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On line technology to predict beef and sheep meat quality traits using near infra red and other predictive techniques

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Abstract

Visible near infrared spectroscopy (VISNIR) is a fast, environmentally friendly analytical method that has gained widespread acceptance in the food industries in recent years to measure and importantly, pay suppliers on the quality attributes of their products. The study found that VISNIR spectroscopy could accurately measure and classify sheep meat on a number of commercially important traits that influence the appearance and/or palatability of red meat. Positive results are reported for VISNIR spectroscopy abilities to measure objective colour, intramuscular fat and pH at 24 hours post slaughter and muscle glycogen and/or glycolytic potential and muscle heme pigment levels at 30 minutes post slaughter. In this study VISNIR spectroscopy did not provide a reliable prediction of objective tenderness.

Executive Summary

Near infrared spectroscopy (NIR) is a fast, environmentally friendly analytical method that has gained widespread acceptance in recent years. Visible/NIR spectroscopy (VISNIR) is an expansion on NIR in that it collects spectra across both the visible and near infrared spectral wavelength regions. Providing the spectral information can be calibrated to one or more specific chemical constituents of meat VISNIR technology offers the potential to measure these constituents in real time. Application of NIR to “on line” uses in the meat industry has lagged behind other food based industries. NIR is widely used in the food industry to measure and importantly, pay suppliers on the quality attributes of their products

The results provide strong support for the continued development of VISNIR spectroscopy to accurately measure and classify carcasses on a number of commercially important traits of red meat. These include the ability to measure objective colour, intramuscular fat and pH at 24 hours post slaughter, muscle glycogen and/or glycolytic potential and muscle heme pigment levels at 30 minutes post slaughter. All these traits influence the appearance and palatability of red meat which are factors known to influence consumers purchasing decisions In this study, VISNIR spectroscopy did not provide a reliable prediction of objective tenderness. Currently most of these traits are not routinely objectively measured by the commercial red meat processing industry largely because the current measurement technology, where available, is manual and must be done on the cold carcass to reliably grade carcasses on pH or colour defects. The results of this project suggest VISNIR technology can overcome some of these limitations to provide the red meat industry with an alternative multi trait measurement technology to better manage and reduce the direct costs associated with carcasses downgraded for colour or pH related defects. Industry surveys estimate the incidence of such defects to be between 8 to 10% of the national kill.

Collectively the results support the concept that VISNIR technology can be developed to rapidly, objectively and cost effectively measure and grade carcasses on a suite of meat quality traits. The meat quality traits most suited to being measured by VISNIR are those controlled by the chemical status of muscle. Whilst most processors do not recognise it the conversion of muscle to meat is a complex chemical process which they control through their processing and chilling management practices. If coupled with MLA’s substantial investment in livestock traceability and automated processing systems VISNIR technology has the potential to add considerable value to lamb and beef supply chains. It could achieve this by providing supply chain participants with objective feedback on meat quality that has greater diagnostic benefits for supply chains to enhance the more consistent achievement of end user product specifications. Other food industries that now regularly use VISNIR technology have done so on the basis of if you can measure it you can manage it. This philosophy has equal relevance to the meat industry. For the red meat industry to get the best return from their investment in any further development of meat measurement technology using VISNIR it is critical it be done following a well executed business plan. Some of the key issues that need early resolution include the meat characteristics to be measured of highest priority to industry, availability of expertise given that it requires the skills of a multi disciplinary team which are predominantly available external to the meat industry, instrument manufacturer selection to ensure a seamless and cost effective interchange from research to commercialisation.

Contents

1 Background

Near infrared spectroscopy (NIR) is a fast, environmentally friendly analytical method that has gained widespread acceptance in recent years. Visible/NIR spectroscopy (VISNIR) is an expansion on NIR in that it collects spectra across both the visible and near infrared spectral wavelength regions. Providing the spectral information can be calibrated to one or more specific chemical constituents of meat VISNIR technology offers the potential to measure these constituents in real time. The meat industry has used NIR (near infrared spectroscopy) in the laboratory for routine fat and protein determinations where its precision and accuracy are fully equal to the wet chemical methods it has replaced. The advantages of NIR include:

- it gives faster results at a reduced cost,
- it is non destructive and requires no sample preparation, solvents or reagents,
- several NIR calibrations may also be used simultaneously on a single sample to give rapid, accurate multiple constituent analysis

Application of NIR to “on line” uses in the meat industry has lagged behind other food based industries. NIR is widely used in the food industry to measure and importantly, pay suppliers on the quality attributes of their products. Grain growers are paid on NIR estimates of the protein content of their grain. Similarly, sugarcane farmers are paid on NIR estimates of the sugar content of their cane.

Published reports indicate a variety of potential commercial applications aimed at enhancing the red meat industry’s commercial competitiveness and its ability to improve the consistency of its products to consumers. Downey and Beauchene (1997) reported NIR technology could discriminate between fresh and frozen/thawed beef. Consequently, NIR could be used by food regulatory bodies to guarantee the authenticity of labelling of fresh versus frozen beef. Moss et al. (2000) reported that NIR could be used to discriminate between irradiated and non-irradiated pork with a high degree of accuracy. Forrest et al. (2000) showed that NIR spectra collected in pork carcasses 30 minutes post exsanguination were highly correlated with 24 hr drip loss (r=0.84, RMSEP of 1.8% drip loss). Anderson et al. (1999) investigated whether pH in pork could be measured spectroscopically in reflectance using the visual and near infrared regions. A moderate correlation (r=0.791 RMSEP 0.077 pH units) was reported between NIR predicted pH and measured pH using a standard glass electrode pH meter for two pork muscles measured 24 hours post exsanguination. Josell et al (2000) reported NIR could be used to measure the glycolytic potential of pork immediately post slaughter and in doing so differentiate between RN – and RN+ phenotype pigs. Glycolytic potential of muscle immediately post slaughter is a key factor influencing the rate of biochemical events that occur in the conversion of muscle to meat post slaughter

NIR could also be used to measure fresh meat colour which is strongly influenced by the concentration and oxidative status of the muscle pigments myoglobin (Mb) and haemoglobin (Hb) the latter which can partially remain in muscle tissue as residual blood. Muscles containing a high concentration of haemoglobin show a more rapid discolouration, especially during freezing causing an undesirable product appearance (Hunt & Kromf, 1987). Although the majority of haemoglobin in carcass muscles is removed at slaughter the haemoglobin content remaining in the meat is dependant on stunning method, stun to stick interval and bleeding time (Lawrie, 1998; Ahn and Maurer, 1989). Bleeding is considered to be enhanced if the stun to stick interval is less than 5 seconds and a thoracic stick is used although the latter may exclude the product from some halal markets. Currently, there is no objective method to monitor whether carcasses have been effectively bled even though it is considered to influence several commercially important meat quality traits including colour, retail display life, microbial shelf life and the incidence of blood splash (or eccymosis). VISNIR spectroscopy applications have been developed for the chicken processing

industry to detect meat quality defects attributed to ineffective bleeding. This is possible because the major pigments in meat have unique optical absorption characteristics. In well bled red meat the major pigment is myoglobin. Elevated haemoglobin pigment levels indicate poorly bled meat. Rickansrud and Henrickson (1967) reported that the proportion of haemoglobin of total pigments in beef muscle were 20.1%, 25.0%, 31.8% and 37.0% for the longissimus dorsi, biceps femorus, semitendinosus and psoas major muscles respectively when compared on a dry fat free weight basis. Abramson (1962) suggested that the observed differences between muscle groups in both myoglobin and residual haemoglobin levels is associated with vascular patterns which vary widely from one muscle to another. A more detailed review of the literature supports the concept that VISNIR technology could be used to measure both haemoglobin and myoglobin pigments in muscle and their various oxidative states. It is based on the unique spectral characteristics of myoglobin and haemoglobin that have been extensively studied since the 1920's. Kryzwicki (1979) suggested that the colour image of beef was dependant on both the total concentration of the myoglobin pigments in muscle and the relative status of the different oxidative forms of myoglobin. Kryzwicki (1979) also recommended that the unique optical properties of the meat surface also needed to be considered when estimating meat colour that were independent of muscle pigment influences on meat colour image. Kryzwicki (1979) defined the spectral absorption and diffusion wavelengths that could be used to estimate both the pigment and non pigment related factors that influence meat colour. The wavelengths defined by Krzywicki (1979) are the same as those recommended by the American Meat Science Associations Guidelines for Meat Colour Evaluation (1991). Similarly Warris (1976) has described an optical method to measure muscle haemoglobin levels. Mancini & Hunt (2005) conducted an extensive review of the options for measuring instrumental colour including VISNIR spectroscopy. They suggest that spectral information may potentially provide greater diagnostic benefits to identify and manage the key factors that influence meat colour and pH than the more widely used colour coordinate systems (Lab or XYZ).
This project aims to evaluate the potential of VISNIR spectroscopy to measure on line muscle objective tenderness, glycolytic potential and myoglobin and haemoglobin pigment levels. All 3 are recognised as key factors that impact on the visual and functional properties of red meat. It also proposes to investigate some other non VISNIR technology options for the low cost measurement of meat pH. The study was conducted at the MRTC Werribee using lamb and mutton

2 Project Objectives -

1. Assess the predictive accuracy of VISNIR spectroscopy to measure the objective tenderness and intra-muscular fat content of lamb;
2. Investigate the predictive accuracy of VISNIR spectroscopy to measure muscle heme pigment levels as indicators of blood content (sticking efficiency) and their effect on ultimate pH and meat colour
3. Validate a method for using VISNIR spectroscopy to accurately measure and classify carcasses on their pre-rigor muscle glycogen and lactate levels on the slaughter floor with 90% accuracy;
4. Develop and assess the accuracy of an accelerated freeze/thaw method to induce and measure ultimate muscle pH within 30 minutes of slaughter;
5. Review, develop and validate a new generation low cost data logger that can record time and temperature of muscle(s) at the onset of rigor.

3 VISNIR spectroscopy to measure objective tenderness and intra-muscular fat content

3.1 Methodology -

3.1.1 Animals

VISNIR spectra and reference samples were collected on 529 lamb carcasses slaughtered at G& V Hardwick's Pty Ltd, Kyneton. The lambs were part of the scheduled slaughter of over 900 lambs from the MLA/ AWI sheep genomics project. The carcasses were transported to the Meat Research & Training Centre Werribee within 24 hours of slaughter for boning and further processing.

3.2.2 Reference measurements

Detailed individual carcass information was collected on the 529 lamb carcasses assessed. Carcass weight, P8 fat depth and sex were recorded for all individual carcasses on the day of the kill. Muscle ultimate pH and objective colour (Hunter Lab L, a & b values) were measured at 24 hours post slaughter. Table 3.1 summarises the key meat quality reference measurements selected to calibrate with the multi wavelength spectral scan information collected on the loin muscle (*M. longissimus thoracis et lumborum)* at 30 minutes, 1 and 5 days post slaughter. The 30 minutes spectral measurements were collected on the kill floor during processing at a chain speed rate of 5.5 carcasses per minute.

Table 3.1 Summary of the meat quality traits and time points measured for each loin

Meat quality Attribute	Time post slaughter		
	0.5 hours	1 day	5 days
MWS scan			
Shear force			
Compression (hardness, chewiness and cohesiveness)			
Intra muscular fat %			
Ultimate pH			
Objective colour (Hunter Lab L, a & b)			

The loin muscle (*M. longissimus thoracis et lumborum*) from the left side of each carcass was boned and cut into four sections. Each section was allocated to one of the four reference measurements in accordance with a pre-determined allocation procedure. Objective measurements of shear force at 1 and 5 days post slaughter, compression values (hardness, cohesiveness & chewiness) at 1 day only

and intramuscular (IM) fat content of the loin muscle were conducted using standard SMEQ protocols.

