Premium grains for livestock: an update on NIR calibration development and implementation

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BACKGROUND

One of the major objectives of the national Premium Grains for Livestock Program (PGLP) is to develop rapid methods for measuring the determinants of grain quality relevant to the nutritional requirements of ruminants, pigs and poultry.

This paper provides a brief update on the development of NIR calibrations for a range of chemical, physical, \textit{in vitro} and \textit{in vivo} analyses. The major emphasis in this section of the PGLP from now on will be the implementation of NIR tests at strategic locations within the grain and animal industries. This would allow the test results to be used for pricing grains in accordance with their suitability as an animal feed, and also as a real time decision tool in grain utilisation and diet formulation.

MATERIALS AND METHODS

Approximately 100 different grains were selected over six years, either from breeding programs or acquired commercially. Grain selection was based on a combination of NIR spectra, input from plant breeders and seasonal factors such as rain, drought and frost damage. The objective was to assemble a set of grains having as wide a range as possible in nutritional value. Sufficient quantities of each grain were either grown out or purchased to enable both laboratory analysis and animal feeding trials. The grains in this study included wheat, barley, oats, triticale, sorghum, maize and a small number of pulses (lupins, peas, chickpeas and faba beans).

All grains have been analysed for 76 chemical components, 27 physical properties and 21 functional characteristics specific to ruminants, pigs or poultry.

During the course of the PGLP, grain samples have been scanned on NIR instruments at both Hamilton and Roseworthy, SA (SARDI’s Pig and Poultry Production Institute). Whole grain samples were scanned in a natural product cell using the sample transport module in a Foss-NIRSystems model 6500 scanning monochromator (both locations). Ground samples were scanned in small ring cups using either a spinning sample module in a model 5000 monochromator (Hamilton) or the sample transport module in a model 6500 instrument (Roseworthy). Recently, the instruments at both locations have been standardised to each other. In all cases, ISI software was used.

Preliminary NIR calibrations have been obtained for 52 selected chemical and physical parameters. Two separate sample populations were selected: ruminant (all grains included) and monogastric (omitting the oats and pulses). Within each population, calibrations were developed...
using SNV and Detrend scatter correction, and MPLS regression (1,4,4,1). Calibrations were compared using two different wavelength ranges (408-1092,8 and 1108-2492,8 or 1108-2492,8 only); two sample treatments (whole or milled); and two outlier options (two outlier passes or no outliers omitted).

Separate calibrations for functional characteristics relevant to monogastrics have been developed at Roseworthy. These include whole tract and ileal digestible energy (pigs and poultry) and feed intake, response to enzyme addition and apparent metabolisable energy (poultry). This work is not covered in this paper.

In the case of ruminants, in vivo digestion trials have been conducted at Hamilton since 1997 on 82 different grains using sheep, and of these, 14 have been fed to cattle. An NIR calibration based directly on percentage in vivo dry matter digestibility (DMD) has been progressively updated as new data has been obtained. A calibration for in sacco digestibility of whole oats, based on measurements at NSW Agriculture, Wagga, has also been investigated. Calibrations have also been attempted at the University of New England for a number of in vitro measurements, and additional reference analysis in this field is currently in progress.

RESULTS AND DISCUSSION

The calibrations derived for chemical and physical parameters using the treatments described resulted in a large number of statistics. To give a greatly simplified impression of these preliminary results, the histograms in Figures 1 to 6 compare coefficients of variation (CV) between milled and whole grain for certain groups of measurements. The examples shown are for the “ruminant” population, and in each case the CV values represent the “best” treatment in terms of spectral range and outlier option.
Figure 2. Coefficient of variation plot (CV%) for amino acids

Figure 3. Coefficient of variation plot (CV%) for anti-nutritional factors
Figure 4. Coefficient of variation plot (CV%) for fatty acids

Figure 5. Coefficient of variation plot (CV%) for starch components
Previous studies have suggested that CV values need to be less than 20% for an NIR calibration to be worth considering, and much lower to be used routinely with confidence. Figure 1 indicates that, apart from lignin, calibration performance was satisfactory for proximate constituents using the mixed grain population in this study. NIR was clearly more accurate on milled grain for crude protein, fat and crude fibre, but there appeared to be no benefit in milling grain for the other constituents.

Calibration accuracy appeared to be either high or satisfactory for all amino acids except tryptophan (Figure 2). The benefits of milling versus testing whole grain were not clear-cut, with lower error in milled grain in only 50% of cases.

There appeared little justification for attempting to predict anti-nutritional factors using NIR, with the possible exception of phytic acid and total tannins (Figure 3). Milling did not improve accuracy.

Figure 4 shows that NIR had considerable promise in predicting three of the four fatty acids measured. Stearic acid could not be predicted satisfactorily and milling seemed to decrease error only in the case of oleic acid.

