AusScan® Calibration Description

This document is provided to AusScan users as a guide in understanding the AusScan NIR calibrations made available online via the website www.aunir.com

Calibration Development

Over 3,400 grains, primarily wheat, barley, oat, triticale, sorghum and maize, with a wide range in chemical and physical characteristics thought to influence their nutritional value for livestock have been collected from germplasm archives, plant breeders, specifically grown cultivars and farmers. The samples included frost damaged, partially germinated and drought affected grains as well as more normal well-grown and irrigated grains. All grains were scanned with NIR and the extent and rate of digestion of components in selected grains examined with *in vitro* systems simulating rumen fermentation and intestinal digestion. Approximately 504 grains selected on the basis of NIR scans, *in vitro* analyses, genetic background or growing conditions were fed to animals including sheep, cattle, pigs, broiler chickens and laying hens.

Animal feeding data was used with NIR spectra to develop NIR calibrations as described below:

1. RUMINANTS

1.1.1. Cattle Estimated Metabolisable Energy (ME) MJ/kg dry matter

The NIR calibration established for the Cattle ME content of grains reflects the energy availability expected from cereal grains or pulses fed to growing cattle ad libitum. Cattle ME was derived for grain samples based upon rations fed containing 70% dry rolled grain and 30% lucerne hay.
The ME calibration accounts for 88% of the observed variation in ME across all in vivo observations.
The calibration predicts ME with an accuracy of ± 0.26 MJ/kg.
It should be noted that the AusScan ME predictions being based upon ad libitum cattle feeding will provide ME data for grains in the order 0.4 to 0.6 MJ/kg lower than that predicted based upon use of the sheep fed at maintenance ME calibration.
The AusScan ME prediction takes account of the inherent characteristics of the grain and the manner in which the grains constituents are digested. This is in contrast to more traditional predictions based upon wet chemistry equations utilising nutrients measured such as protein, fat and fibre components.

The ME prediction is based upon typical dry rolling of grain and it does not account for more variable grain processing, this either being poor rolling with a presence of whole grains or higher levels of processing such as steam pelleting, steam flaking or extrusion.

1.1.2. Sheep Estimated Metabolisable Energy (ME) MJ/kg dry matter

The NIR calibration established for the Sheep ME content of grains reflects the energy availability expected from cereal grains or pulses fed to mature sheep at maintenance. Sheep ME was derived for grain samples based upon rations fed containing 70% dry rolled grain and 30% lucerne hay. The ME calibration accounts for 88% of the observed variation in ME across all in vivo observations. The calibration predicts ME with an accuracy of ± 0.28 MJ/kg.
The sheep ME calibration is based on feeding at a maintenance energy intake and provides higher ME results for grain than those for cattle fed *ad libitum*. The calibration does not account for heat processing of grains.

1.2. Acidosis ‘hotness’ Index

An index predicting the relative potential for a grain to cause acidosis in ruminants was calculated from the rate of starch disappearance from rolled grains held in a bag within the rumen of cattle for 6 hrs, the starch content of the grain, total acid production during a 5 hr *in vitro* fermentation assay, the lactic acid production during the 5 hr fermentation assay and the buffering effect of the NDF content of the grain through saliva release during mastication. The values obtained for each grain were divided by the highest value to provide an index with potential values from 0 to 100+.
The ‘hotness’ index as calculated from the grains fed to sheep and cattle has a value ranging from 12 for a sample of sorghum to 100 for a naked oat grain sample.

The calibration was used to select 21 grains with predicted ‘hotness’ index ranging from 24 to 89 for evaluation in cattle. The predicted values were highly correlated (P=0.0003) with observed acidosis values measured in cattle. A decision was made to develop a new calibration based on the results from cattle and this is the one that is being made available under the AusScan licence. The calibration accounts for 90% of the variation in calculated Acidosis index and has an accuracy of prediction of ± 6.93 units. The evaluation of the calibration for cattle fed a range of grains varying widely in their capacity to develop acidosis is given in the following paper: Lean, I.J., Golder, H.M., Black, J.L., King, R. and Rabiee, A.R. (2013). In vivo indices for predicting acidosis risk of grains in cattle: comparison with in vitro methods. *Journal of Animal Science* 91:2823-2835.