3.1.2 VISNIR spectroscopy:

Spectral data (350 to 2500nm) was collected at the nominated time points using an ASD Field Spec Pro instrument operating in reflectance mode. An optical fibre probe was placed on the loin muscle sample. An instrument setting of 15 scans per measurement site was used with the spectral measurement collected only once at 30 minutes post slaughter, due to chain speed constraints, with the fibre optic probe orientated perpendicular to muscle fibres. At subsequent time points (1 and 5 days post slaughter) the spectral measurements were collected twice with the probe either orientated perpendicular or parallel to the muscle fibres.

3.1.3 Statistical analysis

The data has been analysed using the Unscrambler chemometrics software package. Partial least square regression (PLS) was used to derive calibrations for all objective meat quality traits measured with the VISNIR spectra collected. A standard data pre treatment procedure was used for all PLS regressions with all raw reflectance data converted to the 1st derivative. Spectral data under 380nm and over 2100nm was excluded from the analysis on the basis of there being a high level of noise in these areas of the spectrum. Also when developing the regression model estimates the jackknife statistical procedure was used. This procedure statistically selects the spectral wavelengths that are most informative to the reference measurement trait of interest and excludes those that are statistically least informative. No outliers were removed from the analysis. The predictive accuracy of the models is given by the root mean square error of prediction (RMSEP). This value indicates the average uncertainty that can be expected when predicting Y values for new samples expressed in the same units as the Y variable. The number of PLS factors indicates the complexity of the calibration model with a lower number indicative of a more stable and robust model across data sets.

3.2 Results

3.2.1 Summary statistics

A statistical summary of the means, standard deviations and range in the objective meat quality traits measured are summarised in Table 3.2

Table 3.2 Statistical summary of the carcass and meat quality measurements recorded for the lambs used in the study (n=529)

Carcass/ meat quality trait	No.	Mean	Std deviation	Range
Hot carcass weight (kg)	529	18.48	2.05	12.90 to 26.30
GR fat depth (mm)	529	6.9	3.16	1 to 18
Shear force 1 day (kg)	517	4.04	1.12	1.37 to 7.52
Shear force 5 days (kg)	527	2.63	0.74	1.31 to 6.30
Hardness (kg)	529	3.109	0.54	1.22 to 6.20
Cohesiveness	528	0.27	0.03	0.16 to 0.38
Chewiness	528	0.84	0.18	0.24 to 1.67
Hunter lab L	529	33.0	2.71	23.70 to 41.01
Hunter lab a	529	7.21	1.29	2.92 to 10.60
Hunter lab b	529	8.23	1.33	2.97 to 11.49
pHu	529	5.69	0.23	5.40 to 6.98
Intra muscular fat	311	4.09	1.66	1.25 to 10.92

3.2.2 VISNIR calibration models

Partial least squares calibration model estimates using the VISNIR spectral data (380 to 2100nm) to predict the objective meat quality traits measured are summarised in Tables 3.3 & 3.4.

Table 3.3 Prediction model estimates for loin muscle shear force, hardness, cohesiveness and chewiness values using multi wavelength spectra collected at 30 minutes,1 & 5 days post slaughter

Meat Quality Trait	Time spectra acquired post slaughter	Probe orientation to muscle fibres	No. of samples	Model estimates			
				r^2	RMSEP	Ratio of RMSEP:SD	PLS factors
Shear force 1 day (kg)	30 mins	Perp	479	0.08	1.07	0.96	1
	1 day	Perp	423	0.23	1.01	0.91	5
	1 day	Parallel	517	0.33	0.92	0.82	6
Shear force 5 days (kg)	30 mins	Perp	488	0.02	0.74	1.00	1
	1 day	Perp	432	0.20	0.70	0.95	8
	1 day	parallel	527	0.28	0.63	0.85	8
	5 days	Perp	514	0.10	0.71	0.96	2
	5 days	parallel	526	0.19	0.67	0.91	6
Hardness 1 day (kg)	30 mins	Perp	490	0.10	0.54	0.73	1
	1 day	Perp	434	0.16	0.45	0.80	5
	1 day	parallel	529	0.15	0.52	0.93	3
Cohesiveness 1 day (joule)	30 mins	Perp	489	0.10	0.03	1.00	1
	1 day	Perp	433	0.19	0.03	1.00	1
	1 day	parallel	528	0.19	0.03	1.00	5
Chewiness 1 day (kg× joule)	30 mins	Perp	489	0.12	0.17	0.94	1
	1 day	Perp	433	0.23	0.14	0.78	4
	1 day	parallel	528	0.30	0.15	0.83	5

Muscle shear force models

Mean loin shear force values measured at 1 and 5 days post slaughter for 529 lamb loins declined, as expected, with time post slaughter. The mean shear force value was 4.04 ±1.12 kg at 1 day post slaughter and 2.63 ± 0.74kg by 5 days post slaughter. Such shear force values indicate that the majority of the lamb was tender particularly by 5 days post slaughter. Muscle VISNIR spectral measurements were taken at 30 minutes, 1 and 5 days post slaughter. Statistical estimates for r^2, Root Mean Square Error of Prediction (RMSEP) and the ratio of RMSEP/Std Deviation for PLS calibration models shear force at both 1 and 5 days indicated that the spectral data were of limited value. The best calibration model derived for the prediction of loin shear force values at 1 day post slaughter was using muscle spectral data acquired at day 1 post slaughter with the probe orientated parallel to the muscle fibres (r^2=0.33, RMSEP 0.92kg, PLS factors 6). Calibration models estimates of shear force were consistently lower using spectral data acquired at 30 minutes post slaughter. The likely explanation for this is that at 30 minutes post slaughter muscle tissue is still pre-rigour hence undergoing considerable chemical and physical change in comparison to post rigour muscle that has reached a much more stable chemical and physical status. Calibration model estimates of muscle 5 day shear force values were lower than comparable 1 day shear force values. The likely explanation is because of the effect of ageing on substantially reducing the variation in shear force values of 5 day compared to 1 day samples as evidenced by the lower standard deviation values. Under such circumstances calibration models will have lower r^2 value.

Compression measurement models

Muscle compression values for hardness, cohesiveness and chewiness were measured on the 529 loins at 1 day post slaughter only (Table 3.2). Mean values were 3.09 ±0.56, 0.27 ±,0.03kg and 0.84 ± 0.18kg respectively. PLS calibration model estimates of hardness, cohesiveness and chewiness indicated that the spectral data was again of limited value to predict any of these muscle compression values. The fact that all three traits are mathematically derived from the same base data means it is not surprising that the predictive accuracy of the PLS models generated for hardness, cohesiveness and chewiness are comparable.

Table 3.4 Prediction model estimates for loin muscle intramuscular fat percentage, ultimate pH and objective colour values using multi wavelength spectra collected at 30 minutes,1 & 5 days post slaughter

Trait	Time post slaughter (days)	Probe orientation to muscle fibres	No. of samples	Model estimates			
				r^2	RMSEP	RMSEP/SD	PLS factors
Intramuscular fat %	0	perp	306	0.45	1.23	0.74	5
	1	perp	287	0.64	0.96	0.57	9
	1	parallel	311	0.51	1.13	0.68	2
	5	perp	301	0.51	1.13	0.68	5
	5	parallel	311	0.44	1.24	0.74	5
Ultimate pH	0	perp	490	0.29	0.20	0.87	4
	1	perp	429	0.76	0.12	0.52	9
	1	parallel	529	0.71	0.11	0.48	7
	5	perp	527	0.71	0.13	0.57	6
	5	parallel	527	0.64	0.14	0.61	9
Hunter lab L	0	perp	489	0.26	2.37	0.88	3
	1	perp	434	0.53	1.62	0.60	2
	1	parallel	529	0.72	1.41	0.44	3
Hunter lab a	0	perp	490	0.34	1.06	0.82	4
	1	perp	434	0.66	0.78	0.60	5
	1	parallel	529	0.69	0.73	0.57	6
Hunter lab b	0	perp	490	0.21	1.19	0.89	3
	1	perp	434	0.59	0.81	0.61	4
	1	parallel	529	0.74	0.69	0.52	3

Objective meat colour models

The Hunter lab co-ordinate system was used to measure loin muscle objective meat colour at 24 hours post slaughter after a 30 minute bloom time. Mean meat colour measurements of the 529 lamb loins were L (lightness) 33.0 ± 2.71, a (redness) 7.21 ± 1.29 and b (yellowness) 8.23 ± 1.33.

Lightness and redness are known to be important indicators of the products appearance that influence appeal to consumers. VISNIR spectral data provides an alternative approach to measuring meat colour. The PLS calibration models generated for L, a & b values using the muscle spectral reflectance data acquired at 1 day post slaughter indicate that the VISNIR system could be reliably used to sort carcasses on colour. The best models for L and a colour measurements were obtained when the spectra was acquired at 1 day post slaughter with the probe parallel to the muscle fibres (r^2 values of 0.69 & 0.74 , RMSEP 1.41 & 0.73 and RMSEP/SD 0.52 & 0.57). Once again calibration model estimates of L,a & b were much lower for spectra collected at 30 minutes post slaughter most probably for the same reasons suggested previously for shear force values.

Ultimate pH models
Mean loin muscle pH measured at 24 hours post slaughter (pHu) was 5.69 ±0.23. The best calibration model developed for pHu was attained when spectra was collected at 1 day post slaughter (r^2=0.77, RMSEP 0.11 pH units, RMSEP/SD 0.48). Orientation of the optical fibre probe to the muscle fibres made no statistical difference to model accuracy. There was a substantial decline in the accuracy of the calibration model for pHu for spectra acquired at 30 minutes post slaughter but not at 5 days post slaughter.

Intramuscular fat percentage models
Objective reference measurements of intramuscular fat levels were limited to 311 of the 529 loins for budgetary reasons. Mean intramuscular fat content was 4.09% ± 1.66. The best calibration model developed for intramuscular fat percentage was attained when spectra was collected at 1 day post slaughter perpendicular to the muscle fibres(r^2=0.64, RMSEP 0.96%, RMSEP/SD 0.57). Calibration model estimates were lower but still useful when measured at either 30 minutes or 5 days post slaughter.

3.3 Discussion

Objective tenderness
This study found that VISNIR technology had limited ability to measure the objective tenderness (shear force & compression values) of lamb with the calibration models calculated being of limited predictive accuracy but comparable to those reported in other previous studies. VISNIR technology is considered to be a particularly valuable tool to measure one or more unique chemical characteristics of a substance. It has particular limitations for measuring the mechanical properties of a substance such as objective tenderness unless the mechanical properties of the substance are largely dictated by one or more distinct chemical bonds. Conversely, if the mechanical properties of the substance are controlled by a more amorphous group of chemical bonds then VISNIR technology is less likely to be able to be calibrated to the trait(s) of interest. An alternative approach to enhance the ability of VISNIR to predict muscle tenderness would be to identify one or more of the specific chemical bonds in the muscle matrix that have a large influence on tenderness. If such biomarkers do exist then this would render shear force and compression values obsolete as measures of objective tenderness.

It is interesting to note that a VISNIR based tenderness classification system has been developed in the USA to grade select beef carcasses. The application is designed to grade select carcasses into an above or below average tenderness category based on predicted tenderness after 14 days ageing using the loin muscle spectra acquired at 1 day post slaughter. The application has been developed by ASDI in collaboration with USDA meat scientists Steve Shackelford and Mohamed Koohmaraie after the commercial meat industry rejected a conventional shear force assessment process as impractical. ASDI is the same manufacturer of the spectrometer used in this study,.

Published results indicate that the VISNIR prediction model estimates that underpin this particular application are comparable to the best prediction model reported in this study. However, the application has still progressed on the basis that the commercial advantages of being able to rapidly measure, sort and brand select carcasses with above average tenderness more than offsets the costs of some misclassification errors. It is still too early to judge whether the USA VISNIR tenderness application will be a commercial success.

