NEAR INFRARED REFLECTANCE SPECTROSCOPY PREDICTS FATTY ACID CONTENT IN BACKFAT FROM PIGS FED REDUCED-OIL CORN DRIED DISTILLERS GRAINS WITH SOLUBLES

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Abstract – The aim of this study was to test the ability of near infrared reflectance spectroscopy (NIRS) to estimate the proportion of major fatty acids (FA) groups in backfat from carcasses of pigs fed reduced-oil corn dried distillers grains with solubles. The outer layer of backfat from 96 pigs was sampled immediately after slaughter, scanned at both 35 and 4 °C over a NIR spectral range from 400 to 2,498 nm using benchtop equipment, and analysed for FA composition. NIRS calibrations, tested by cross-validation, accurately predicted saturated FA (SFA), monounsaturated FA (MUFA), polyunsaturated FA (PUFA), omega-3 and omega-6 FA proportions, in warm samples, with R^2 (RMSECV,% FA) of 0.86 (1.37), 0.82 (1.31), 0.86 (1.08), 0.80 (0.26) and 0.83 (1.03), respectively. The NIRS predictability in cold fat samples was slightly higher for SFA, MUFA, PUFA, omega-3 and omega-6 FA with R^2 ranging from 0.82 to 0.89 and RMSECV from 0.25 to 1.11% FA. The slightly lower NIRS predictability found for the warm samples could be due to increased spectral scattering at a higher a temperature. These results showed that NIRS could be a useful technology for the early, fast and relatively inexpensive estimation of FA groups proportions in backfat from pigs carcasses.

Key Words – Backfat, DDGS, Fatty acid, NIRS, pork

I. INTRODUCTION

For the last 15 years, the North American ethanol industry has increased the availability of corn dried distillers grains with solubles (cDDGS) for livestock feeding. Hence, this alternative feedstuff has been successfully included in swine diets [1].

Feeding cDDGS, however, increases the proportion of dietary unsaturated fatty acids (FA), which can in turn influence carcass fat quality, since carcass fat composition is affected by dietary FA [2]. The increased concentration of unsaturated FA in the pig carcasses results in softer fat, which can lead to processing problems, affect the quality of processed pork products, and influence their ability to meet pork export specifications [3].

In the last 2 years, US ethanol plants have begun to partially remove oil from cDDGS, reducing it from 10–12 to 6–9%. This in turn requires a reassessment of both its net energy (NE) value and effects on carcass quality.

Traditional quantitative chemical techniques for the comprehensive determination of FA profiles involves solvent extraction of total lipids, followed by conversion of FA to their methyl esters and then analysis by GC [4], a time-consuming and costly process. In contrast, near infrared reflectance spectroscopy (NIRS) is a rapid and non-destructive method, neither requiring reagents nor producing waste. The structure of FA can produce individual spectral characteristics and they are, therefore, very accessible for detection and classification by NIRS [5].

This paper examines the potential of NIRS technology to predict the proportion of major FA groups in intact pork backfat using benchtop equipment, and explores the possibility of using

portable equipment for measurements directly from the carcass.

II. MATERIALS AND METHODS

A. Animals and diets

Animals used were a subset of a larger study where the estimated NE value of cDDGS was narrowed down using 1,056 pigs housed in 48 pens, split by gender (barrows or gilts), and fed diets containing cDDGS with assumed NE values of 1.70, 1.85, 2.00, 2.15, 2.30 and 2.45 Mcal/kg over 5 feeding phases. Diets were formulated to provide equal grams of standardized ileal digestible Lys:Mcal NE within phase. Canola oil was added at assumed low NE values of cDDGS and greater inclusions of barley replaced wheat grain as the assumed NE value of cDDGS increased.

B. Slaughter and sample collection

A subset of 96 animals was slaughtered (124.9 kg) at Agriculture and Agri-Food Canada Lacombe Research Centre (Lacombe, Alberta, Canada). Immediately after slaughter, the temperature of the outer layer of backfat was recorded and approximately 200 g of backfat over the grading site (7.5 cm of the mid-line, between the 10-11th rib) were collected and refrigerated at +2 ºC for solidification.

