

Copyright is owned by the Author of the thesis. Permission is given for a copy to be downloaded by an individual for the purpose of research and private study only. The thesis may not be reproduced elsewhere without the permission of the Author.

# In-plant, non-invasive spectral imaging for the prediction of lamb meat quality attributes

A thesis presented to Massey University  
for the partial fulfilment of the requirements of the degree of  
Masters of Food Technology

Massey University, Manawatū, New Zealand

Adam Douglas Stuart

2016



# CERTIFICATE OF REGULATORY COMPLIANCE

This is to certify that the research carried out in the Masterate Thesis entitled:

Thesis title: *In-plant, non-invasive spectral imaging for the prediction of lamb meat quality attributes*

in the academic unit of:

Academic unit: *Food Technology*

at Massey University, New Zealand,

- (a) is the original work of the candidate, except as indicated by appropriate attribution in the text and/or in the acknowledgments
- (b) that the text, excluding appendices/annexes, is within the recommended 40,000 word count
- (c) all the ethical requirements applicable to this study have been complied with as required by Massey University and other organisations and/or committees

Insert name(s) of the external organisation(s)/committee(s) if applicable:

*Beef and Lamb Genetics NZ  
Alliance Group Limited  
AgResearch Limited*

which had a particular association with this study, and relevant legislation.

Ethical Authorisation code(s) if applicable:

Student ID number: 

1	5	2	1	3	5	6	6
---	---	---	---	---	---	---	---

Student name: *Adam Douglas Stuart*

Students signature: *Adam Stuart*

Chief supervisor's name:

*Ally Thompson*

Signature: *Ally Thompson*



**COPYRIGHT FORM AND DECLARATION CONFIRMING CONTENT OF DIGITAL VERSION OF THESIS**

Student ID number: 

1	5	2	1	3	5	6	6
---	---	---	---	---	---	---	---

Student's name: Adam Douglas Stuart

Thesis title: In-plant, non-invasive spectral imaging for the prediction of lamb meat quality attributes

Have you published articles/material from your thesis?  Yes  No

If 'Yes', have you received copyright permission from the third party to include this published material in your thesis which will be placed in the Library's electronic repository?  Yes  No

Signed on: 

Day	Month	Year
2,7	0,4	2,0,1,6

Student's signature:

**COMPLETE THIS SECTION ONLY IF YOU OPTED TO SELF-PRINT YOUR THESIS**

I confirm that the content of the digital version of this thesis is the final amended version following the examination process, and is identical to the bound paper copy.

Signed on: 

Day	Month	Year

Student's signature: \_\_\_\_\_



## Acknowledgements

Firstly I would like to thank the Beef + Lamb New Zealand Genetics project, funded by Beef +Lamb NZ and the Ministry of Business, Innovation and Employment, for not only making this project possible but whose scholarship allowed me to further my studies in such a new and exciting area and expand my understanding of the world of science, industry and food technology as a whole. Namely I would like to thank Eleanor Linscott and Graham Alder. The contribution of Mike Tate from Mike Tate Consultancy was instrumental in securing the project and funding.

Alliance Group Limited whose supply of staff and processing facilities were invaluable for sample collection and processing. Gemma Milne, Sonja Lindsay and Johanna Phillips became not only work colleagues but good friends with wise words and an amazing working knowledge of all things lamb and production related. A special thank you should be reserved for Gary Maclennan for accepting me into his home, always making me feel welcome and being there whenever I had need of him.

To my supervisors Abby Thompson, Marlon Reis and Tricia Johnson, I would like to offer many thanks for working through the many dramas and issues that followed in my wake and offering guidance where needed and a firm but positive prod in the right direction when it looked like I was missing my way.

I would also like to thank AgResearch for allowing me the opportunity to take time away from work to focus on this thesis and also to the many staff that supported this work with information, skills and time in my support. Cameron Craigie for fighting the good fight and navigating the maze that is bureaucracy, while the hands on skills and support of Kevin Taukiri, Kathryn McRae, Michelle Challies, Paul Shorten, Robert Wieliczko and Michael Agnew when my two were not enough was invaluable.