The calibration is considered useful for distinguishing between grains that have reasonably wide differences in potential 'hotness'. Grains with 'hotness index' values greater than 75-80 could potentially cause acidosis and a reduction in feed intake when fed *ad libitum* to ruminants.

The Acidosis Index prediction is based upon typical dry rolling of grain and it does not account for more variable grain processing, this either being poor rolling with a presence of whole grains or over processing resulting in faster fermentation.

1.3. Whole Oats - Dry Matter Digestibility, Hull Acid Detergent Lignin and Hull Content

The disappearance of dry matter during a 48 hr suspension of whole oats in nylon bags in the rumen of cattle ranged from 3.1 to 85.9% when over 400 oat grain samples were examined. The *in sacco* DMD was shown to be negatively related to the lignin content of the hull, with all grains having more than around 6% hull lignin being poorly digested. There is a strong relationship between whole oat *in sacco* DMD, *in vivo* DMD and animal performance.

The lignin content of hulls from 227 oat grain samples was analysed. The whole grains were scanned before dehulling and a calibration for hull lignin content derived.

The percent hull in oat grains is inversely related to starch content and energy availability. Although 48 hr *in sacco* DMD is a more reliable measure of energy availability, the percentage of hull in oat grains can be estimated with reasonable accuracy using NIR.

The calibration statistics were:
Oat grain *in sacco* digestibility (% dry matter): R^2 0.87; accuracy ± 4.92
ADL in hull (% hull): R^2 0.83; accuracy ± 1.93
Hull content (% whole grain): R^2 0.86; accuracy ± 2.69

Whilst use of these calibrations has direct application for cattle and sheep fed whole oats, there is potential application for oats utilised in horse feeding, with these calibrations having capacity to identify higher energy, lower lignin and lower hull containing oats.

1.4 Faecal Starch, % of DM

Over 300 samples of faeces were collected from individual steers offered feedlot rations containing barley, wheat, triticale and sorghum grains that had been processed to various degrees. The faeces were dried for 24 hours in a laboratory oven at 55-60ºC. The dried material was then ground using a laboratory mill with a 1mm screen. The dried samples were scanned with a NIR and analysed for starch content. A faecal starch calibration was established using faecal samples in which the starch content ranged from 0.01 to 48.7% of dried faeces.

The calibration provides a means of measuring the amount of undigested starch remaining in cattle faeces. It predicts starch in faeces with an R^2 of 0.99 and an accuracy of ± 1%. Values greater than 5% indicate that substantial amounts of starch in the grains are not being digested and processing of the grain could be improved.

PIGS

2.1. Faecal Digestible Energy (FDE) and Ileal Digestible Energy (IDE), MJ/kg as fed

The faecal and ileal DE content of grains including wheat, barley, sorghum, triticale and rice was measured in pigs weighing 35-40 kg and fed diets containing 94% grain and added dicalcium phosphate, salt, minerals and vitamins with a celite marker.

The faecal DE NIR calibration accounts for 89% of the observed variation in faecal DE across all in vivo observations. The calibration predicts DE with an accuracy of ± 0.26 MJ/kg. A difference of 1 MJ/kg is worth from approximately $7.50 to $21.00 per tonne depending on the base cost of the grain (up to $300/t in the simulations) and the relative costs of high and low energy ingredients.

The ileal DE NIR calibration accounts for 83% of the observed variation in faecal DE across all in vivo observations. The calibration predicts DE with an accuracy of ± 0.48 MJ/kg.

2.2. Ileal/Faecal Digestible Energy Ratio

The ileal:faecal DE ratio can be used to predict the proportion of available energy from grains that is digested in the small intestines of pigs. This ratio can be calculated from the individual NIR predictions for Ileal DE and Faecal DE.

