PORK LOIN FATTY ACID CONTENT ESTIMATION FROM PIGS FED REDUCED-OIL CORN DRIED DISTILLERS GRAINS WITH SOLUBLES USING NEAR INFRARED SPECTROSCOPY

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Abstract - This study tested the ability of near infrared reflectance spectroscopy (NIRS) to estimate the proportion of fatty acids (FA) in pork loin from pigs fed reduced-oil corn dried distillers grains with solubles. At 24 h post mortem, a mid-loin chop was collected from 96 carcasses, scanned both intact and homogenized from 400 to 2,498 nm on benchtop equipment, and analysed for FA composition. For intact meat, predictions for SFA and MUFA were the most reliable (R^2 = 0.81 and 0.87; RMSECV = 0.85 and 1.03 mg FA/g muscle), followed for PUFA, CLA and omega-6 FA (R^2 = 0.73, 0.72 and 0.75; RMSECV= 2.28% FA, 0.003 mg FA/g muscle and 1.99% FA), but the predictions for omega-3 FA were unreliable (R^2 = 0.44; RMSECV = 0.24% FA). In homogenized form, higher accuracy was found for SFA, MUFA, PUFA, CLA and omega-6 (R^2 = 0.95, 0.95, 0.84, 0.85 and 0.93; RMSECV = 0.41, 0.50 and 0.002 mg FA/g muscle for SFA, MUFA and CLA, and 1.33 and 1.22% for PUFA and omega-6 FA), omega-3 proportion was still not although accurately predicted (R^2 = 0.60; RMSECV = 0.21% FA). These results showed that NIRS can estimate rapidly and accurately the content of major FA groups in pork loin.

Key Words - DDGS, Fatty acid, NIRS, Pork

I. INTRODUCTION

With the rapid development of the fuel ethanol industry, the production of major co-products of ethanol production such as corn dried distillers grains with solubles (cDDGS) has been increased tremendously [1].

cDDGS is often used as a dietary energy source in pig feed [2]. However, the high proportion of unsaturated fatty acids (FA) in its oil has a detrimental effect on pork quality and shelf life given that unsaturated fats are more vulnerable to oxidation [3]. Lipid oxidation may lead to an increase in off-flavors, odors, and may damage sensitive fat-soluble vitamins.

Recently, the partial removal (4-6%) of oil from cDDGS has resulted in a need to reassess both its dietary net energy (NE) value and effects on carcass and pork quality.

The determination of FA profile in the muscle of swine is a long and tedious task involving extraction of total lipids and determination of FA methyl esters by gas chromatography [4]. Hence, there is an urgent need to find fast and efficient alternative methods to estimate FA composition. Near infrared reflectance spectroscopy (NIRS) has shown to be a rapid and non-destructive method, neither requiring reagents nor producing waste, to estimate accurately the FA composition in beef [5].

The aim of this study was to compare the reliability of NIRS technology for predicting FA composition from intact and homogenized pork loin samples of pigs fed 30% cDDGS.

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, 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 equal grams of standardized ileal digestible Lys:Mcal NE within phase. Canola oil was added at assumed low NE values 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). At 24 h post mortem, a chilled chop from the grading site (7.5 cm of the mid-line, between the 10-11th rib) was collected for scanning over the near infrared region and for FA analyses.

C. Spectra collection

For each chop, duplicate intact circular lean cores were obtained using a custom-constructed stainless steel device [6] to create a lean disc of an appropriate diameter (38 mm) and thickness (7 mm) to fit the ring cups of the NIRS machine. Each disc was placed in a ring cup, all visible air bubbles removed by squeezing, and the cup backed with thin black foam. After NIR spectra collection, the two lean disks and the remainder of each chop were homogenized using a Robot Coupe Blixir BX3 (Robot Coupe USA Inc., Ridgeland MS, USA). Duplicate ring cups were then prepared with premeasured volumes of homogenate and the help of a modified syringe in order to avoid air bubbles, backed with foam, and spectra collected. Every 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. The pair of spectra per animal 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

From each homogenized sample, 50 g were subsampled and frozen until used for FA analysis by gas chromatography according to Mapiye et al. [4].

E. Data analysis

Calibration and validation were performed using Unscrambler program (v10.2, The Camo. Trondheim, Norway). Two passes of elimination of outliers (H and T) were allowed. Spectral data were subjected to standard normal variate and detrend (SNVD) [7] to reduce multicolinearity and the confounding effects of baseline shift and curvature on spectra arising from scattering effects due to physical effects. First or second-order derivatives (1D/2D) were applied to the spectra to heighten the signals related to the organic compounds of the meat samples [8]. Partial least square regression (PLSR) was used for predicting FA proportions using NIR spectra as independent variables. Internal full cross-validation was performed in order 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 crossvalidation 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

The content of the major FA groups in intramuscular fat of pork loin in this study (Table 1) agree with those shown by Wang et al. [9] in pork samples from pigs finished with 30% cDDGS.

Table 1 Descriptive statistics for fatty acids (mg FA/g muscle and % total FA) in pork loin samples (n = 96)

Fatty acid	Range	Mean	SD	CV (%)
mg FA/g muscle				
SFA	2.18-9.40	5.24	1.668	31.83
MUFA	2.20-12.05	5.91	2.050	34.71
PUFA	1.72-3.04	2.29	0.324	14.16
CLA	0.01-0.03	0.01	0.006	37.27
Omega-3	0.08-0.25	0.13	0.038	29.08
Omega-6	1.75-3.76	2.28	0.325	14.24
% total FA				
SFA	31.33-43.70	37.63	2.657	7.06
MUFA	33.27-51.57	42.01	3.594	8.55
PUFA	11.20-26.11	17.43	3.545	20.34
CLA	0.06-0.15	0.11	0.018	17.28
Omega-3	0.49-2.02	1.00	0.333	33.23
Omega-6	11.66-26.35	17.30	3.326	19.22

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

Table 2 shows the statistics corresponding to the best equations for the prediction of each major FA group in both intact and homogenized pork loin samples.