Objective meat colour

VISNIR spectral data provides an alternative approach to measuring meat colour. The PLS prediction models generated for L, a & b values using the muscle spectral reflectance data acquired at 1 day post slaughter indicate that the VISNIR system could be reliably used to sort carcasses on colour. It is not essential that spectral estimates of L, a & b are highly correlated to colorimeter L, a & b co-ordinate systems (Hunter lab and CIE). Instrumental metamerism is a common and serious defect of colorimeters , even for the same make and model. This means it is difficult to get the same spectral response from two colorimeters of the same make and model. It has also been shown that the measurement of spectral reflectance measurement to be superior to colorimeters in their ability to discern discolouration defects in meat on retail display. This is because of significant overlapping in colorimeter L & a values measured for red and brown coloured meat. This problem was evident in MLA project VBCSH 017 where CIE values of L and a could not discern quite marked visual colour differences in MAP and overwrapped packs of lamb associated with metmyoglobin accumulation that impacted on their retail acceptability. VISNIR technology potentially offers an alternative option to the current visual system used to assess meat colour. With further development it may also provide a more reliable method to determine the best before colour date on packaged meat. Colour and best before dates are frequently the only factors by which consumers judge the quality of a product.

Muscle pH

The results of this study indicate that VISNIR spectroscopy could provide a more rapid option for measuring ultimate pH with comparable accuracy to conventional methods but would be a higher cost option unless VISNIR spectroscopy was used for measuring multiple meat quality traits and not just muscle pH alone. Conventional methods for measuring pH are labour intensive, slow and prone to error due to instrument calibration drift and/or operator error. Industry is seeking a low cost rapid method that would enable carcasses to be graded on ultimate pH whilst still on the kill floor. Such an option is being investigated in studies 3 & 4. Without such a low cost method industry will continue with their current practise of not routinely measuring ultimate meat pH even though national audits have shown that around 8 to 10% of cattle and lamb carcasses processed have a high ultimate pH (pH >5.8). In some weeks of unfavourable weather this can spike to 25% of the daily kill.

Intramuscular fat

The best prediction model developed for intramuscular fat percentage was attained when spectra was collected at 1 day post slaughter perpendicular to the muscle fibres(r^2=0.64, RMSEP 0.96%, RMSEP/SD 0.57). Prediction model estimates were lower but still useful when measured at either 30 minutes or 5 days post slaughter.

A rapid accurate method of measuring intramuscular fat levels in lamb would enable commercial industry the ability to offer high value markets a differentiated lamb product with above average intramuscular fat levels. Comparable markets are already well established for marbled beef (high intramuscular fat) that attract clear price premiums.

4 VISNIR spectroscopy to measure muscle heme pigments and their effect on ultimate pH and colour

4.1 Methodology

4.1.1 Animals

VISNIR spectra and objective meat quality reference samples were collected on a total 24 lamb and 24 mutton carcasses slaughtered at the MRTC Werribee over three separate kill dates of 16 head. All sheep processed were from commercial flocks of known vendor histories. Different vendors were selected for each kill group to facilitate variation in the meat quality traits measured**.**

4.1.2 Treatments

A 4 × 2 factorial study was designed to assess the effects of age category, stick method and muscle type on muscle pH and meat colour
age category a total of 24 lambs and 24 mutton were processed over 3 separate kill dates for the study
stick method all lambs and mutton slaughtered were allocated to either a normal stick or a thoracic stick method
muscle type- measurements were collected on 4 muscle groups that varied according to fibre type and position within the carcass. The four muscles measured were the loin (*M. longissimus thoracis et lumborum)*, chuck tender muscles *(M. supraspinatus)* silverside *(M.semitendosis)*, and shin *(M biceps brachii)*.
Sex 31 ewes and 17 wethers processed over the 3 kill dates

In addition to the above 4 treatments, probe orientation to the muscle fibres was also assessed with regard to its effect on light absorption and diffusion by pigment and non pigment related factors of muscle. Spectra were collected with the probe held both parallel and perpendicular to the muscle fibres.

4.1.3 Carcass measurements

Individual carcass weights, sex and vendor ID were recorded on the day of kill. Muscle pH and temperature profiles for all four muscles were recorded at 30 minutes, 3, 6 and 24 hours post slaughter. At 24 hours post slaughter objective meat colour (Hunter Lab L, a & b values) were recorded after a 30 minutes bloom for all muscles.

4.1.4 Muscle myoglobin and haemoglobin levels

Spectral absorption and diffusion (A) values for total myoglobin (A525nm less A730nm) , total myoglobin and haemoglobin (A550nm-A730) and non pigment related factors (A730nm) were measured at 30 minutes, 3 hours and 24 hours post slaughter using an ASD Lab Spec Pro instrument. Reflectance (R) values were converted to absorption and diffusion values according the formula A= log 1/R. An optical fibre probe was placed on the surface of the muscle and a setting of 15 scans per measurement site used. Spectra were collected with the fibre optic probe orientated both parallel and perpendicular to muscle fibres.

4.1.5 Statistical analysis

Carcass traits muscle pH, meat colour (Hunter Lab L, a & b values) and muscle pigment and non pigment absorption and diffusion values for defined wavelengths were analysed using the method of restricted maximum likelihood (REML) with age category, stick method, muscle type and sex as fixed effects and week of kill, carcass and side of carcass as random effects. The predictive ability of muscle pigment levels measured at 30 minutes on pH and objective colour values (L, ,a &b) measured at 24 hours was assessed by regression allowing for week of kill, age category, sex and muscle type. Genstat statistical program version 7.1 (2003) was used for analyses.

4.2 Results & Discussion

4.2.1 Summary statistics

Table 4.1 summarises the key meat quality reference measurements collected to assess the effect of age category on meat colour, muscle pH and muscle pigment levels at nominated time points post slaughter.

Table 4.1 Statistical summary of the carcass weight, fat depth, muscle pH ,objective meat colour, light absorption (A) values attributable to muscle heme pigments and other non pigment related factors pooled for four primal muscles

Carcass/ meat quality trait	Time post slaughter (hours)	No. of samples	Mean	Std deviation	Range
Hot carcass weight (kg)		48	25.3	3.01	17.6 to 33.1
L Hunter lab	24	144	34.54	3.86	26.48 to 42.45
a Hunter lab	24	144	7.23	2.24	1.94 to 14.14
b Hunter lab	24	144	8.76	1.93	4.16 to 15.11
pH	0.5	288	6.61	0.21	6.04 to 6.99
	3	288	6.43	0.23	5.76 to 6.94
	6	288	6.23	0.19	5.66 to 6.79
	24	288	5.87	0.22	5.47 to 6.70
A Mb (A525-A730 nm)	0.5	288	0.538	0.125	0.060 to 0.873
	3	288	0.561	0.092	0.105 to 0.909
	24	288	0.565	0.080	0.318 to 0.819
A Mb/Hb(A550-A730nm)	0.5	288	0.670	0.146	0.159 to 1.099
	3	288	0.720	0.112	0.153 to 1.183
	24	288	0.745	0.090	0.445 to 1.013
A Nil pigment (A730nm)	0.5	288	0.566	0.084	0.296 to 0.765
	3	288	0.597	0.101	0.360 to 0.923
	24	288	0.464	0.086	0.294 to 0.726

4.2.2 Muscle pH

Muscle pH was significantly influenced by animal age category, sex and muscle type but not by stick method (Table 4.2). Muscle pH was significantly lower ($P<0.001$) in lamb compared to mutton carcasses at 24 hours post slaughter. There were no significant differences between lamb and mutton in muscle pH prior to 24hours post slaughter. There were significant differences ($P<0.001$) in muscle pH between the four muscles types at all time points measured with the loin muscle having

the highest initial pH of the four muscles measured at 30 minutes post slaughter but by 24 hours post slaughter it had the lowest pH.

Table 4.2 Summary of the effects of age category, stick method, sex and muscle type on muscle pH at 30 minutes, 3, 6 & 24 hours post slaughter

Treatment		No.	pH 0.5 hrs	pH 3hrs	pH 6hrs	pH 24hrs
Age category	lamb mutton sed p*	24 24	6.61 6.61 0.034 ns	6.42 6.43 0.03 ns	6.19 6.26 0.03 0.056	5.82 5.92 0.03 <0.05
Stick method	normal thoracic sed p*	24 24	6.62 6.61 0.032 ns	6.43 6.42 0.03 ns	6.24 6.22 0.03 ns	5.87 5.87 0.03 ns
Sex	ewe wether sed p	31 17	6.60 6.63 0.034 ns	6.38 6.47 0.03 <0.05	6.18 6.28 0.03 <0.001	5.84 5.91 0.03 0.06
Muscle	chuck tender loin shin silverside sed p*	96 96 48 48	6.74 6.80 6.41 6.50 0.022 <0.001	6.47 6.66 6.33 6.24 0.02 <0.001	6.22 6.39 6.20 6.09 0.02 <0.001	5.92 5.67 6.04 5.85 0.02 <0.001
Interactions			AMSex*** *	AMSex*** *	MSex* ASex** AMSex**	ASex**

* P<0.05
** P<0.01
*** P<0.001

4.2.3 Objective meat colour

Muscle lightness (L Hunter lab), redness (a Hunter lab) and yellowness (b Hunter lab) were measured at 24 hours post slaughter after a 30 minute bloom time. Table 4.3 shows that Hunter lab L & a 24 hour colour measurements were significantly influenced by age category, sex and muscle type but not stick method. Hunter lab 24 hour b colour measurements were only significantly influenced by muscle type and sex. Lamb muscle was significantly lighter (P<0.001) and less red (P<0.001) compared to mutton at 24 hours post slaughter. Significant differences (P<0.001) in muscle L, a & b colour values occurred between the three individual muscle types measured. There was insufficient shin muscle available for objective colour measurements at 24 hours post slaughter.

Table 4.3 Summary of the effects of age category, stick method, sex and muscle type on objective meat colour at 24 hours post slaughter

Treatment		No.	L Hunter lab 24 hrs	a Hunter lab 24hrs	b Hunter lab 24hrs
Age category	Lamb	24	35.77	6.25	8.78
	mutton	24	33.32	8.20	8.75
	sed		0.44	0.31	0.27
	p		<0.001	<0.001	ns
Stick method	normal	24	34.64	7.32	8.77
	thoracic	24	34.45	7.13	8.75
	sed		0.42	0.29	0.25
	p		ns	ns	ns
Sex	ewe	31	35.00	7.62	9.20
	wether	17	34.08	6.84	8.32
	sed		0.43	0.31	0.27
	p		0.06	<0.05	<0.01
Muscle	chuck tender	96	36.56	8.33	10.03
	loin	96	30.58	7.73	7.26
	silverside	48	36.48	5.63	8.99
	sed		0.44	0.25	0.29
	p		<0.001	<0.001	<0.001
Interactions					ASex* MASex*

4.2.4 Light absorption at 525nm attributable to muscle myoglobin

Light absorption attributable to muscle myoglobin was significantly influenced by animal age category (P<0.001) and muscle type (P<0.001) but not by stick method or sex (Table 4.4). Lamb muscle had significantly lower absorption values (P<0.001) compared to mutton at all time points measured. Significant differences (p<0.001) existed between individual muscles with the *M.semitendosis* having the lowest values and *M. longissimus thoracis et lumborum* the highest at 24 hours post slaughter.

