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Technical Report

Program and KPI: Sub-program 2.2 Summary Report

Report Title: Develop and validate new eating quality (EQ) technologies

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Contents

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Executive Summary

This report summarises the key scientific deliverables that was achieved, which was to develop and validate new eating quality (EQ) technologies in beef, lamb and pork. This included cut surface, probe and non-invasive technologies. As a result, several devices have progressed towards commercial prototype phases and installation in-plant to facilitate on-line calibration and validation work.

- SOMA NIR device
- Optical Coherence tomography probe
- Optical coherence elastography
- Nuclear Magnetic Resonance
- ASD NIR device
- Dual energy X-Ray absorptiometry (DEXA)

In addition, the role of fatty acids and distribution of marbling on eating quality in beef was also investigated.

1.1 SOMA NIR device

The experimental work was designed to develop and validate the SOMA NIR device for predicting IMF% in lamb. The Soma S-7090 device uses NIR spectroscopy using the wavelength range of 885 to 1015nm. The S-7090 is portable, non-destructive and is a relatively fast NIR instrument powered by rechargeable batteries. The SOMA had been previously calibrated using partial least squares regression using chemical IMF% data and scans (n = 1318) collected in 2020 and 2021 (R^2 = 0.77, RMSEC = 0.75).

Validation predictions from the SOMA NIR device demonstrated that it could predict lamb chemical IMF% with moderate precision (R^2 = 0.46 – 0.61, RMSEP = 0.56 – 0.74) and accuracy (Bias = 0.02 – 0.86), however differences in prediction existed between experimental sub-groups. In addition, repeatability of the SOMA NIR device was high up to 4 hours post quartering, indicating some flexibility in scan time may be possible in a commercial setting. Further work is required to understand the effect of aging and environmental factors on NIR measurement and prediction of IMF% in lamb.

This validation project was successful, and the device progressed to commercialisation phase and AUS-MEAT accreditation (as was reported in sub-program 2.1). Full details of the experimental work are provided in the appendix.

- Report on a plan for the validation of the SOMA NIR device in lamb
- Report on the updated calibration and validation of the SOMA NIR device for IMF% in lamb
- Report on the independent validation and repeatability of the SOMA device

1.2 Nuclear Magnetic Resonance (NMR)

The objective of this project was to design and build a beta Marbl™ prototype system for non-destructive and automated intramuscular fat (IMF) measurement of lamb and to test it in a processor's facility. In prior work, a prototype single-sided NMR sensor (alpha Marbl™) that was specifically designed for the application, was shown to successfully measure

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intramuscular fat non-invasively and non-destructively. For a full description of the experimental work, the final report is included in the appendix

- Report on the independent validation of Nuclear Magnetic Resonance prototype device for the prediction of IMF, pH and WBSF in lamb).

On-line validation testing of approximately 1000 lambs of a range phenotypes at JBS Bordertown was conducted in 2022 and showed that the NMR was able to meet AUS-MEAT accreditation on a narrow range of IMF% (2-6%). Further samples will need to be collected outside this range for a more thorough test data set. Currently iNMR have been focusing on developing and commercialising the device for fast, accurate measurement of IMF%as this is their primary business goal. The measurement of pH and shear force will be trained and tested at a future date. Future work will involve the use of two new in-line systems, commissioned at JBS Bordertown which will enable the device(s) to operate at abattoir line speed.

1.3 Optical Coherence Tomography

The aim of this study was to demonstrate the feasibility of using OCT to measure the IMF% of intact hot carcases and to examine whether using deep learning algorithms could improve accuracy of IMF% predictions.

Optical coherence tomography (OCT) is a technology commonly used in human medicine to produce high resolution cross-sectional images of the microstructure of tissue. OCT is analogous to ultrasound technology, however near infrared light (NIR) is used instead of soundwaves. The OCT imaging probe developed by Miniprobes is miniaturised and encased within a hypodermic needle, enabling imaging deep within muscle tissue.

Early calibration work demonstrated excellent potential of the OCT device to predict IMF%, with a with a mean absolute error of 0.87%, an RMSE of 1.00% and R^2 of 0.62. The outcomes of this preliminary study were intended to endorse/decline progression to a prototype of the device specifically designed for use in a meat processing plant and more extensive calibration and validation trials. Full detail of the experimental work is included in the following reports in the appendix:

- Report on a plan for the calibration and validation of the commercial OCT prototype
- Report on the initial calibration performance of Optical Coherence Tomography predicting chemical IMF% lamb

This project has progressed, and significant development work has been undertaken in image analysis, the design of the scanner handpiece and the back-end OCT scanner. With support from the MLA and Dept. Industry CRC-P funding, Miniprobes are planning to have a proof-of-principle system ready for installation in a commercial plant by mid-2023. This first system will be used to assess robustness of the system and scan data reliability. This is a critical early step in development of a robust commercial system to predict IMF% in lamb.

