VALIDATION OF RAMAN SPECTROSCOPY TO VERIFY PREMIUM AUSTRALIAN BEEF CARCASES

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I. INTRODUCTION

Due to the increased content of health beneficial fatty acids of Australian grass -fed beef and increased eating quality of long-term grain-fed beef both attract a premium at retail. Verification of grass-fed carcases is currently achieved through individual supply chains which not only represents a significant cost to the industry, but also risks market access should a failure occur. Consequently, Raman spectroscopy has been utilised as a rapid method for verifying feeding systems given it is capable of real time analysis of chemical composition [1]. Yet the models created have not been validated on independent data which is required to ensure the robustness of models and the repeatability of predictions required for industry application. Therefore, the aim of this study was to validate models previously created used to verify feeding systems for Australian grass-fed beef.

II. MATERIALS AND METHODS

Calibration models were created by measuring the subcutaneous fat on the brisket from carcases of long- and short-term grain fed cattle as well as grass- and grass-fed cattle in northern and southern Australia 3 times using a 785nm Mira Raman device (Metrohm®). Spectra collected were reduced to 600 – 2000cm^{-1} before they were pre-processed and partial least squares discrimination analysis was undertaken [2]. Subsequent spectra for model validation were collected from 100-day grain fed (278) and grass fed carcases (350) and pre-processed as per calibration model. The PLS-DA calibration model was then applied to these samples not known to the model to classify them as “grass” or “grain”. Like the calibration models, the validation models were cross validated using random subsets and permutation testing.

III. RESULTS AND DISCUSSION

Overall, validation model results indicated the calibration model was accurate as 82.2% of grass-fed carcases and 90.3% of grain-fed carcases were correctly classified and precise given the precision value (P) of 0.85 and 0.87 for grain-fed and grass-fed carcases respectively (Table 1). The high precision demonstrates the benefit of the model as falsely classifying cattle fed with grain as grass fed poses the greatest risk to market access.

Table 1. Predictive model accuracy and performance metrics for the validation of Raman spectroscopy to classify carcases based on production system of origin.

Class:	TPR	FPR	TNR	FNR	N	Err (%)	P	F1
Grain	0.903	0.177	0.823	0.097	278	14.2	0.802	0.849
Grass	0.822	0.097	0.902	0.177	350	14.1	0.914	0.866

Abbreviations: true positive rate (TPR); false positive rate (FPR); true negative rate (TNR); false negative rate (FNR); number of test samples (N); class error (Err), Precision (P), F1, accuracy of model.

The performance of validation models is only marginally lower than the calibration models which reported accuracies of 88% for grass fed and 96% for grain fed carcases indicating the calibration models are robust over time [2]. However, this may be further increased by validating models for northern and southern cattle separately, given they accounted for regional differences in feed and therefore fatty acid composition [2].
The VIP Score plot (Fig 1) show spectral features at 1050 -1150 cm^{-1}, 1300 -1380 cm^{-1}, 1450 - 1500 cm^{-1} and 1650 - 1700 cm^{-1} underpin this classification. This demonstrates the model is based on signals including the C-C stretch at 1060 – 1140cm^{-1}, the CH_2 twisting bond at 1305 cm^{-1} and 1309 cm^{-1} as well as the scissoring CH_2 and CH_3 bonds which are characterised at 1450 cm^{-1} [3]. With further spectral differences at 1650 - 1700 cm^{-1} identifed as the C=C bond of unsaturated fatty acids, it is evident these models are derived from differences in fatty acid composition related to the finishing diet which are being detected by the spectra [3]. However, further research is required to quantify these differences.

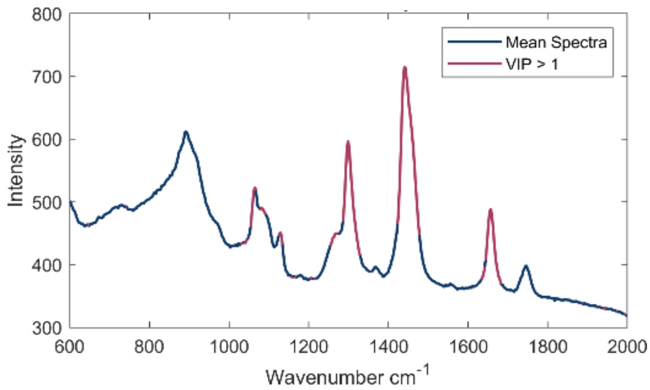

Figure 1. VIP Score plot for the validation of Raman spectroscopy to classify carcases based on feeding system.

IV. CONCLUSION

Overall, calibration models created to verify Australian grass and grain fed beef carcases are repeatable and robust as shown by the high accuracy and precision of the validation model with temporal stability. However, the performance of the model may be increased by completing separate models for carcases from northern and southern production regions. Key spectral features have been identified including 1050 -1150 cm^{-1}, 1300 -1380 cm^{-1}, 1450 - 1500 cm^{-1} and 1650 - 1700 cm^{-1} although further research is required to determine how these relate to the fatty acid composition.

ACKNOWLEDGEMENTS

The authors would like to thank Meat and Livestock Australia and the NSW Department of Primary Industries for funding this research as well as Matt Kerr (NSW DPI), Richard Meyers (NSW DPI), Kerrie Graham (NSW DPI) and Bridgette Logan (NSW DPI) for their assistance with sample collection and analysis.

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