Table 4.4 Summary of the effects of age category, stick method, sex and muscle type on muscle light absorption and diffusion (A) values attributable to myoglobin (Mb) at 30 minutes, 3 & 24 hours post slaughter with the probe orientated perpendicular to the muscle fibres

Treatment		No.	AMb (A525-A730) 0.5hrs	AMb (A525-A730) 3hrs	AMb A525-A730) 24hrs
Age category	Lamb	24	0.505	0.535	0.528
	mutton	24	0.569	0.587	0.603
	sed		0.017	0.014	0.009
	p		<0.001	<0.001	<0.001
Stick method	normal	24	0.536	0.557	0.560
	thoracic	24	0.539	0.565	0.571
	sed		0.016	0.003	0.009
	p		ns	ns	ns
Sex	Ewe	31	0.549	0.558	0.570
	wether	17	0.526	0.564	0.561
	sed		0.017	0.014	0.009
	p		ns	ns	ns
Muscle	Chuck tender	96	0.631	0.616	0.599
	loin	96	0.667	0.592	0.621
	shin	48	0.390	0.531	0.561
	silverside	48	0.463	0.506	0.479
	sed		0.015	0.016	0.008
	p		<0.001	<0.001	<0.001
Interactions				MSex*	AM** SM*

4.2.5 Light absorption at 550nm attributable to total muscle heme pigments (haemoglobin & myoglobin**)**

Table 4.5 shows that the light absorption and diffusion measured at 550nm, attributable to total muscle heme pigments, was similarly significantly influenced by animal age category (P<0.001) and muscle type (P<0.001) but stick method or sex were not significant.. Lamb muscle had significantly lower absorption values (P<0.001) compared to mutton at all time points measured. Significant differences (p<0.001) also existed between individual muscles with the *M.semitendosis* having the lowest values and *M. longissimus thoracis et lumborum* and *M. supraspinatus* the highest at 24 hours post slaughter. To a large extent the results on light absorption at 550nm and attributed to total heme pigments mirrors that reported for 525nm and attributed to myoglobin alone.

Table 4.5 Summary of the effects of age category, stick method, sex and muscle type on light absorption and diffusion values for total muscle heme pigments (haemoglobin & myoglobin) at 30 minutes, 3 & 24 hours post slaughter with the probe orientated perpendicular to the muscle fibres

Treatment		No.	AMb/Hb (A550-A730) 0.5hrs	AMb/Hb (A550-A730) 3hrs	AMb/Hb A550-A730) 24hrs
Age category	lamb	24	0.671	0.699	0.707
	mutton	24	0.729	0.741	0.784
	sed		0.022	0.017	0.011
	p		<0.01	0.053	<0.001
Stick method	normal	24	0.697	0.714	0.738
	thoracic	24	0.702	0.725	0.752
	sed		0.021	0.017	0.010
	p		ns	ns	ns
Sex	ewe	31	0.712	0.714	0.749
	wether	17	0.687	0.726	0.741
	sed		0.022	0.017	0.011
	p		ns	ns	ns
Muscle	chuck tender	96	0.816	0.782	0.795
	loin	96	0.824	0.720	0.773
	shin	48	0.540	0.697	0.756
	silverside	48	0.620	0.680	0.656
	sed		0.020	0.021	0.012
	p		<0.001	<0.001	<0.001
Interactions				AM* MSex*	AM** MS* MASex*

4.2.6 Light absorption at 730nm attributable to non pigment related muscle factors

Light absorption at 730nm attributable to non pigment related muscle factors was significantly influenced by animal age category (P<0.001)and muscle type (P<0.001) but not stick method or sex (Table 4.6). Lamb muscle had significantly lower absorption values (P<0.001) compared to mutton at all time points measured. Significant differences (p<0.001) also existed between individual muscles with the *M.supraspinatus* having the lowest and *M. longissimus thoracis et lumborum* consistently the highest values at all time points.

Table 4.6 Summary of the effects of age category, stick method, sex and muscle type on muscle light absorption and diffusion values attributed to non pigment related factors at 0.5, 3 & 24 hours post slaughter with the probe orientated perpendicular to the muscle fibres

Treatment		No.	Nil pigment (A730) 0.5hrs	Nil pigment (A730) 3hrs	Nil pigment (A730) 24hrs
Age category	lamb	24	0.538	0.575	0.441
	mutton	24	0.594	0.620	0.488
	sed		0.008	0.011	0.010
	p		<0.001	<0.001	<0.001
Stick method	normal	24	0.563	0.594	0.462
	thoracic	24	0.568	0.601	0.467
	sed		0.007	0.010	0.010
	p		ns	ns	ns
Sex	ewe	31	0.568	0.590	0.456
	wether	17	0.564	0.605	0.473
	sed		0.008	0.011	0.010
	p		ns	ns	ns
Muscle	chuck tender	96	0.481	0.488	0.406
	loin	96	0.655	0.690	0.554
	shin	48	0.543	0.562	0.469
	silverside	48	0.584	0.650	0.430
	sed		0.006	0.008	0.010
	p		<0.001	<0.001	<0.001
Interactions			AM* MSex* AMSex*		MSex*

4.2.7 Regression models to predict muscle pH and objective colour

To assess the effect of muscle pigment and non pigment related factors on muscle pH and colour regression models were calculated using muscle light absorption values at 525, 550 and 730nm measured at 0.5 hours post slaughter to predict muscle pH and objective colour L, a& b values at 24 hours post slaughter (Table 4.7). The regression models with all terms fitted is detailed below.

> **Response variates(s) (24 hour):** pH, Hunter Lab L,a & b colour values
> **Fitted terms:** constant + week+ age category+ muscle+ significant interactions + absorption value @ nominated wavelength (0.5 hours) i.e. 525nm or 550nm or 730nm

Muscle pH (24 hour) The regression models indicate that non pigment related factors measured at 0.5 hours post slaughter (730nm absorption values) had a significant influence (P<0.001) on muscle pH at 24 hours post slaughter. Pigment related factors (525 & 550nm absorbance values) also measured at 0.5 hours post slaughter also had a significant influence (P<0.001) on muscle pH at 24 hours but their influence was very small compared to non pigment related factors

Objective colour (24 hour) Similarly the regression models indicate that non pigment related factors measured at 0.5 hours post slaughter (730nm absorption values) had a significant influence (P<0.001) on muscle lightness (L) and yellowness b values at 24 hours post slaughter. Pigment related factors (525 & 550nm absorption values) measured at 0.5 hours post slaughter had a significant influence (P<0.001) on muscle lightness but not yellowness at 24 hours and their influence was smaller compared to non pigment related factors. Conversely the pigment related factors had a bigger influence on muscle redness a values than non pigment related factors.

Table 4.7 Regression model estimates for predicting muscle pH and objective colour L, a & b values at 24 hours post slaughter using all fitted terms with or without light absorption values at either 525, 550 and 730nm wavelengths measured at 0.5 hours post slaughter

Response variate	Fitted terms in model	Significance	Percent variation accounted for ($r^2 \times 100$)	Std error	Non significant factors
pH 24hr	all plus A525nm	<0.001	69.0	0.122	A525nm
	A525nm only	<0.001	7.2	0.212	
	all plus A550nm	<0.001	67.1	0.126	A550nm
	550nm only	<0.001	4.9	0.215	
	all plus A730nm	<0.001	67.8	0.125	
	A730nm only	<0.001	25.9	0.190	
L 24hr	all plus A525nm	<0.001	65.8	2.26	A525nm
	A525nm only	<0.001	20.3	3.45	
	all plus A550nm	<0.001	65.7	2.26	A550nm
	A550nm only	<0.001	13.7	3.59	
	all plus A730nm	<0.001	66.4	2.24	A730nm
	A730nm only	<0.001	39.9	3.00	
a 24 hr	all plus A525nm	<0.001	64.5	1.34	A525nm
	A525nm only	<0.001	25.4	1.94	
	all plus A550nm	<0.001	64.4	1.34	A550nm
	A550nm only	<0.001	19.6	2.01	
	all plus A730nm	<0.001	64.7	1.33	A730nm
	A730nm only	ns			
b 24 hr	all plus A525nm	<0.001	48.6	1.39	A525nm
	A525nm only	0.052	1.9	1.91	
	all plus A550nm	<0.001	48.6	1.39	A550nm

	A550nm only	ns			
	all plus A730nm	<0.001	48.7	1.39	A730nm
	A730nm only	<0.001	21.7	1.71	

4.3 Discussion

The results indicate that VISNIR spectroscopy can objectively measure muscle heme pigments and non pigment related factors of muscle that collectively influence meat colour. Age category and muscle type both significantly influenced pH, objective meat colour and spectral absorption values at wavelengths attributed to muscle heme pigments (525 and 550nm) and non pigment factors (730nm). Stick method had no significant effect on any of these traits. It is interesting to note that when muscle type is included in the regression model it can largely account for variation in spectral absorption values otherwise attributed to either pigment or non pigment related factors. This demonstrates that the inherent differences that exist between the descriptor of muscle type in the regression model accounts for pigment and non pigment related factors The relationship between spectral absorption values attributed to muscle heme pigments and other non pigment factors has previously been reported by Krzywicki (1979) who suggests that two distinctively separate modes of action on how light absorption at these wavelengths influences meat colour and/or pH.

The first mode appears to occur via a direct influence on the level of total myoglobin and/or haemoglobin pigments retained in muscle tissue after slaughter. Numerous studies have previously reported and explained the significant differences in muscle myoglobin levels that exist between animals of differing ages and different muscle types within the same carcass. Published studies on stick method are more limited and where reported have been for poultry, fish and frogs. Whilst not actually measured the thoracic stick method used in this study appeared to only marginally improve bleeding at slaughter hence it is not surprising that stick method had no significant effect on the optical properties of the muscle. The regression models calculated indicate that muscle pigment related factors did have a significant influence on objective colour redness scores. This is to be expected and is one of the key characteristics of red compared to white meat. However the regression models suggest that the muscle pigment levels had little or no effect on muscle lightness, yellowness or pH measured at 24 hours post slaughter.

According to Krzywicki (1979) light absorption at 730nm provides a measure of the optical properties of meat that influence light and oxygen penetration beyond the meat surface. A combination of factors is considered to influence light absorption at 730nm. These include pH, hydration status, fibre type, presence of marbling and/or connective tissue etc and are independent of muscle heme pigment effects.. The colour perception of the meat is essentially influenced by the more advanced of the two penetrating factors- light and oxygen. When oxygen penetration prevails, the meat surface appears to be completely covered by bright oxymyoglobin. Otherwise, if light penetrates the surface layer of meat beyond the oxymyoglobin the meat appears darker due to increased achromatic absorption due to the appearance of purplish myoglobin in the light path. Light absorption at 730nm differed significantly between age categories and muscle types at all time points measured. The regression models indicate that high 730nm absorption values at 0.5 hours post slaughter will contribute toward a low pH and muscle lightness values at 24 hours post slaughter.

5 VISNIR spectroscopy to measure pre-rigor muscle glycogen and lactate levels

5.1 Methodology

5.1.1 Animals

The same animals described in section 4 were used for this study.

5.1.2 Treatments

The same experimental design described in section 4 was used for this study.

5.1.3 Carcass & reference measurements

Individual carcass measurements recorded on the day of kill included carcass weight, and GR fat depth. Muscle pH and temperature profiles for all four muscles were recorded at 0.5, 3, 6 and 24 hours post slaughter. Muscle biopsies were collected from all four muscles at 0.5, 3 and 24 hours post slaughter. The samples were snap frozen in liquid N at collection and held at –80C until homogenised. A sub sample of 1g of muscle was homogenised in 10ml of 30mM HCL using the homogenising drill. Levels of glycogen were determined in the homogenate using enzymatic methods, following the hydrolysis of glycogen with amyloglucosidase (Dalrymple & Hamm 1973). The same homogenate was used to measure muscle lactate at the same time points nominated for the glycogen. After homogenisation lactate was determined in the muscle extract by the method described by Noll 1985. Muscle glycolytic potential (GP) was derived by the sum of the main compounds producing lactic acid post mortem and calculated according to the formula proposed by Monin and Sellier (1985)
GP= 2(glycogen) +(glucose) + (glucose 6-phosphate) +(lactate)
In this study the calculated GP excluded both glucose and glucose 6-phoshate. This decision was base on data from previously reported studies, which suggested that both glucose and glucose-6-phosphate were small and consistent contributors to GP (Josell et al 2000). Their exclusion substantially reduced laboratory assay costs

At 24 hours post slaughter objective meat colour (Hunter Lab L, a & b values) were recorded after a 30 minutes bloom for all muscles except the shin which was excluded due to its small size. VISNIR spectral reflectance measurements (380 to 2100nm) were collected on each muscle at the same nominated time points recorded for pH and temperature except 6 hours. The spectra were collected using an ASD Lab Spec Pro instrument. operating in reflectance mode and fitted with a fibre optic probe. The optical fibre probe was placed on the surface of the muscle and a setting of 15 scans per measurement site used with the probe orientated both parallel and perpendicular to muscle fibres.