1.4 Optical coherence elastography

This was a small project which investigated whether objective coherence elastography (OCE) technology could predict lamb tenderness. Optical coherence elastography (OCE) is a high-

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resolution imaging technique of generating 3D maps of tissue mechanical properties, such as stiffness and surface stress. OCE has long been used in biomedical imaging for its high imaging resolution and rapid acquisition. Full details of the experiment are included in the appendix in the following report:

- Report on measuring lamb meat stiffness using optical coherence elastography

Briefly, meat samples were acquired from 20 lambs which were aged for 1 and 5 days. The OCE device was able to collect high-resolution images with fine muscle structure presented. The device demonstrated a difference in stiffness values (kPa) between day 1 and 5 aged lamb, with overall stiffness of the sample on Day 1 is higher than that on Day 5 (kPa). This aligns with the tenderness results, which showed that Warner Bratzler Shear Force (WBSF) was also (tougher) on day 1. These results support that OCE has the potential to measure the difference in mechanical properties of the meat. However further work was not undertaken to validate or commercialise this technology.

1.5 ASD NIR device

The aim of the current study was to determine the potential for Near Infrared (NIR) spectroscopy to predict IMF using Partial Least Squares (PLS) and machine learning analysis methods. Full detail of this experimental work is presented in the appendix in the following report:

- Report on PLS and Machine Learning for the ASD NIR prediction of IMF in lamb

The ASD NIR fibre optic device was used to scan 299 lamb loins and a sample was excised for Soxhlet determination of IMF content. Prediction models were created using either PLS or machine learning analyses methods. IMF prediction model outcomes were similar between analysis methods with an R^2 = 0.6 and RMSE = 0.84 and R^2 = 0.65 and RMSE = 0.72, respectively. This study highlighted that spectra from one slaughter varied greatly from the two succeeding slaughters and wavenumbers selected between studies are not consistent. Further work on the ASD device did not progress as the company did not develop a commercialisation plan for developing and supporting a device to measure IMF% in lamb.

1.6 Live animal Ultrasound

This work investigated quantitative ultrasound techniques to estimate the intramuscular fat content of NZ and Australian Lamb loins and beef striploins. Full detail of this work is presented in the appendix in the following report:

- KPI 3.26.3 Ultrasonic assessment of intramuscular fat percentage in beef and lamb loins at 37ºC

Ultrasound is an extensively studied technology for objective measurements in a range of different applications. The driving motivation behind recent improvements in ultrasonic image reconstruction for soft tissues has come from the medical industry. However, few of these improvements have translated into meat quality assessment. This study demonstrated that new algorithms and imaging protocols could be implemented on existing ultrasound hardware, providing the industry with improved decision support tools for grading live animals. In order to validate these findings in a commercial setting a device should be developed which can be deployed on-farm or in-plant.

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1.7 MasterBeef to predict IMF% in lamb

The MasterBeef cut surface camera utilises a Samsung phone mounted to a 3d printed shroud, designed to grade Beef Carcasses. It is handheld and can be used by a single operator predicting a range of traits with intramuscular fat % the single one utilised for this data set. This experimental work has been described in the following report presented in the appendix:

- Report on the ongoing validation and repeatability of the MasterBeef system for measuring IMF% in lamb

In this work, Masterbeef camera images and IMF% samples were collected from 50 commercial lamb loins. Camera predictions of IMF% were compared to reference values using the AMILSC accuracy standards for measuring IMF% in lamb. Accuracy performance did not meet the required standard. Further work was required to improve the AI models and be re-trained to determine the smaller loin face compared to the rib eye of beef. Significant hardware changes would also need to be made to facilitate optimal image acquisition. As MasterBeef were primarily focused on predicting traits in beef, work in this area was finalised.

1.8 Effect of fatty acids on eating quality in beef

This study aimed to evaluate the potential effect of fatty acid content on eating quality when carcases were selected in a case-control fashion based on marbling. Three cohorts of 36 carcases were selected from Angus, Wagyu Angus F1 cross, purebred Wagyu, and Wagyu *Bos indicus* F1 cross, all of which had been long fed (≥200 DOF). The chuck roll, bolar blade, striploin, D-rump, and outside flat were consumer tested using the grill cook method. Sensory scores for CMQ4 were analysed within cohorts against cut and carcase traits for Australian consumers. The results showed that cut, IMF%, rib fat and all the fatty acids except linoleic acid have a significant effect on CMQ4. The inclusion of muscle explains 47% of variation in the model and IMF% explains a further 12% of variation. The inclusion of oleic and palmitic fatty acids explains a further 11% and 12% of variation in the model, whereas palmitoleic, myristic and stearic explain only a further 2%, 4% and 5% of the model and linolenic explains no significant variation in the model. Inclusion of monounsaturated (MUFA) and polyunsaturated (PUFA) had significant effect on CMQ4, whereas, saturated fatty acids (SFA) was not significant when included in the model.