C. Spectra collection

When backfat was solidified enough to core (approximately 1 h), skin was removed and duplicate intact circular fat cores from the outer layer of backfat were obtained using a custom-constructed stainless steel device [6] to enable consolidation of fat and fat discs of an appropriate diameter (38 mm) and thickness (7 mm) to fit the ring cups of the NIRS machine. Each fat disc was placed in a ring cup, all visible air bubbles removed by squeezing, and the cup backed with thin black foam. Subsequently, the samples were placed in open plastic bags and heated in a water bath at 39 °C. A DuaLogR model 600–1050 (Barnant Company Barrington, USA) thermocouple was inserted into the centre of each fat sample for temperature monitoring during warming. As soon as the core sample reached the average temperature of backfat immediately after slaughter (35 °C), samples were removed from the water bath and NIR spectra were collected. Afterwards, fat cores were kept under refrigeration over night and then scanned cold at 4 °C. Each backfat sample was scanned 32 times over the range (400–2,498 nm) using a NIRSystems Versatile Agri Analyzer (SY-3665-II Model 6500, FOSS, Hillerød, Denmark) benchtop equipment and spectra were averaged by the equipment software. Two fat samples per animal were scanned using two different cells. The two spectra were visually examined for consistency and then averaged. The spectrometer interpolated the data to produce measurements in 2 nm steps, resulting in a diffuse reflectance spectrum of 1,050 data points. Absorbance data were stored as log (1/*R*), where *R* was the reflectance. Instrument control and initial spectral manipulation were performed with WinISI II software (v1.04a; Infrasoft International, Port Matilda, MD, USA).

D. Fatty acid analysis

After NIR spectra collection, the two fat cores from each animal were stored at −80 °C for subsequent FA determinations. From the backfat collected, 5 g was sampled and 50 mg subsamples were freeze dried and used for FA analysis by gas chromatography according to Dugan et al. [7].

E. Data analysis

Calibration and validation were performed using The Unscrambler program (version 10.2, Camo, Trondheim, Norway). Two passes of elimination of outliers (H and T) were allowed. Spectral data were subjected to standard normal variate and detrend (SNV-D) and/or first or second-order derivatives (1D/2D) to reduce multicolinearity and the confounding effects of baseline shift and curvature on spectra arising from scattering effects due to physical effects, and to heighten the signals related to the organic compounds [8]. Partial least square regression (PLSR) was used for predicting FA proportions using NIR spectra as independent variables. Internal full cross-validation was performed to avoid over-fitting the PLSR equations. Thus, the optimal number of factors in each equation was determined as the number of factors after which the standard error of cross-validation no longer decreased. The accuracy of prediction was evaluated in terms of coefficient of determination (R^2) and root mean square error of cross-validation (RMSECV).

III. RESULTS AND DISCUSSION

Table 1 summarises the range, mean, standard deviation (SD) and coefficient of variation (CV) of major FA groups analysed in this study. The values found were similar to those indicated by Benz et al. [9] in carcass backfat from pigs fed corn-soybean meal diet with 15% DDGS and choice white grease.

Table 1. Descriptive statistics for fatty acids (% total FA) in backfat samples from pigs (n = 96)

Fatty acid	Range	Mean	SD	CV (%)
SFA	28.0-41.3	34.9	3.16	9.1
MUFA	37.5-48.3	42.8	2.41	5.6
PUFA	16.6-29.4	22.3	2.37	10.6
Omega-3	0.8-3.0	1.6	0.52	31.8
Omega-6	15.7-26.3	20.6	2.10	10.2

SD: standard deviation; CV: coefficient of variation; SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids.

NIRS calibrations, tested by cross-validation, accurately predicted the proportions of major FA groups when spectra were collected on warm samples (Table 2). The best prediction equations were obtained after applying the SNV-D+2D to the spectra and showed R^2 (RMSECV, % FA) of 0.86 (1.37), 0.82 (1.31), 0.86 (1.08), 0.80 (0.26) and 0.83 (1.03) for SFA, MUFA, PUFA, omega-3 and omega-6 FA proportion, respectively.