5.1.4 Statistical analysis

All statistical analyses of the reference data were performed using GenStat Committee version 7.1 2003. Laboratory reference data for muscle glycogen, lactate and glycolytic potential data were analysed using the method of restricted maximum likelihood (REML). Age category, stick method, muscle type and sex were treated as fixed effects and week of kill, carcass and side of carcass as random effects. The predictive ability of muscle glycogen measured at 30 minutes on pH measured

at 24 hours was assessed by regression allowing for week of kill, age category, sex and muscle type. For this analysis pH measured at 24 hours was natural log transformed.

Spectral data was processed using Unscrambler v7.6 and WINISI v3.1 software. Spectra were converted to absorbance. Glycogen and glycolytic potential calibration models using principal least squares analyses (PLS) were calculated over the range 400 to 1900nm using every second data point i.e. 2nm data pitching. The spectra data was pre-treated using the SNV and detrend scatter correction functions. The PLS analyses were conducted using the 2nd derivatives of the spectral absorbance data calculated using a 16nm gap and 16nm smooth function. Spectral data under 400nm and over 1900nm was excluded from the analysis on the basis of there being a high level of noise in these areas of the spectrum.

Additional analysis of the glycogen and glycolytic potential calibration models was performed using the bootstrapping statistical technique. This technique is used to assess the performance of a calibration where the accuracy of the reference data is in question. Bootstrapping can provide a more representative estimate to be made of the true value of the laboratory reference measurement where a high repeatability in a single laboratory estimate is difficult to achieve. A high repeatability in the muscle glycogen and lactate reference measurements can be difficult to achieve because of non homogenous nature of muscle tissue. Bootstrapping is performed on the data to determine and estimate of the parameter which is then utilised as if it were the true value. It is not to be confused with cross-validation where the aim is to estimate the prediction error of a calibration in the absence of an independent validation set. In cross-validation, each sample is removed from the calibration in turn, a model developed using the remaining samples, and the removed sample predicted. The cycle is repeated until all samples have been removed and predicted. The calibration models are then averaged and the performance of the averaged model estimated by calculating the standard deviation of the differences between the reference value and the predicted values for the set of samples. In bootstrapping, each sample is removed, and a number of different models built from the remaining samples. Each model is used to estimate the removed sample, and the estimates averaged to give a new reference value. The calibration results from the 7 calibrations for each sample are average. This is an estimate of the true value and can be compared to the result obtained from a calibration developed using the full data set and the original reference data.

5.2 Results

5.2.1 Muscle glycogen, lactate and glycolytic potential reference data

Table 5.1 provides a statistical summary of muscle glycogen, lactate and calculated glycolytic values. The values are the pooled means of the four muscles sampled at 3 nominated time points post slaughter. The changes observed in their values with time post slaughter generally reflect the expected trend of a high initial muscle glycogen levels immediately post slaughter that progressively decline over time until rigour This represents the normal cellular process that occurs post slaughter where muscle glycogen is converted to lactic acid in an anaerobic environment.

Table 5.1 Means and standard deviations values for the glycogen, lactate and calculated glycolytic potential pooled for *M. longissimus thoracis et lumborum*, *M. supraspinatus M.semitendosis*, and *M. biceps brachi* muscles at 3 nominated time points post slaughter (n=282)

Carcass/ meat quality trait	Time post slaughter (hours)	Mean	Std deviation	Range
Glycogen	0.5	66.4	33.9	1.1 to 162.0
	3	57.4	29.4	11.3 to 140.4
	24	34.0	17.7	4.9 to 110.1
Lactate	0.5	42.7	11.5	15.1 to 75.5
	3	55.3	10.0	28.6 to 86.7
	24	80.0	14.8	44.9 to 111.4
Glycolytic potential	0.5	175.6	70.7	39.0 to 370.8
	3	170.3	56.9	81.1 to 333.0
	24	148.1	43.6	72.9 to 326.2
pH	0.5	6.66	0.21	6.04 to 6.99
	3	6.46	0.23	5.76 to 6.94
	24	5.84	0.22	5.47 to 6.70

5.2.2 Treatment effects on muscle glycogen, lactate and glycolytic potential levels

Considerable variation existed in muscle glycogen, lactates and calculated GP values. Muscle type had a significant influence ($P<0.001$) on muscle glycogen, lactate and calculated GP values at all time points (Tables 5.2, 5.3 & 5.4). Stick method and sex had no significant effect. Lambs had higher muscle glycogen levels compared to mutton at all three time points measured but the differences only approached significance at 0.5 hours ($p=0.07$) and 24 hours (0.09) post slaughter. Of the four muscles assessed the loin muscle had consistently the highest glycogen levels at 0.5, 3 and 24 hours post slaughter. Conversely, the shin muscle had the lowest glycogen levels at 0.5 and 3 hours post slaughter but by 24 hours post slaughter glycogen levels in the silverside muscle were the lowest of the four muscles. There was a significant interaction between age category and muscle type at 0.5 and 3 hours post slaughter and it approached significance ($P=0.06$) at 24 hours post slaughter.

The treatment effects reported for glycogen also had comparable effects on both muscle lactate and glycolytic potential levels. In addition muscle lactate levels were significantly higher ($P<0.05$) in ewes compared to wethers at 0.5 and 3 hours post slaughter but the differences attributed to sex were not significant by 24 hours post slaughter.

Table 5.2 Summary of the effects of age category, stick method, sex and primal cut on muscle glycogen at 30 minutes, 3 & 24 hours post slaughter

Treatment		No.	Glycogen 0.5 hrs	Glycogen 3hrs	Glycogen 24hrs
Age category	lamb mutton sed p	24 24	69.70 63.13 3.93 0.07	60.27 54.61 4.01 ns	36.77 31.29 2.85 0.09
Stick method	normal thoracic sed p	24 24	65.65 67.18 3.67 ns	56.83 58.05 3.75 ns	34.35 33.71 2.67 ns
Sex	ewe wether sed p	31 17	67.79 65.05 3.93 ns	59.43 55.46 4.01 ns	34.48 33.59 2.85 ns
Muscle	chuck tender loin shin silverside sed p	96 96 48 48	62.54 91.92 48.85 62.36 2.92 <0.001	52.10 82.78 35.70 59.20 2.76 <0.001	34.15 44.17 31.15 26.66 2.45 <0.001
Interactions			AM (<0.001) MSex (0.03)	AM (0.009) MSex (0.006)	AM (0.06) ASex (0.06)

Table 5.3 Summary of the effects of age category, stick method, sex and primal cut on muscle lactate at 30 minutes, 3 & 24 hours post slaughter

Treatment		No.	Lactate 0.5 hrs	Lactate 3hrs	Lactate 24hrs
Age category	lamb mutton sed p	24 24	41.47 44.0 1.19 0.013	55.19 55.37 1.43 ns	80.88 79.17 1.75 ns
Stick method	normal thoracic sed p	24 24	42.99 42.48 1.12 ns	55.17 55.38 1.35 ns	80.82 79.24 1.64 ns
Sex	ewe wether sed p	31 17	44.50 40.97 1.19 0.02	57.07 53.49 1.43 0.03	80.81 79.24 1.75 ns
Muscle	chuck tender loin shin silverside sed p	96 96 48 48	32.00 38.86 53.23 46.85 0.96 <0.001	46.00 51.47 61.37 62.27 1.25 <0.001	69.69 92.78 73.86 83.79 2.52 <0.001
Interactions			AM (0.06) MSex (0.04)		AMSex (0.01)

Table 5.4 Summary of the effects of age category, stick method, sex and primal cut on muscle glycolytic potential (GP) at 30 minutes, 3 & 24 hours post slaughter

Treatment		No.	GP 0.5 hrs	GP 3hrs	GP 24hrs
Age category	lamb mutton sed p	24 24	181.0 170.3 8.0 ns	175.9 164.7 8.0 ns	154.4 141.8 6.2 ns
Stick method	normal thoracic sed p	24 24	174.3 177.0 7.5 ns	168.9 171.6 7.5 ns	149.5 146.7 5.8 ns
Sex	ewe wether sed p	31 17	180.1 171.1 8.0 ns	176.1 164.4 8.0 ns	149.8 146.4 6.2 ns
Muscle	chuck tender loin shin silverside sed p	96 96 48 48	157.1 222.7 151.1 171.7 5.8 <0.001	150.1 217.0 133.0 181.0 5.3 <0.001	138.0 181.1 136.2 137.1 5.3 <0.001
Interactions			AM (<0.001) MSex (0.04)	AM(0.002) MSex (0.004) AMSex (0.003)	AM(0.08) ASex (0.03)

5.2.3 Relationship between muscle glycogen and 24 hour pH

Regression modelling of muscle glycogen measured at 0.5 hours post slaughter with 24 hour pH transformed to the natural log indicates a highly predictive relationship (P<0.001) when week of kill, age category, sex and muscle type all significant factors are included in the model. The model described below accounted for 81.2% of the variation in 24 hour pH with a standard error of 0.015 units.

Response variate: log_pH_24hr
Fitted terms: Constant + Week + Gly_0_5 + Muscle + Gly_0_5.Muscle + Age category + Sex + Age_category.Sex

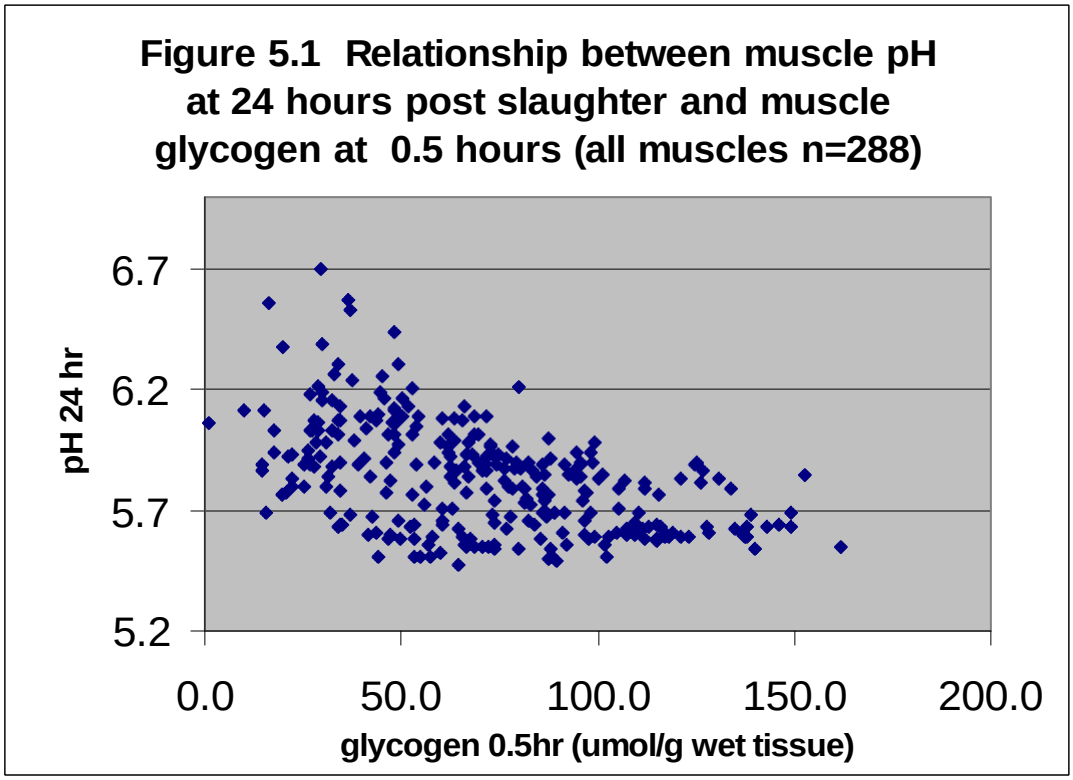
The comparable model for muscle glycolytic potential accounted for 76.9% of the variation. From an animal modelling perspective this is high when compared to other animal based models of biological systems. However as Table 5.5 demonstrates if the intended use of the models is to sort individual