I would also like to offer a thank you to the great, unnamed many hiding behind the scenes from a multitude of companies and places that provided the little bits and pieces that are always needed when any great endeavour is made.

## Abstract

Muscle foods such as meat are a perishable, nutritious, relatively expensive food commodity, a great source of human nutrition and are a large part of the New Zealand economy, as well as overseas. Currently, New Zealand's meat producing companies measure meat quality attributes by using a different technology for every trait, with no overarching way to combine them, with many of the technologies requiring collection and destruction of the product. There is a desire by the meat industry to find a single way to measure and compare meat quality parameters in a single process or technology. The development of an in-line (within the normal production line of an abattoir or meat processor), real time, non-destructive quality control system could help define multiple meat traits in a way that can guarantee the product in terms of composition, safety and consistency. These guarantees not only help the producer to ask a higher premium for their product, but also give assurances to the consumer that they are getting exactly what they are expecting and paying for.

This thesis focussed on determining whether the spectral imaging technologies of near infrared and hyperspectral imaging, and relevant pre-processing and modelling techniques were suitable for use in an in-plant situation for the prediction of lamb meat quality attributes.

Data was collected on 2511 lambs from 10 separate kills. The lambs were slaughtered through three abattoirs owned by Alliance Group Limited with near infrared and hyperspectral imaging of intact *M. Longissimus thoracis et lumborum* muscle surface collected at 24 hours *post-mortem*. Traditional meat quality measurements were also collected; tenderness using a MIRINZ tenderometer, CIELab colour using a CR-400 colour meter, ultimate pH using an Eutech Cyberscan pH 300 meter, marbling using subjective scoring by trained personnel and intramuscular fat content using gas chromatography – flame ionisation detector. The resulting data were split and used to generate calibration and validation data sets. The calibration data was used together with the spectral data that was processed using a variety of chemometric techniques including partial least squares, variable selection and neural networks to generate predictive models. The accuracy of the predictive models was then tested using the validation data set.

This work found that not all meat quality traits were able to be predicted accurately and certain techniques worked better for differing traits. The best predictive models for ultimate pH using the near infrared and hyperspectral data achieved  $R^2$  values (a measure of goodness of fit) from the validation data sets of 0.63 and 0.48 respectively. For near infrared the best predictive models were achieved using partial least squares with pre-processing (standard normal variate, orthogonal signal correction and mean centring) applied, while for hyperspectral imaging neural networks provided the best model using a decay of 0.00004 and a node size of 2. The best predictive models for intramuscular fat using the near infrared and hyperspectral data achieved  $R^2$  values from the validation data sets of 0.56 and 0.75 respectively. For near infrared this was achieved using partial least squares with pre-processing (normalisation, multiplicative scatter correction and mean centring) applied, while for hyperspectral imaging neural networks provided the best model using a decay of 0.0009 and a node size of 4. This performance of these two traits in particular, shows that that the prediction abilities are of a quality that future work on implementing these into an in-line system at a pilot scale should be considered.

Overall, the use of novel modelling techniques such as neural networks showed potential to increase the predictive abilities of the resulting models, over more traditional modelling techniques. Additionally, it was demonstrated that the number of predictors needed to create a calibration model could be reduced, increasing the speed of analysis with only minimal loss in the accuracy of the resulting model.

Results obtained during this study suggest that the calibration models are not abattoir dependent and the transfer of one calibration model to multiple abattoirs could decrease the costs and allow for faster development and implementation of an in-line, in-plant system.