When NIR spectra were collected on cold backfat samples (Table 2), slightly higher predictability than that reported for warm samples was observed for the proportion of SFA, MUFA, PUFA, omega-3 and omega-6 FA, where R^2 (RMSECV, % FA) were 0.89 (1.11), 0.86 (1.11), 0.89 (0.97), 0.82 (0.25) and 0.86 (0.94), respectively, mostly after applying a 2D to the spectra.

The slightly lower NIRS predictability found for the warm samples in this study could be due to changes in spectral scattering associated with temperature during NIR spectra collection. This suggests that in practical conditions, when NIRS is to be used on-line in the abbatoir, the NIRS predictability would probably be higher when collecting the NIR spectra in chilled carcasses 24 h post mortem than immediately after slaughter on warm carcasses. Nevertheless, given that difference in accuracy between the prediction equations with warm vs. cold backfat samples was small, the application of NIRS when backfat is still warm should be considered due to the advantages of NIRS collection in a more optimum operator and equipment environment, and the subsequent ability to sort carcasses according to fat hardness for marketing purposes as they enter the cooler.

Table 2. Prediction of fatty acid proportion in pigs backfat using NIR spectra collected on warm and cold samples

	p	Treatment	R^2	RMSEC	RMSECV
Warm samples					
SFA	7	SNV-D+2D	0.86	1.15	1.37
MUFA	7	SNV-D+2D	0.82	1.00	1.31
PUFA	6	SNV-D+2D	0.86	0.87	1.08
Omega-3	4	SNV-D+2D	0.80	0.23	0.26
Omega-6	6	SNV-D+2D	0.83	0.84	1.03
Cold samples					
SFA	5	2D	0.89	1.00	1.11
MUFA	7	2D	0.86	0.86	1.11
PUFA	7	2D	0.89	0.78	0.97
Omega-3	5	SNV-D+2D	0.82	0.22	0.25
Omega-6	7	2D	0.86	0.76	0.94

p: number of PLS terms utilized in the calibration equation; R^2: coefficient of determination of calibration; RMSEC: root mean square error of calibration; RMSECV: root mean square error of cross-validation; SNV-D: standard normal variate and detrend; 2D: second-order derivative.

Predictions by Pérez-Juan et al. [10] had lower accuracy than those observed in this study for SFA (R^2 = 0.85, RMSECV = 1.4%) and PUFA proportions (R^2 = 0.77, RMSECV = 1.2%) in ham backfat from carcasses of pigs fed a commercial diet enriched with antioxidants or conjugated linoleic acid. However, they reported a higher NIRS predictability for MUFA percentage (R^2 = 0.92, RMSECV = 1.3%), probably due to a wider range (32.99-48.91%) and higher CV (12%).

The performance of predictions on warm backfat opens possibilities of using portable NIRS equipment to capture spectra directly on the carcass. Such possibilities are intriguing but should consider the challenging operational environment at abattoirs such as fluctuations in temperature and humidity, which can compromise the reliability of NIR spectra.

Our research group has pursued such a path, and preliminary results from using a fibre-optic probe directly on pork backfat on-line show low noise levels in the spectral absorption regions below 2,350 nm when the spectra were collected on both warm (immediately after slaughter) and cold (after 24 post mortem) backfat (Fig. 1). This approach suggests that NIRS might be a promising technology to monitor and control fat quality at initial processing via remote on-line detection.

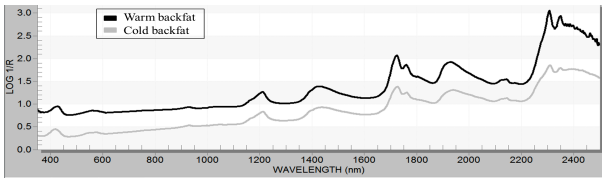

Figure 1. NIR spectra collected on-line on warm and cold backfat from pig carcasses

IV. CONCLUSION

NIRS technology has the potential to quickly and accurately estimate the proportions of the major FA groups in backfat from carcasses of pigs fed reduced-oil cDDGS. Further work remains to be carried out to develop robust NIRS models to be implemented at large scale, fast processing lines (1200 hogs/h), where portable equipment applied directly on the carcass would provide clear advantages in speed of analysis and carcass sorting for marketing.

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