When loin samples were scanned intact, reliable NIRS equations, after applying the 2D to the spectra, were obtained to estimate the content of SFA and MUFA ($R^2 = 0.81$ and 0.87; RMSECV = 0.85 and 1.03 mg FA/g muscle). The NIRS predictability was lower, although still acceptable, for PUFA, CLA and omega-6 FA content with R^2 (RMSECV) of 0.73 (2.28% total FA), 0.72 (0.003 mg FA/g muscle) and 0.75 (1.99% total FA), respectively. In contrast, unreliable equation was found to predict the proportion of omega-3 FA ($R^2 = 0.44$; RMSECV = 0.24% total FA).

Table 2 Prediction of fatty acid content in pork loin using NIR spectra collected on intact and homogenized samples

	р	Trt	\mathbb{R}^2	RMSEC	RMSECV
Intact samples					
SFA (mg FA/g muscle)	7	2D	0.81	0.71	0.85
MUFA (mg FA/g muscle)	7	2D	0.87	0.74	1.03
PUFA (% total FA)	6	2D	0.73	1.82	2.28
CLA (mg FA/g muscle)	5	2D	0.72	0.003	0.003
Omega-3 (% total FA)	6	SNVD +2D	0.44	0.20	0.24
Omega-6 (% total FA)	5	2D	0.75	1.68	1.99
Homogenized samples					
SFA (mg FA/g muscle)	4	1D	0.95	0.36	0.41
MUFA (mg FA/g muscle)	4	2D	0.95	0.45	0.50
PUFA (% total FA)	3	2D	0.84	1.19	1.33
CLA (mg FA/g muscle)	4	2D	0.85	0.002	0.002
Omega-3 (% total FA)	3	2D	0.60	0.18	0.21
Omega-6 (% total FA)	5	2D	0.93	0.82	1.22

p: number of PLS terms utilized in the calibration equation; Trt: treatment; R²: coefficient of determination of calibration; RMSEC: root mean square error of calibration; RMSECV: root mean square error of cross-validation; SNVD: standard normal variate and detrend; 2D: second-order derivative; 1D: first-order derivative.

Prediction equations with higher accuracy were observed for all the major FA groups when homogenized pork samples were scanned. In this regard, the percentage of variance explained by the model was over 84% for SFA, MUFA, PUFA, CLA and omega-6 ($R^2 = 0.95$, 0.95, 0.84, 0.85 and 0.93, respectively). Additionally, the RMSECV was low when compared to the SD of the population (RMSECV = 0.41, 0.50, 0.002 mg FA/g muscle for SFA, MUFA and CLA, respectively; and 1.33 and 1.22% total FA for PUFA and omega-6 FA, respectively). Although the NIRS predictability for omega-3 in homogenized samples was higher than in intact lean samples, it was not high enough to consider the equation as acceptable ($R^2 = 0.60$; RMSECV = 0.21% total FA).

Despite the differences in accuracy between intact and homogenized samples, it should be noted that SFA, MUFA and CLA were better predicted in both intact and homogenized samples when expressed in absolute concentrations (mg FA/g muscle). In contrast, PUFA, omega-3 and omega-6 FA were better estimated when expressed on a percentage basis (% total FA). This discrepancy could be due to differences in the range of variation and CV of the FA reference data, the data range being stretched and CV being higher when those FA were expressed as percentage of total FA (Table 1).

The results obtained for both intact and homogenized samples in the current study were better than those indicated by Gonzalez et al. [10] to estimate the proportion of SFA ($R^2 = 0.65$; RMSECV = 1.76% total FA) in intramuscular fat when a fibre-optic probe was applied to intact pork loin samples. For MUFA and PUFA, although the NIRS predictability found in this study when using homogenized samples was much better than that reported by those authors ($R^2 = 0.89$ and 0.74; RMSECV = 1.29 and 1.25%, respectively), similar results were obtained when NIR spectra were collected on intact samples.

Sample preparation has an important effect on the reliability of the NIRS prediction [11]. Therefore, the lower NIRS predictability found in this study for the intact samples could be due to a lack of homogeneity. Moreover, homogenization disrupts the structure of the muscle, destroying and randomizing the fibre arrangement of the muscle and, therefore, averaging the effects of scattering by fibres.

However, despite the slight loss in accuracy, the ability of NIRS technology to predict the content of the major FA groups from intact loin samples is still sufficiently high to be of interest from an industry point of view, because it would have the evident advantage of speed of analysis and provide opportunities for pork product selection and differentiation.

IV. CONCLUSION

The content of major FA groups in pork loin from pigs fed reduced-oil cDDGS were rapidly and accurately estimated from NIR spectra collected on homogenized samples. Lower accuracy was found for FA groups in intact samples, but NIRS predictions were still reliable for SFA and MUFA and acceptable for PUFA, CLA and omega-6 FA content. Thus, these benchtop results showed that NIRS is a promising technology that could be used to grade pork loin. Further research is required to develop robust NIRS models to implement at an industrial scale for rapid pork selection and differentiation.

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