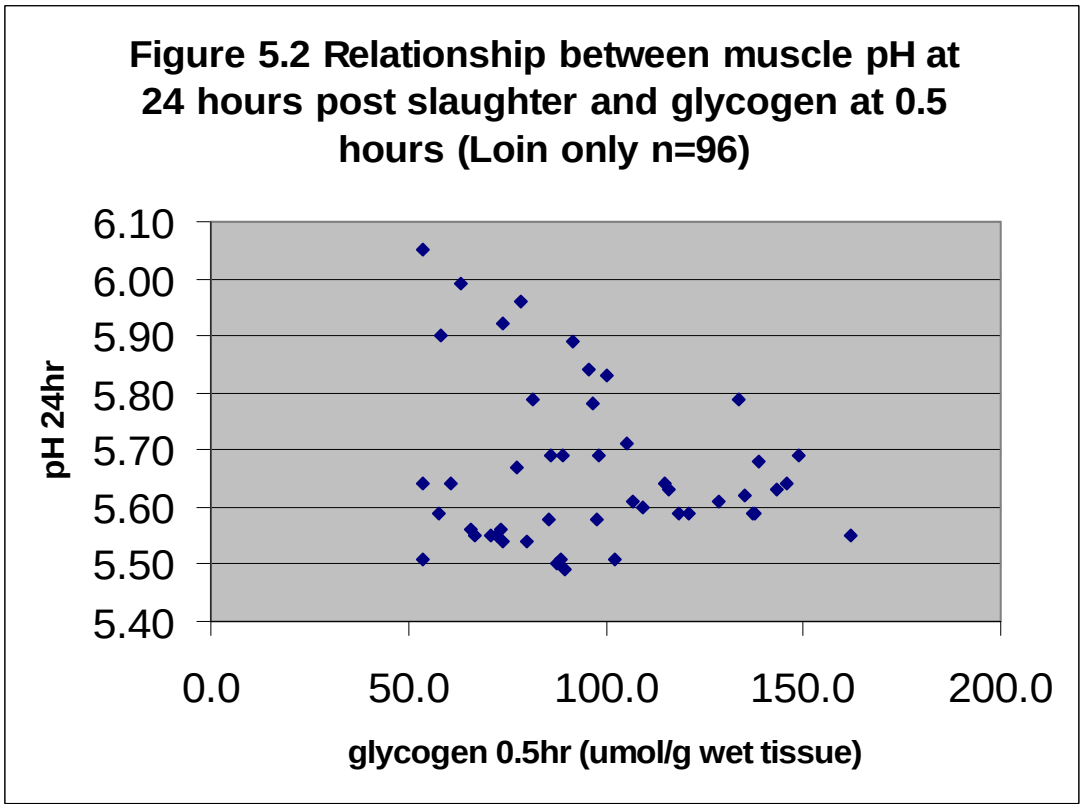
carcasses and/or carcasses on predicted pH at 24 hour post slaughter then the model(s) are not sufficiently accurate based on the 95% confidence limit to achieve the desired commercial outcome. The 95% confidence interval is calculated as the mean prediction ± 2× standard error

Table 5.5 Glycogen model prediction and 95% confidence interval for muscle pH at 24 hours using muscle glycogen measured at 0.5 hours post slaughter

Age category	Sex	Muscle	Muscle glycogen @ 0.5hrs post slaughter	24 hour pH		
				Low Confidence limit	Predicted	High Confidence Limit
Lamb	ewe	Chuck tender	60	5.75	5.93	6.12
			80	5.72	5.89	6.08
			100	5.68	5.85	6.04
Lamb	ewe	Loin	60	5.54	5.72	5.90
			80	5.52	5.69	5.87
			100	5.49	5.66	5.84

Figure 5.1 illustrates the relationship between muscle pH at 24 hours post slaughter and glycogen measured at 0.5 hours for all four muscles. Figure 5.2 illustrates the same relationship for the loin muscle only.





5.2.4 VISNIR calibration models

Spectral and muscle glycogen, lactate and glycolytic potential reference data were analysed to determine the applicability of VISNIR spectroscopy to measure muscle glycogen and/or glycolytic potential status. PCA indicated that there was no difference between the spectra when grouped according to any of the experimental treatments i.e. muscle type, sex, age category, stick method or probe position. Therefore, different spectral models are not required for different muscles, age categories etc. Calibration models were developed using PLS are summarised in Tables 5.6, 5.7 & 5.8.

Glycogen

PLS analyses of the data indicates the best calibration models developed for muscle glycogen were attained when spectra was collected at 0.5 hours post slaughter with the fibre optic probe placed perpendicular to the cut surface of the loin muscle (Table 3.6). The correlation coefficient for the prediction model was r=0.77 with a SEP of 21.55 µmoles/g for spectra collected at 0.5 hours post slaughter with the probe held in the perpendicular position to the cut surface. No outliers had been removed at this stage of the analysis.

Table 5.6 Calibration model estimates for muscle glycogen using VISNIR spectra collected at 0.5, 3 and 24 hours post slaughter(n=287)

Hours post slaughter Scan / Reference measurement	Probe orientation (no. scanned)	Model estimates R	SEP	SEP/SD	PLS Factors
0.5/ 0.5	perp*	0.77	21.55	0.63	4
	parallel	0.72	23.67	0.70	4
3/ 3	perp	0.72	20.67	0.70	2
	parallel	0.71	20.63	0.70	2
24/ 24	perp	0.60	14.20	0.80	2
	parallel	0.55	14.77	0.83	2

* Perpendicular

Lactate The calibration models generated for lactate were also highest at 0.5 hours post slaughter. A correlation co-efficient of r= 0.76 with a SEP of 7.57 umoles/g was obtained for lactate at 0.5 hours post slaughter with the probe orientation perpendicular to the cut surface (Table 3.7). This is surprising considering that lactate values were at their lowest point of the 3 times points measured.

Table 5.7 Calibration model estimates for muscle lactate using VISNIR spectra collected at 0.5, 3 and 24 hours post slaughter (n=287)

Time post slaughter	Probe orientation (no. scanned)	Model estimates R	SEP	SEP/SD	PLS Factors
0.5	Perp	0.76	7.57	0.66	5
	Parallel	0.75	7.52	0.65	2
3	Perp	0.55	8.38	0.84	3
	Parallel	0.43	9.02	0.90	1
24	Perp	0.65	11.23	0.76	3
	Parallel	0.68	10.86	0.73	4

Glycolytic potential

Calibration models of comparable accuracy to muscle glycogen were obtained for glycolytic potential particularly in the pre-rigour phase. A correlation co-efficient of r= 0.82 with a SEP of 39.42 umoles/g was obtained for glycolytic potential at 0.5 hours post slaughter with the probe orientation perpendicular to the cut surface (Table 5.8). This result directly reflects comparable results obtained for both glycogen and lactates and reflects the fact that GP was a calculated value from both glycogen and lactate.

Table 5.8 Calibration model estimates for muscle glycolytic potential using VISNIR spectra collected at 0.5, 3 and 24 hours post slaughter (n=287)

Time post slaughter	Probe orientation (no. scanned)	Model estimates			
		R	SEP	SEP/SD	PLS Factors
0.5	Perp	0.82	39.42	0.56	4
	Parallel	0.77	44.44	0.63	3
3	Perp	0.72	39.30	0.69	2
	Parallel	0.69	41.03	0.72	2
24	Perp	0.65	33.35	0.76	2
	Parallel	0.62	34.21	0.78	2

5.2.5 VISNIR model classification accuracy

Table 5.9 summarises the classification accuracy of the VISNIR calibrations models to measure the glycogen and glycolytic potential status of carcass muscles using spectra collected at 0.5 hours post slaughter. The calibrations models were calculated with and without the bootstrap method. Their accuracy was assessed by assessing their ability to sort muscles into one of two categories either above or below a nominated threshold value. Glycogen calibration models used a classification threshold value of 70 µmoles/g of glycogen at 0.5 hours post slaughter. The value was selected on the basis that for this dataset it represented a threshold value associated with high muscle pH at 24 hours post slaughter. The comparable threshold value used for glycolytic potential calibration models was 180µmoles/g.

Table 5.9 Classification accuracy of VISNIR calibration models to predict whether muscle glycogen and glycolytic potential values at 0.5 hours post slaughter are above or below a nominated threshold value associated with high pH

Calibration model	Bootstrap method used	Model statistical estimates		
		r^2	SECV	% Correctly classified
Glycogen 0.5 Hour	No	0.44	24.6	78
	Yes	0.87	8.4	81
Glycolytic potential 0.5 hour	No	0.49	49.2	91
	Yes	0.86	20.1	91
Milestone target				90

Figures 5.3, 5.4 & 5.5 illustrate the predictive classification accuracies of the calibration models developed using either single laboratory reference measurements or bootstrap estimates. Whilst the

bootstrap method substantially improves the r^2 and SECV model estimates of the calibration models it only has a relatively minor effect on overall classification accuracies which approach or achieve the stated milestone of 90%. It should be noted that the bootstrap calibration models are probably an over estimate as it relies on the assumption that there is a high degree of linkage between the spectral and analytical data. To confirm this requires further testing involving replicate lab analysis data to confirm the results.

Figure 5.3 Bootstrap estimates and single estimates vs. the NIR cross-validation error for glycogen at 0.5 hours post slaughter.

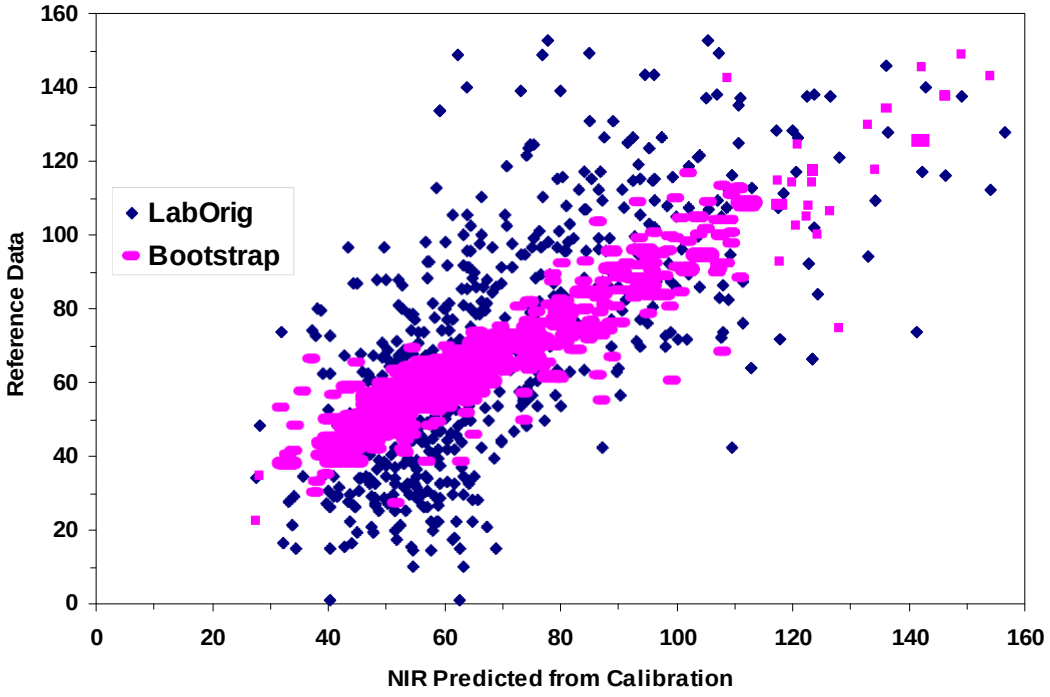


Figure 5.4 Prediction of Glycogen at 30 minutes showing the missclassification rate