## Table of Contents

Acknowledgements.....	4
Abstract.....	5
Figures, tables and equations .....	10
1 Introduction .....	13
Adding value through quality: The need for monitoring quality in the meat industry .....	13
Monitoring quality: the challenges in the meat industry .....	13
1.1 Meat Quality: a background .....	17
Meat Quality Measurements used by Industry .....	17
1.1.1 Pre and during production .....	17
1.1.2 Post production.....	19
pH.....	19
Meat colour.....	25
Meat tenderness.....	30
Intramuscular fat.....	32
Sensory evaluation by panel .....	36
1.2 Spectroscopic based sensors .....	38
1.2.1 Spectroscopy in the Visible and Near Infrared spectral range .....	39
1.2.2 Hyperspectral imaging .....	42
Line scanning or pushbroom imaging.....	45
1.2.3 Univariate and Multivariate model types .....	45
1.2.4 Current spectral imaging considerations .....	46
1.3 Modelling .....	48
1.3.1 Principal Component Analysis (PCA).....	48
1.3.2 Partial Least Squares (PLS) .....	49
1.3.3 Support Vector Machine (SVM) .....	49
1.3.4 Artificial Neural Networks (ANN) .....	50
1.3.5 Pre-processing.....	52
Multiplicative Scatter Correction (MSC) .....	52
Standard Normal Variate (SNV) .....	53
1.3.6 Selection of model parameters and validation.....	53
1.3.7 Expressions of model fitness/robustness .....	55



Scope.....	57
2 Materials and methods .....	58
2.1 Sample Acquisition.....	58
2.2 Sample collection and preparation .....	59
Loin Collection.....	61
Loin Processing.....	63
2.3 Meat quality measurements.....	65
2.3.1 Ultimate pH.....	65
2.3.2 Tenderness.....	65
2.3.3 Colour stability .....	65
2.3.4 Marbling.....	67
2.3.5 Intramuscular fat.....	67
2.4 Spectral acquisition.....	72
2.4.1 VIS-NIRS.....	72
2.4.2 HSI .....	74
2.5 Data analysis .....	75
2.5.1 Software .....	76
2.5.2 Spectra selection.....	76
2.5.3 Calibration, Validation .....	77
2.5.4 Pre-processing.....	77
2.5.5 Variable selection.....	78
2.5.6 Neural Network.....	79
2.5.7 Testing predictive abilities .....	79
3 Results.....	81
3.1 Physical meat quality characteristics .....	81
3.2 Modelling .....	86
3.2.1 VIS-NIRS.....	86
3.2.2 HSI .....	94
3.2.3 VIS-NIRS and HSI .....	105
4 Discussion.....	107
4.1 Prediction of intramuscular fat .....	108
4.1.1 VIS-NIRS.....	108
4.1.2 Exploratory analysis .....	111
4.2 Prediction of marbling .....	113

4.3 Prediction of objective tenderness; shear force.....	115
4.4 Prediction of ultimate pH.....	117
4.5 Prediction of colour.....	120
5 Conclusion.....	123
6 Recommendations.....	125
6.1 Industry implementation.....	125
6.2 Future research.....	126
Sample population variation.....	126
Intramuscular fat.....	126
Tenderness and eating quality.....	127
Between year differences.....	127
Between abattoir differences.....	127
Colour.....	127
Whole loin variation.....	128
Appendix one: meat quality data summary.....	129
Appendix two: pre-processing and PLS as applied to near infra-red spectral data.....	132
Bibliography.....	147

