analyzed by standard Kjeldahl procedure (Lab) and NIRS (FOSS 6500 apparatus and WinISI II software). Prior to analysis samples were ground in 1 mm sieves. For Kjeldahl method 0.6 grams (for cereals) and 0.4 grams (for oilseed meals) in three replicates were analyzed standard procedures (AOAC, 1990). Nitrogen content is converted to CP by multiplying with 6.25. For NIRS analysis three 5 x 6.5 cm cups were filled and scanned. Commercial calibration equations were for wheat: mean=14.92, Standard error of calibration (SEC)=0.39, $r^2=0.98$, Standard error of cross validation (SECV)=0.42; corn: mean=9.49, SEC=0.33, $r^2=0.93$, SECV=0.34; rye: mean=15.27, SEC=0.47, $r^2=0.98$, SECV=0.58; barley: mean=12.53, SEC=0.31, $r^2=0.96$, SECV=0.39; oat: mean=13.59, SEC=0.54, $r^2=0.91$, SECV=0.62; soybean meal mean=54.71, SEC=0.59, $r^2=0.92$, SECV=0.61; sunflower meal: mean=36.76, SEC=0.87, $r^2=0.97$, SECV=0.91, respectively.

Statistical analysis of data were done by using analyses of variance with the General Linear Models (GLM) procedure of SAS (1994). Correlation coefficients were computed using Proc Corr.

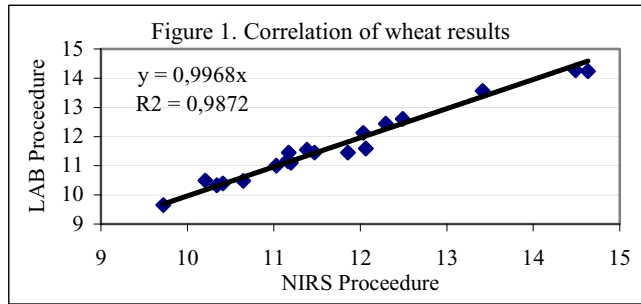
Results and discussion

For all ingredients included in this study, there is no significant effect of method used to predict CP content (Table 1).

Table 1. Descriptive statistics for the protein of 7 ingredient samples as determined by different methods

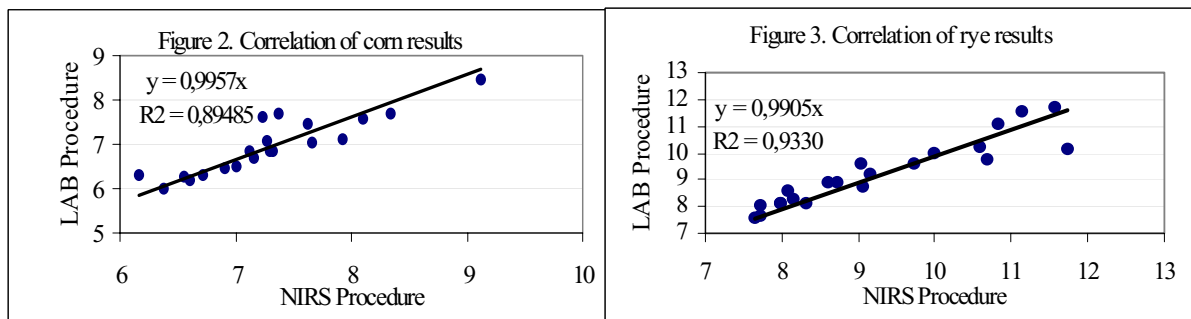
Feeds	Lab Procedure					NIRS Procedure					Pr>F
	Average	Min	Max	Sd	CV%	Average	Min	Max	Sd	CV%	
Wheat	11.63	9.65	14.28	1.26	10.88	11.66	9.72	14.63	1.32	11.30	0.9418
Corn	6.95	6.01	8.47	0.64	9.28	7.29	6.16	9.12	0.70	9.67	0.1158
Rye	9.19	7.56	11.71	1.23	13.38	9.26	7.66	11.73	1.36	14.67	0.8779
Barley	10.97	9.03	12.77	1.05	9.52	10.80	8.57	12.51	1.16	10.74	0.6142
Oat	10.60	8.95	12.41	1.04	9.78	10.54	8.53	12.25	1.02	9.68	0.8343
SBM	43.33	38.16	46.79	2.24	5.17	44.05	39.35	47.43	2.20	5.01	0.3435
SFM	31.32	22.33	35.54	3.49	11.14	31.80	25.93	35.82	2.93	9.23	0.6370

There were no significant differences ($P=0.9418$) in average CP content in wheat between two methods (11.63 vs 11.66 for LAB and NIRS, respectively). NIRS method has slightly higher CV than Lab procedure (0.43%) There was high correlation between the conventional chemical methods and NIRS technique (0.99).