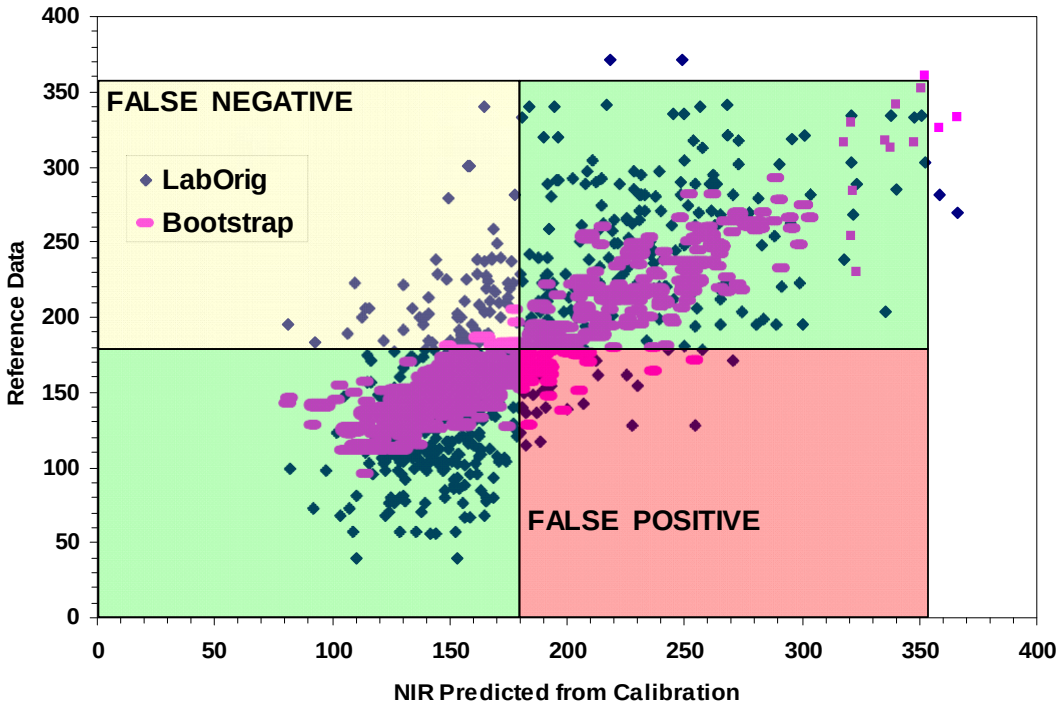
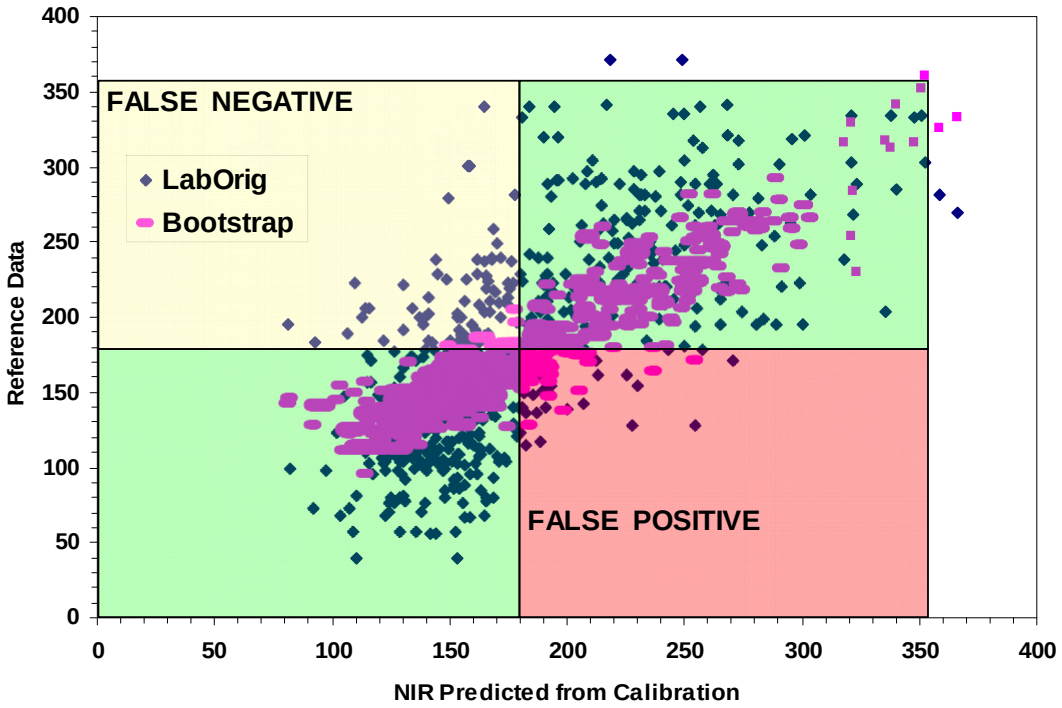


Figure 5.5 Prediction of Glycolytic Potential at 30 minutes showing the misclassification rate.



5.3 Discussion

The primary objective of this study was to develop an application using VISNIR technology that could reliably sort carcasses with 90% accuracy on their ultimate pH measured at 24 hours post slaughter. To achieve this requires a two stage process. The first stage requires evidence that glycogen or glycolytic potential measured immediately post slaughter can be used to predict muscle pH some 24 hours later. The second stage requires evidence that VISNIR technology can be calibrated and used to measure in vivo muscle glycogen levels on the slaughter floor

The results confirm that a strong predictive relationship existed between laboratory reference muscle glycogen levels at slaughter and muscle ultimate pH at 24 hours post slaughter that accounts for over 80% of the variation. However when used to predict 24 hour pH in individual carcasses with a 95% confidence limit the range in predicted muscle pH values at 24 hours was too high to reliably sort muscles on their predicted ultimate pH from a commercial industry perspective. It is considered this is largely a reflection of the inherent measurement error associated with both the laboratory reference measurement of muscle glycogen and instrumental error in measuring pH.

VISNIR calibration models developed for muscle glycogen indicate that the spectra can be used to reliably sort muscles on their glycogen level.. The results support the use of VISNIR spectroscopy to accurately measure and classify carcasses on their pre-rigor muscle glycogen and glycolytic potential levels on the slaughter floor. Classification accuracies of 78% for glycogen and 91% for glycolytic potential were achieved between the VISNIR predicted and laboratory reference value using calibration models developed using standard cross validation procedures. Again some further improvement to both the VISNIR calibration models could be achieved by reducing the inherent error that exists in the laboratory reference values of muscle glycogen and glycolytic potential.

With further refinement to both the VISNIR glycogen prediction model(s) and the underpinning muscle glycogen/pH prediction model it appears feasible that VISNIR technology could be used by processors to sort carcasses and/or individual muscles on line on their predicted ultimate pH as early as 30 minutes post slaughter with 90% accuracy. The technology has immediate commercial application for hot boning plants which need a reliable real time method to identify and exclude high pH primal cuts from their graded chilled products destined for high value domestic and export markets because of the reduced shelf life and inherent colour problems of high pH meat.

It also has commercial relevance to conventional processing plants who are still confronted with the downgrading costs of high pH meat which in beef plants represents around 8 to 10% of their production. To date there has been no technology available that can operate at chain speeds to measure muscle glycogen levels to provide real time feedback to integrated supply chains that would enable them to better diagnose and strategically manage the problem.

6 Accuracy of an accelerated freeze/thaw method to induce and measure ultimate muscle pH

6.1 Methodology

6.1.1 Animals

The same animals described in sections 4 & 5 were used for this study.

6.1.2 Treatments

The same experimental design described in sections 4 & 5 were used for this study.

6.1.3 Muscle pH measurements

Muscle samples (g) were collected from two muscles namely the loin (*M. longissimus thoracis et lumborum)* and chuck tender (*M. supraspinatus)* muscles at 0.5 hours post slaughter. The samples were snap frozen in liquid N at collection and held at –80C for 20 minutes then thawed in warm water. Muscle pH was measured for all the rapid frozen/ thawed samples when they attained a thawed temperature of 4 to 5 ˚C which was approximately 1 hour post slaughter. Muscle pH and temperature were also measured at 24 hours post slaughter on the original muscles that had remained intact in the carcass since slaughter. Muscle temperature at the 24 hour measurement time points was around the 4 to 5˚C temperature of the comparable to freeze/ thaw samples.

6.1.4 Statistical analysis

The ability of the freeze thaw technique at 30 minutes post slaughter to predict muscle pH measured at 24 hours was assessed by regression allowing for week of kill, age category, sex and muscle type. Genstat version 7.1 (2003) was used for the analyses.

6.2 Results

Muscle pH measured using the rapid freeze/thaw technique within 30 minutes post slaughter did not provide an accurate predictor of muscle pH measured conventionally at 24 hours post slaughter. Regression analysis of the freeze/thaw pH with the conventional 24 hour pH for the chuck tender muscle was not significant. A comparable regression analysis for the loin muscle was significant ($p<0.05$) but accounted for only 6.5% of the variation. Correlation coefficients between the rapid freeze/thaw and conventional pH measurements for either the chuck tender and loin muscles were both low (Table 6.1)

Table 6.1 Correlations coefficients (r^2) between a rapid freeze/thaw method and conventional muscle pH measurement method taken at 24 hours post slaughter for the chuck tender and loin muscles

Muscle	No. of samples	r^2 Freeze/thaw & conventional 24 hour pH
Chuck tender		0.02
Loin		0.09

Figures 6.1 and 6.2 further illustrate the limited predictive relationship between the rapid freeze/thaw pH method and conventional method to measure muscle pH at 24 hours post slaughter.

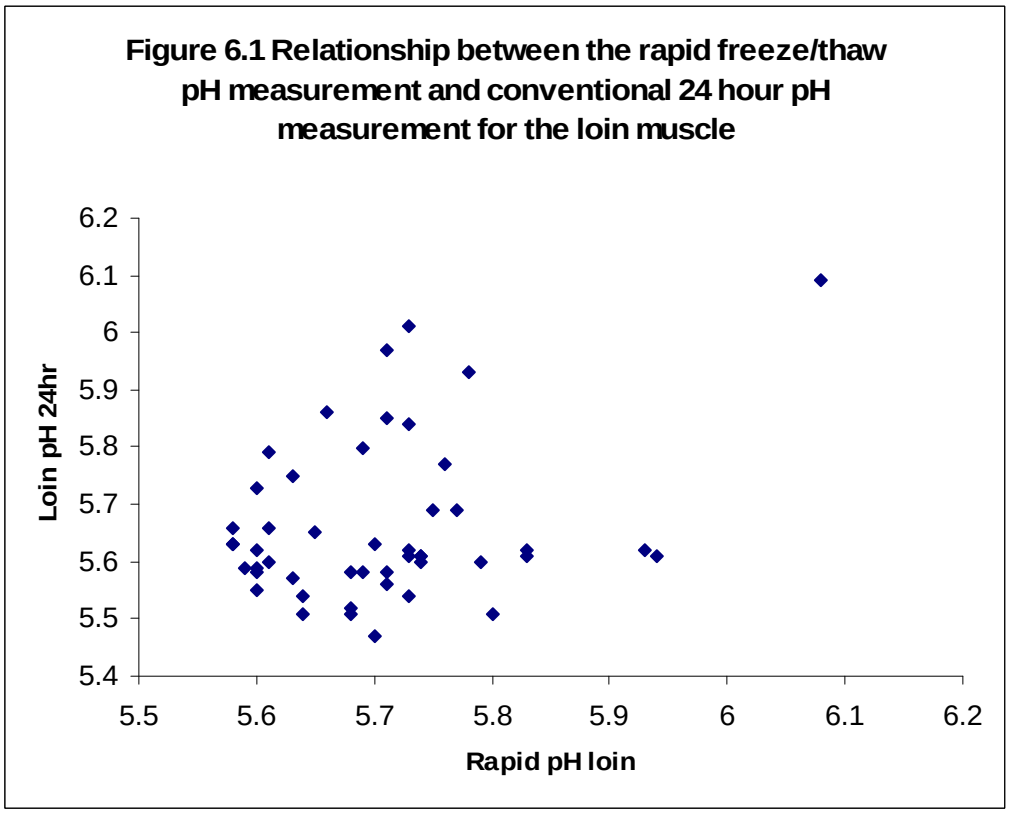
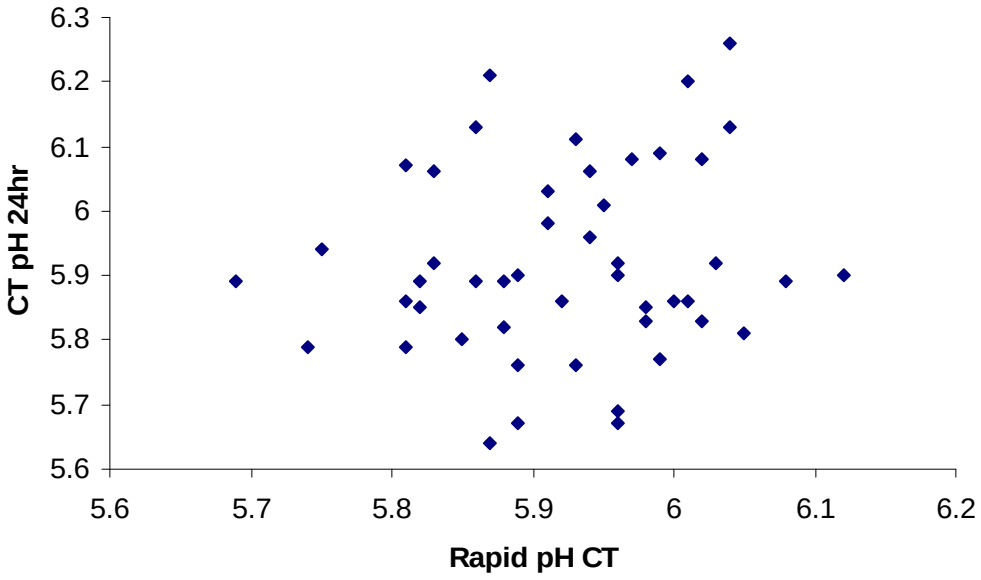