## Figures, tables and equations

Figure 1: Representation of the complex relationship between intrinsic and extrinsic factors involved in defining what is meat quality, specifically beef in this example. GHG = greenhouse gas. Intrinsic quality includes physical (colour, shape, appearance, tenderness, juiciness, flavour) and nutritional. Extrinsic quality comes from factors affecting the product from external sources <sup>9</sup> .....	15
Figure 2: Current New Zealand abattoir grading system for lamb carcass quality assessment <sup>12</sup> .....	19
Figure 3: Example of common household items and their corresponding pH levels and corresponding H <sup>+</sup> /OH <sup>-</sup> concentration <sup>14</sup> .....	20
Figure 4: Simplified diagram explaining the Krebs or citric acid cycle showings its major products and by-products .....	22
Figure 5: The colour of beef meat at various pH levels <sup>19</sup> .....	23
Figure 6: A graph showing the shear force (objective value of tenderness measured by the force needed to break through muscle fibres) values in kgf of low, intermediate and high pH <sub>u</sub> lamb meat after 1 day of ageing. Taken from Watanabe, et al. <sup>25</sup> . See meat tenderness section for further explanation. ....	24
Figure 7: A typical electronic pH meter as in use in the majority of food industry and research situations (Thermo Fisher Scientific, Massachusetts, USA) .....	25
Figure 8: Diagrammatical representation of the haem myoglobin (By Mrbean427 [CC BY-SA 3.0 via Wikimedia Commons).....	26
Figure 9: The different chemical states of myoglobin, the colour of the meat is regulated by the oxidative state of iron in the haem group (Mancini et al., 2005) .....	28
Figure 10: Illustration of DMB, OMB and MMB formation in the surface layer of a cut of meat by increasing oxygen exposure (Photo courtesy of Dr. D.H. Kropf, Kansas State University).....	29
Figure 11: A Photo of a cooked lamb loin sample cut perpendicular to fibre direction in a 1 x 1 cm sub-sample ready for tenderness testing .....	32
Figure 12: Example of gas chromatography results showing peaks correlating to different fatty acids. The total fat content can be found by addition of all the peaks .....	35
Figure 13: Simple bond combinations which give rise to VIS-NIRS absorbance <sup>61</sup> .....	40
Figure 14: Incident light entering the meat structure and being scattered by its inherent structure creating errors in the responding absorption/reflectance measurements (By GianniG46 [CC BY-SA 3.0] via Wikimedia Commons) .....	41
Figure 15: Example of a Near Infrared profile exhibiting spectrum scatter before and after correction using various types of pre-processing to eliminate additive and multiplicative scatter creating a tighter, and more uniform band of spectral information <sup>73</sup> . ....	42
Figure 16: A three dimensional representation of a data cube with x and y representing the spatial elements of a sample and λ representing the spectral dimension. The spectrum shown is for an individual pixel. ....	43
Figure 17: A spatial image of a meat sample on the left with the highlighted pixels showing their corresponding spectrum on the right. Meat in blue, plastic label in yellow, paper tape in black, background in green, intermuscular fat in red, intramuscular fat in pink.....	43
Figure 18: (a) Configuration of a typical hyperspectral imaging system (b) Schematic of line scan hyperspectral imaging system used to collect hyperspectral optical scattering images: 1) camera; 2) spectrograph; 3) Field Of View of line scan camera offset 5 mm from centre of incident beam of light;	

4) steak sample; 5) automated vertical stage; 6) linear slide, moved the sample under the line scan in the direction of travel; 7) incident fibre optic cable; 8) light source tungsten halogen lamp; 9) photoelectric switch. ....	44
Figure 19: Graphical representation of hypercube data collection along a moving conveyor belt <sup>77</sup> ..	45
Figure 20: Conversion of a three dimensional matrix with multiple variables into a two dimensional component space showing the relation between principal components <sup>82</sup> .....	49
Figure 21: A basic radial basis function network outlay (By Kjell Magne Fauske [CC BY-SA 3.0] via TEXample.net).....	52
Figure 22: Graphical representation and explanation of common types of cross-validation <sup>93</sup> .....	54
Figure 23: An example of a nominally linear trend having an overfitted model applied (By Ghiles [CC BY-SA 4.0, via Wikimedia Commons].....	56
Figure 24: Flow diagram of typical New Zealand lamb processing (B+LNZ, 2015) .....	60
Figure 25: Diagram of the location of the saddle on the lamb carcass (a); the removed saddle showing the ribs, <i>M. longissimus thoracis et lumborum</i> , otherwise known as the loin muscle (outlined) and <i>Psoas major</i> , or tenderloin, all attached (b) and the excised loin with its corresponding carcass tag (c).....	61
Figure 26: Loin sample and fat cap as placed on the hyperspectral machine prior to scanning. Light source is from the top of the image .....	64
Figure 27: Top down view of meat samples packed into modified atmosphere Packaging and placed into a 4°C chiller for colour display life measurement .....	66
Figure 28: Example gas chromatography output with peak height related to each specific fat signal intensity and therefore prevalence in the lamb loin sample.....	72
Figure 29: ASD LabSpec 5000 VIS-NIRS machine with custom probe head as used in this current research .....	73
Figure 30: Image of the Headwall hyperspectral setup as used for this study.....	75
Figure 31: A graphical representation of a loin sample after an algorithm had been run to separate regions of interest. Grey is background, blue is intramusclar fat, red is lean meat, green is subcutaneous fat .....	77
Figure 32: A Tukey boxplot for spread of pH values for all 2473 lamb <i>M. longissimus thoracis et lumborum</i> samples from all kills .....	83
Figure 33: A Tukey boxplot for spread of tenderness values in kgf for all 1716 lamb <i>M. longissimus thoracis et lumborum</i> samples from all kills .....	84
Figure 34: A Tukey boxplot for spread of IMF values in %TAG for all 1678 lamb <i>M. longissimus thoracis et lumborum</i> samples from all kills .....	85
Figure 35: $R^2_{\text{cross-validation}}$ for all pre-processing models applied to the VIS-NIRS pH data analysed using partial least squares. Red cross highlights the best model found (highest $R^2$ ).....	88
Figure 36: $R^2_{\text{cross-validation}}$ for all pre-processing models applied to the VIS-NIRS IMF data analysed using partial least squares. Red cross highlights the best model found (highest $R^2$ ).....	88
Figure 37: $R^2_{\text{cross-validation}}$ for all pre-processing models applied to the VIS-NIRS tenderness data analysed using partial least. Red cross highlights the best model found (highest $R^2$ ) .....	89
Figure 38: Measured (as by pH meter) versus predicted (by PLS) pH values of 2015 data, separated by abattoir. Different colours represent different kills.....	91
Figure 39: Graph showing the measured pH (as by pH meter) at Abattoir B versus the predicted (developed using VIS-NIR spectra and PLS modelling obtained from Abattoir B) for all abattoirs. Different colours represent different kills. SEP = Standard Error of Prediction. ....	92