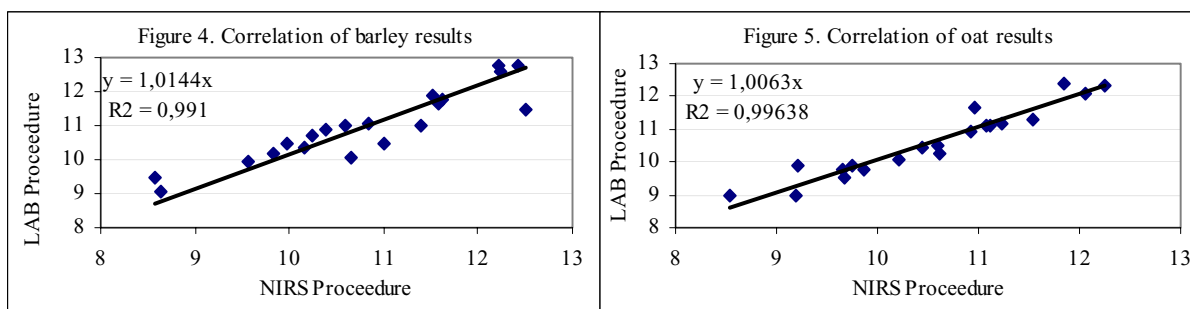
Differences in average protein contents in corn between procedures were higher (6.95 vs 7.29% for LAB and NIRS, respectively), but differences were not significant ($P=0.1158$). Coefficients of variation were similar (9.67 vs 9.28% for NIRS and Lab, respectively). Correlation in prediction of protein contents in corn between analytical procedures were lower but still very strong (0.89).

Content of CP in rye samples analyzed by Kjeldahl and NIRS is very similar (9.19 vs 9.26) and they are not significantly different ($P=8779$) with high correlation between procedures (0.93).



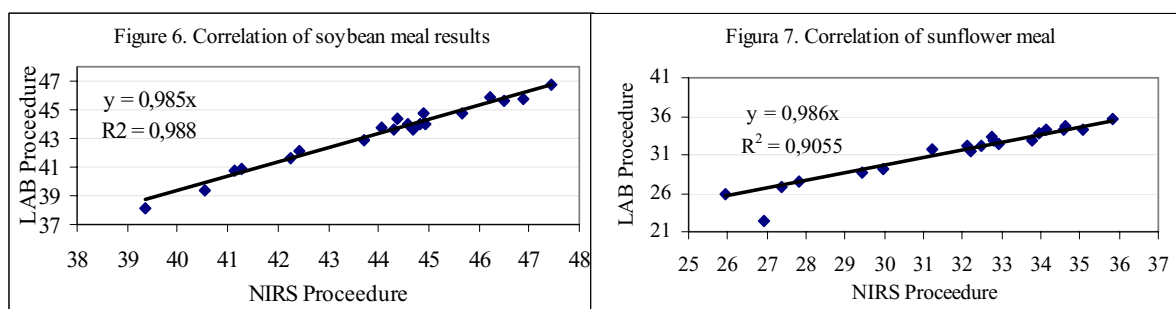
Similar to previous cereals, in barley samples NIRS technique shows some higher values for all parameters measured but average content is not significantly different among used methods ($P=0.6142$) with very high correlation between two analytical procedures ($R=0.99$).

Oat, the last cereal used in this research shows some differences in response of the used method. All parameters are slightly lower, but still not significant ($P=0.8343$) with very high correlation (0.99).



Average CP content in soybean meal analyzed by LAB procedures are slightly lower than NIRS (43.33 vs 44.05% but differences were no significant ($P=0.3435$)). NIRS values showing lower CV comparing with Lab procedure for LAB (5.17 vs 5.01%). Correlation between two procedures is very high (0.99).

Similar to soybean meal, CP of the sunflower meal are lower when analyzed by LAB procedure (31.32 vs 31.80% for LAB and NIRS, respectively). Difference between methods were not significant ($P=0.6370$). Correlation in protein prediction between Kjeldahl methods and NIRS techniques was very high (0.91).



Similar to our results, Hymowitz et al., (1974) reported high correlations between the conventional chemical methods and NIRS techniques for protein from soybeans (ranging from 0.91 to 0.99). Bakalli et al. (2000) that protein values obtained by NIRS were similar to results from standard laboratory procedures with very high correlation (0.99).

From the results of our study it can be concluded that protein values of cereal grains and oilseed meals obtained by NIRS are similar to results from standard laboratory technique with very high correlation. Results indicate that the accuracy of commercial calibration of NIRS could be very acceptable for prediction of crude protein levels to wet chemical methods.

References

- ASSOCIATION OF OFFICIAL ANALYTICAL CHEMISTS, 1990. *Official methods of Analysis* 15th Edition. Assn. Offic. Anal. Chem., Washington, DC.
- BAKALLI, R.I., G.M. PESTI, and R.D. ETHERIDGE, 2000. Comparison a commercial Near-Infrared Reflectance Spectroscopy and standard chemical assay procedures for analyzing feed ingredients: Influence of grinding methods. *J. Appl. Poultry Res.*, 9:204-213.
- BEN-GERA, I., and K.H. NORRIS, 1968. Direct spectrophotometric determination of fat and moisture in meat products. *J. Food Sci.*, 33:64-67.
- HYMOWITZ, T., J.W. DUDLEY, F.I. COLLINS, and C.M. BROWN, 1974. Estimations of Protein and Oil Concentration in Corn, Soybean and Oat Seed by Near-Infrared Light Reflectance. *Crop Science*, 14:713-715
- PESTI, G.M., R.I. BAKALLI, J.P. DRIVER., A. ATENCIO, and E.H. FOSTER, 2005. *Poultry nutrition and Feeding*. Trafford Publishing.
- POND, W.G., D.C. CHURCH, and K.R. POND, 1995. *Basic Animal Nutrition and Feeding*, 4th Edition.
- SAS INSTITUTE, Inc., 1994. SAS User's Guide, Cary, NC.