Figure 6.2 Relationship between the rapid freeze/thaw pH measurement and the conventional 24 hour pH measurement for the chuck tender muscle (CT)



6.3 Discussion

The results of this study found that the accelerated freeze/thaw technique does not appear to provide a reliable method of measuring pH at 24 hours in muscles conventionally chilled.
The study did find that rapid freezing and thawing did greatly accelerate a decline in loin and chuck tender muscle pH but there was a very low correlation between the actual pH values measured in the rapid freeze/thaw samples with comparable samples measured conventionally at 24 hours post slaughter. The explanation offered for the accelerated decline in muscle pH from rapid freezing and thawing is attributed to a rupturing of the sarcoplasmic reticulum as well as the sarcolemma associated with freezing. This releases free calcium to stimulate ATP-consumption. Also at temperatures below 10°C the Ca-ATP pump in the sarcoplasmic reticulum looses its capacity which further stimulates ATP consumption. Rigour is attained when all ATP is consumed. However, it appears that the cascade of chemical events that are triggered and accelerated from freeze/ thaw technique differ substantially from those that occur in muscle undergoing conventional chilling As a consequence the muscle pH end point at the conclusion of the two processes differs significantly.

7 New generation pH/ temperature data loggers

7.1 Background

Compliance to an optimal pH/ temperature window at rigour is one of the conditions required for eligibility for grading under the MSA beef grading program. As a consequence this has substantially increased the commercial meat industries requirement and interest in measuring pH and temperature decline in pre rigor muscle. However the task of measuring the pH component of the pH/temperature decline profile in muscle still remains a manual task using pH measurement technology that has changed little in the past 50 years. The lack of technical development in pH probes is surprising given the quantum leap in electronic technology that has come onto the market in the past decade. This project aimed to review technology options to automate the measurement of muscle pH and if suitable technologies were identified validate their ability to record the time, temperature and pH relationship of pre rigour muscle.

7.2 Results

An international search failed to find any suitable low cost commercially available pH probes that could be used by the meat industry to log muscle pH in multiple carcasses over time comparable to that currently available for logging muscle temperature. Further discussion with some specialist electronic companies indicated that the technology to build an integrated probe to log both muscle pH and temperature decline over time was available but the market for such a product was considered too small and commercially unviable. Consequently given the non availability of any suitable “off the shelf” pH data loggers and a lack of commercial interest in developing such a pH probe no further work was conducted on this milestone.

8 Success in Achieving Objectives -

Objective 1: Assess the predictive accuracy of VISNIR spectroscopy to measure the objective tenderness and intra-muscular fat content of lamb
This objective was successfully achieved. The results support the application of VISNIR technology as an alternative objective measurement method to measure lamb on objective colour (L,a & b), ultimate pH and intramuscular fat content at 24 hours posts slaughter. Moderate PLS calibration model estimates were obtained for these meat quality traits using the spectral data acquired at 24 hours post slaughter. Attempting to predict objective colour and intra muscular fat levels using spectra acquired on pre-rigour lamb at 30 minutes post slaughter was much less reliable and consequently not recommended. The key advantage of using VISNIR technology to measure a suite of meat quality traits including objective colour, ultimate pH and intramuscular fat is its ability to simultaneously measure all traits with the one instrument at extremely rapid speeds of less than 5 seconds per carcass. Furthurmore the process can be automated based on related VISNIR applications already implemented by other sectors of the food industry.

The results do not support the application of VISNIR technology to grade lamb on objective tenderness using loin muscle shear force or compression values. Low PLS calibration model estimates were obtained for these traits using the spectral data acquired at 1 and 5 days post slaughter. As a consequence the VISNIR predictions of objective tenderness were not sufficiently accurate to ensure consumer confidence in a VISNIR objective tenderness classification system.

Objective 2: Investigate the predictive accuracy of VISNIR spectroscopy to measure muscle pigment levels and their effect on ultimate pH and meat colour
This objective was successfully achieved. The results demonstrate that VISNIR technology could be used to measure total muscle heme pigment levels based on absorption values at nominated isobestic wavelengths and these have a significant influence on muscle redness but not pH. Both varied significantly between muscle groups. There was no significant relationship between muscle heme pigment levels and stick method (bleeding efficiency) in this study. In addition VISNIR could also be used to measure non pigment related factors that influence both muscle colour lightness values and ultimate pH based on spectral absorption values at 730nm

Objective 3: Validate a method for using VISNIR spectroscopy to accurately measure and classify carcasses on their pre-rigor muscle glycogen and lactate levels on the slaughter floor with 90% accuracy
This objective was successfully achieved. VISNIR calibration models developed for muscle glycogen indicate that the spectra can be used to reliably sort muscles on their glycogen level. The results support the use of VISNIR spectroscopy to accurately measure and classify carcasses on their pre-rigor muscle glycogen and glycolytic potential levels on the slaughter floor. Classification accuracies of 78% for glycogen and 91% for glycolytic potential were achieved between the VISNIR predicted and laboratory reference value using calibration models developed using standard cross validation procedures. Some further improvement to both the VISNIR calibration models could be achieved by reducing the inherent error that exists in the laboratory reference values of muscle glycogen and glycolytic potential.
The results also confirm that a strong predictive relationship existed between laboratory reference muscle glycogen levels at slaughter and muscle ultimate pH at 24 hours post slaughter that accounts for over 80% of the variation. However when applied to individual carcasses with a 95% confidence limits the predicted range in muscle pH values at 24 hours was too high to reliably sort

muscles on their predicted ultimate pH from a commercial industry perspective. It is considered this is largely a reflection of the inherent measurement error associated with both the laboratory reference measurement of muscle glycogen and instrumental error in measuring pH.

Objective 4: Develop and assess the accuracy of an accelerated freeze/thaw method to induce and measure ultimate muscle pH within 30 minutes of slaughter
This objective was successfully achieved. However based on the results of this study the accelerated freeze/thaw technique does not appear to provide a reliable method of measuring pH at 24 hours in muscles conventionally chilled. The muscle pH end point at the conclusion of the two processes differed significantly for both the loin and chuck tender muscles.

Objective 5: Review, develop and validate a new generation low cost data logger that can record time and temperature of muscle(s) at the onset of rigor
This objective was not achieved. After an international search it was identified that no suitable “off the shelf” new generation pH data loggers with the required specifications were available to test. Also whilst the fundamental technology was already available in other electronic products there was a lack of commercial interest amongst specialist electronic companies to develop a new generation pH probe for the meat industry. Consequently no further work was conducted on this objective.

9 Impact on Meat and Livestock Industry –

The results provide strong support for the continued development of VISNIR spectroscopy to accurately measure and classify carcasses on a number of commercially important traits of red meat. These include the ability to measure objective colour, intramuscular fat and pH at 24 hours post slaughter, muscle glycogen and/or glycolytic potential and muscle heme pigment levels at 30 minutes post slaughter. All these traits influence the appearance and palatability of red meat which are factors known to influence consumers purchasing decisions. In this study VISNIR spectroscopy did not provide a reliable prediction of objective tenderness. Currently most of these traits are not routinely objectively measured by the commercial red meat processing industry largely because the current measurement technology, where available, is manual and must be done on the cold carcass to reliably grade carcasses on pH or colour defects. The results of this project suggest VISNIR technology can overcome some of these limitations to provide the red meat industry with an alternative multi trait measurement technology to better manage and reduce the direct costs associated with carcasses downgraded for colour or pH related defects. Industry surveys estimate the incidence of such defects to be between 8 to 10% of the national kill.

10 Conclusions and Recommendations

Collectively the results support the concept that VISNIR technology can be developed to rapidly, objectively and cost effectively measure and grade carcasses on a suite of meat quality traits. The meat quality traits most suited to being measured by VISNIR are those controlled by the chemical status of muscle. Whilst most processors do not recognise it the conversion of muscle to meat is a complex chemical process which they control through their processing and chilling management practices. If coupled with MLA’s substantial investment in livestock traceability and automated processing systems VISNIR technology has the potential to add considerable value to lamb and beef supply chains. It could achieve this by providing supply chain participants with objective feedback on meat quality that has greater diagnostic benefits for supply chains to enhance compliance to preferred end user specifications. Other food industries that now regularly use

VISNIR technology have done so on the basis of if you can measure it you can manage it. This philosophy has equal relevance to the meat industry.

For the red meat industry to get the best return from their investment in the further development of meat measurement applications using VISNIR technology it is critical it be done following a well executed business plan. Some of the essential issues that need to be clearly articulated in the business plan include:

- the priority of the measurement technologies sought by industry and under what operating conditions (hot or cold carcasses, operating conditions such as chain speeds etc)
- availability of the expertise & skills required to form a multidisciplinary team that can deliver on the project outcomes given that the majority of NIR expertise has been developed and resides outside of the meat industry
- early selection of the preferred instrument manufacturer and specifications of the spectrometer. Majors costs are associated with the development of calibration models. Changing instruments mid stream can change the optical performance of the spectrometer substantially and could make any previous investment in the calibration models obsolete.
- Early selection of the preferred instrument manufacturer will also facilitate a cost effective and seamless interface between research and commercialisation

The red meat industry have long sought a reliable and accurate objective method to grade meat on tenderness. Unfortunately this study found that VISNIR technology had limited ability to measure the objective tenderness (shear force & compression values) of lamb. The most likely explanation is that whilst VISNIR technology is well recognised for its ability to measuring one or more unique chemical characteristics it does have particular limitations for measuring the mechanical properties of a product such as meat unless they are largely dictated by one or more distinct chemical bonds. VISNIR may yet still have the potential to predict muscle tenderness if there are one or more of the specific chemical bonds in the muscle matrix that have a large influence on tenderness. If such biomarkers do exist then VISNIR technology could provide industry with an objective method to grade meat on tenderness.

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