2.3. Faecal Digestible Energy Intake Index for pigs, 0-100+

Feed intake was determined for grains including wheat, barley, sorghum and triticale in weaner pigs approximately 7 kg for 21 days. The pigs were fed the same diets used for determining the DE content of the grains which contained 94% grain and added dicalcium, phosphate, salt, minerals and vitamins with a celite marker. Daily feed intake values were multiplied by the faecal DE content of the diet to calculate faecal DE intake. The values obtained for all grains were then divided by the highest value to calculate the faecal DE intake index. Additional information is currently being added to this calibration because it accounts for only 65% of the observed variation and has an accuracy of ± 10.9 units.

The NIR calibrations for faecal DE intake can be used to predict the relative energy intake from the grain and be used as a guide for ranking grains according to their capacity to stimulate growth rate in pigs.

3. POULTRY

3.1. Apparent Metabolisable Energy (AME), MJ/kg as fed

The AME content of grains including wheat, barley, sorghum, triticale, oats and rice was measured in male and female chickens from 22 days of age when fed diets containing 80% grain, 15.5% casein and added calcium, phosphorus, vitamins and DL-methionine. The calibration accounts for 93% of the observed variation in grain AME across all in vivo observations and predicts AME with an accuracy of ± 0.40 MJ/kg.

The NIR calibrations for grain AME can be used to predict the available energy content of grains for broilers.

3.2. Apparent Metabolisable Energy Intake Index, 0-100

The energy value of a grain to broilers depends both on the available energy content of the grain expressed as AME (MJ/kg) and on the amount of the grain eaten. In general, the daily intake of AME will be positively related to the growth rate and performance of birds.

The intake of grain by broilers was also recorded during experiments determining the AME content of grains within PGLP experiments. Thus, the intake of AME (AME intake/bird, MJ/bird/d) could be calculated for the different grains fed to broilers. Because AME intake is not commonly used to rank the energy value of grains, the values were converted to an index by dividing the AME intake value for each grain by the highest value. The values derived were multiplied by 100, such that grains could be ranked with values potentially from 0 to 100+.

The AME intake index values were then used to develop an NIR calibration AME intake index. The calibration accounts for 87% of the observed variation in intake across all in vivo observations predicts AME intake index with an accuracy of ± 4.1.

4. REACTIVE LYSINE

NIR calibrations for rapid prediction of reactive lysine content has been developed for soybean and canola meal. Reactive lysine content of an oilseed meal is a reliable way to estimate lysine available to the animal for methabolism.

4.1 Soybean Meal

The calibration for the total and reactive lysine contents of soybean meal is based on approximately 300 samples of commercially available soybean samples and soybean and isolated soybean protein samples exposed to heat for varying times. The R^2 values relating NIR predicted values to measured values are 0.94 and 0.95, respectively for total lysine and reactive lysine. Similarly the accuracy of the calibrations is ± 1.02 and ± 0.96 (g/kg), for total and reactive lysine. Thus, unknown soybean samples can be predicted with 95% confidence to within 2.04 and 1.92 g/kg of the actual value, respectively for total and reactive lysine. This unique NIR calibration offers a robust prediction for bioavailable lysine content in soybean meal and soy protein concentrate.

In addition, the calibration has the ability to predict apparent, standardized and true ileal digestible total and reactive lysine contents in soybean meal and soy protein concentrate with similar accuracy. The calibration can be applied in commercial feed mills, nutrition labs and SBM trading companies to accurately evaluate the quality of SBM. Furthermore, use of predicted standardized reactive lysine content for diet formulation will improve nitrogen utilisation efficiency by the Australian Pork industry.

4.1 Canola Meal

Commercial crushing plants demonstrate the effect of heat processing on the quality of the canola meal. This variability between commercial crushing plants identifies the value in utilising NIR calibrations as a rapid method of assessing the level of heat damage in canola meal samples. A recent upgrade of this calibration including 126 canola meal and heat treated samples has resulted in a more robust NIR calibrations for prediction of total and reactive lysine content. The inclusion of more samples, as well as heat damaged autoclaved samples, has provided a greater spread of samples, with improved NIR calibration statistics. The current calibrations have R^2 values relating predicted to measured total and reactive lysine of 0.90 and 0.84, respectively for total and reactive lysine. Similarly, the accuracy of the calibrations is ± 0.76 (g/kg) for both total and reactive lysine.