1.9 The association of DEXA images with lamb eating quality.

This project analysed data collected at 2 commercial abattoirs (sites) that have installed dual energy x-ray absorptiometry (DEXA) systems used to drive carcass cutting devices in plant and concurrently predict carcass composition of fat, lean and bone. Carcasses were subsequently used in eating quality experiments, enabling the relationship between DEXA and eating quality to be explored. An algorithm has recently been established which better identify bone pixels within the DEXA images allowing better determination of all carcass bone DEXA R values. The all carcass bone DEXA R values and those from individual bones that were manually isolated from DEXA images (humerus, lumbar vertebra and femur) were used to predict eating quality from cuts across the lamb carcass. Data from both sites was analysed independently due to the inability to calibrate DEXA images between the two sites,

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however future data acquired will utilise phantoms to allow bone DEXA values to be compared. An increase in all carcass and some individual bone DEXA R Mean terms demonstrated an association with decreasing eating quality (overall liking, tenderness, juiciness and flavour). The best prediction was of the loin grill where a decrease of 10.5 and 9 overall liking scores was observed across the range of all carcass bone DEXA R Mean at site 1 and site 2. The other cut with relatively strong associations with bone DEXA was the shoulder roast, however the prediction of eating quality in other cuts was more tenuous and lacked consistency between sites. This experiment also used whole carcass lean % and loin intramuscular fat % to investigate relationships with eating quality. In this experiment the bone DEXA R Mean terms were generally independent predictors of eating quality to those of loin IMF % and carcass lean %. The biology underpinning the relationship between DEXA and eating quality has not been identified, though is likely associated with an index of maturity. Bone mineral content did not directly relate to eating quality, however there were some relationships of bone minerals (magnesium, calcium and phosphorus) with bone DEXA R and carcass composition. The isolation of bone pixels from the carcass during routine DEXA scanning at abattoirs may be able to provide input into a multi-trait eating quality model in the future, however future research is necessary.

1.10 Calibration and initial validation of devices predicting IMF% and other traits in pork

Intramuscular fat (IMF) is related to eating quality in both beef and sheep, and probably in pork. However, the amount of IMF in pork is much lower than in the red meat species which presents problems in its' measurement. Near infra-red (NIR) and nuclear magnetic resonance (NMR) technologies have been shown to be able to predict IMF in lamb and in beef. However, the relationship between IMF and these technologies in pork is unknown. The objectives of this project was to determine the chemical IMF content of two pork muscles (Longissimus thoracis et lumborum (LTL) and Semimembranosus (SM)) and relate these values of measures obtained using NIR (with a SOMA device) and NMR in 60 pork carcasses.

The chemical IMF % of the LTL (1.04±0.051%) and SM (1.38±0.065%) were very low compared to other red meat but within the expected range. The IMF content of the SM was higher ($p<0.001$) than the LTL. All measures of SOMA LTL IMF were highly correlated ($p<0.001$) with chemical IMF%. Mean SOMA LTL IMF, geometric mean SOMA LTL IMF and the highest SOMA IMF accounted for 36.4%, 31.9% and 36.9% of the variation in chemical IMF %. There was a significant correlation ($p=0.008$) between chemical IMF % and NMR average p2f, although only 9% of the variation was accounted for. Interestingly, measures of SOMA IMF% were correlated with NMR p2f, indicating that they were measuring something similar.

All measures of SOMA SM IMF were correlated ($p<0.05$) with chemical IMF% but not to as great an extent as the LTL, although the correlations were much lower than for the LTL. Mean SOMA, geometric mean SOMA and highest SOMA IMF accounted for 6.7%, 6.4% and 6.4%, respectively, of the variation in chemical SM IMF %.

When muscles were combined, there were highly significant correlations ($p<0.001$) between SOMA measures of IMF and chemical measures of IMF, and these relationships were

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slightly improved by including muscle in the model. For example, the inclusion of muscle in the model relating mean SOMA to IMF described 27.7% of the variation compared to 25.5% in the simple model. Similarly, for the other relationships relating SOMA measures of IMF to chemical IMF in the pooled data set.

While these values are unlikely to be good enough to provide confidence in predicting IMF within the low ranges of IMF encountered in Australian pork LTL and SM, they do provide encouragement that with some finessing of the instrumentation and algorithms, which were specifically developed for lamb, an online tool can be developed.

The major conclusion from this project is that both the SOMA and NMR technology appear to be related to pork IMF, particularly in the LTL. These relationships exist despite the very low levels of observed IMF %. It is recommended that pork carcasses be manipulated nutritionally and genetically to increase the range in IMF to further test both SOMA and NMR over a greater range in IMF %.

2 Conclusion

Program 2 was successful in achieving its outcomes for 2.2 which were to develop and validate new eating quality (EQ) technologies in beef, lamb and pork. Several devices have been developed and advanced to commercial prototype stages, with on-site installations undergoing calibration and validation work. Further support will be required post ALMTech to assist and support adoption and on-line integration of these devices across the supply chain.

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