Figure 40: Graph showing the measured pH (as by pH meter) at Abattoir A versus the predicted (developed using VIS-NIR spectra and PLS modelling obtained from Abattoir B) for all abattoirs. Different colours represent different kills. SEP = Standard Error of Prediction. ....	93
Figure 41: Actual (measured using GCFID) versus predicted (by variable selection) measurements of Intramuscular fat using HSI data.....	96
Figure 42: Actual (measured using pH meter) versus predicted (by variable selection) measurements for pH using HSI data .....	96
Figure 43: First attempt of neural network modelling for intramuscular fat using HSI data (best R <sup>2</sup> values achieved shown in red).....	99
Figure 44: Second attempt of neural network modelling for intramuscular fat using HSI data with smaller decay rates applied (best R <sup>2</sup> values achieved shown in red) .....	99
Figure 45: Third attempt of neural network modelling for intramuscular fat using HSI data (best R <sup>2</sup> values achieved shown in red).....	100
Figure 46: Fourth attempt of neural network modelling for intramuscular fat using HSI data (best R <sup>2</sup> values achieved shown in red).....	100
Figure 47: Fifth attempt of neural network modelling for intramuscular fat using HSI data (best R <sup>2</sup> values achieved shown in red).....	101
Figure 48: Graph of intramuscular fat percentage measured (by gas chromatography) versus predicted (by neural network) from HSI data. Solid trendline = calibration data (1031 samples) Dashed trendline = validation data (515 samples) .....	102
Figure 49: Equations used to calculate predictive ability of final model using neural networking HSI data <sup>101</sup> .....	103
Figure 50: Graph of pH measured (by pH meter) versus predicted (by neural network) from HSI data. Solid trendline = calibration data. Dashed trendline = validation data .....	105
Figure 51: Graph of combined highest R <sup>2</sup> values for all modelling techniques from both VIS-NIRS or HSI data completed to this point and discussed previously. (SVM and PLS-R were performed independently of this current work).....	106
Figure 52: Count and distribution of tenderness data grouped by individual kill .....	129
Figure 53: Count and distribution of Intramuscular fat data grouped by individual kill .....	129
Figure 54: Count and distribution of pH data grouped by individual kill.....	130
Figure 55: a Tukey boxplot showing tenderness data spread grouped by individual kill.....	130
Figure 56: A Tukey boxplot showing intramuscular fat data spread grouped by individual kill.....	131
Figure 57: A Tukey boxplot showing pH spread grouped by individual kill .....	131
 Equation 1: showing the esterification reaction of a carboxylic acid with methanol using a boron trichloride catalyst. ....	36