Of the starch components, calibrations for total starch and enzyme digestible starch were accurate, but it was not possible to use NIR for resistant starch (Figure 5). Little if any benefit was obtained from milling the grain.

In the case of physical properties, satisfactory to average calibration accuracy was obtained, using whole grain, for colour variables, specific weight, 100 grain weight and hydration capacity. Based on the grain population and reference method used for hardness index, NIR did not appear to be satisfactory.
Figure 7 shows the most recent results for the direct NIR calibration against in vivo dry matter digestibility (DMD) measured with sheep. This is an attractive alternative to the more usual but indirect method of estimating DMD (and hence metabolisable energy, ME) using chemical, in vitro or enzymic methods then basing an NIR calibration on these measurements. The major limitation of the direct approach is the difficulty and expense in obtaining enough samples with in vivo data for a robust calibration and the ongoing need for validation and testing of the calibration.

The DMD calibration represented in Figure 7 is now based on in vivo DMD values determined on 82 diverse grains over 4 years using a standard protocol (sheep fed at maintenance on a 70:30 mix of rolled grain and chaffed lucerne hay). This is probably the largest number of grains with in vivo DMD values ever produced in Australia. The SECV for this calibration was 1.8, which suggests that NIR could predict in vivo DMD of grain to within 3.6 DMD% units (or approximately 0.5 MJ of ME) for 95% of samples.

This calibration to estimate grain ME is expected to be of considerable benefit to the beef feedlot industry. However, so far it is based on data from sheep fed at a maintenance level. Results to date indicate that, when fed at maintenance on rolled grain, there was no significant difference in DMD between sheep and cattle on the same grain, with the exception of sorghum. However, industry practice in beef feedlots is to feed cattle at ad libitum levels, and it is important to know the difference in DMD for a given grain between sheep fed at maintenance and cattle fed ad libitum.

To investigate this further, feeding trials at Wagga and Hamilton to compare the measurement of DMD on 15 rolled grains (7 wheat, 6 barley and 2 triticale) between sheep (maintenance) and cattle (ad libitum) have just been completed. Data analysis is now in progress to assess the extent to which differences in DMD between sheep and cattle will vary across grain types, and the implications for the NIR calibration.
DMD measurements have also just been completed on 6 additional oat grains using sheep (maintenance). This brings to a total of 103 different grains with measured in vivo DMD values using sheep fed at maintenance. Of these, 3 grains have also been fed to sheep ad libitum, 11 to steers (maintenance) and 18 to steers (ad libitum). This represents a very valuable database.

There is also considerable potential for NIR estimation of in sacco DMD% of whole oats, a valuable screening tool for oat breeders. This measurement involves suspending samples of whole oats in the rumen of a fistulated ruminant for a specified time to estimate dry matter disappearance. An NIR calibration for in sacco DMD% based on 392 samples of whole oats analysed by NSW Agriculture, Wagga, indicated an SECV of 5.0 and a 1-VR of 0.89 for a population having range of 3 to 82% and an SD of 15% (Figure 8).

![Figure 8. Scatterplot of measured vs NIR-predicted in sacco digestibility of whole oats (data from NSW Agriculture, Wagga)](image)

**IMPLEMENTING NIR CALIBRATIONS – THE NEXT STEP**

The NIR calibrations developed to date for the various grain quality parameters have all been derived on scanning monochromators using ISI software. However, a requirement of this project is to implement appropriate NIR tests at the point of delivery of grains (e.g. a feed mill or silo). This requires calibrations to be transferred to NIR instruments at these locations, but these instruments vary in type and performance, and different software packages are in use. It is therefore not possible in all cases to use a spectral standardisation procedure, recognised as the most accurate method of transferring NIR calibrations from one instrument to another, although this technique will be used where practicable, particularly for instruments used in appropriate
GRDC-funded breeding programs. The alternative is to scan a common set of calibration samples on each instrument and then derive calibrations separately for each instrument.

This presents a challenge, as calibration and prediction accuracy for the diverse quality parameters in this study on different instruments is unknown.

An evaluation of several NIR instruments will shortly be undertaken at Hamilton. A selected set of whole grains with reference values will be scanned in sequence on all instruments in the same laboratory, then calibrations will be developed using the NIR scans from each instrument and the calibration statistics compared. Decisions will then be made on the feasibility or otherwise of conducting more rigorous validations on some or all instrument types in the field.

In addition, a range of grain samples from various suppliers will be used to test the applicability of NIR calibrations for some constituents to grains representing different regions, seasons and varieties. A large population of barley samples from a GxE study in a breeding program will also be used to evaluate these calibrations.

A commercialisation strategy, including a cost-benefit analysis, is currently in progress by GRDC for the implementation of relevant NIR calibrations in the feed grain industry.

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