final report

Project code: A.SCT.0059

Prepared by: Greenleaf Enterprises

Date submitted: April 2010

Comparison between hot and cold CT analysis of bovine muscle

CT OCM Project – risk 3 (hot marbelling) further mitigation

Executive summary

The following trial work was conducted as part of an ongoing investigation by Meat and Livestock Australia (MLA) into the viability of using CT scanning as a measurement technology for objective measurement of bovine muscle and whole carcasses. Previous studies have assessed the various benefits of CT scanning for bovine carcass measurements, and have indicated a viable economic return on investment, provided the function of technology is proven.

The main objectives of the following trial work was firstly to establish a suitable measurement methodology which would allow for accurate comparisons between hot and cold scans, and secondly to assess if there was any difference in the CT measurement results, particularly of fat content, between hot and cold carcases.

The actual trial design consisted of two muscles primals, Strip Loin and Cube Roll. These primals were harvested hot from 5 different carcasses and divided into 5 replicate slices within each muscle and measured at both 2hrs post mortem (pre-rigor), and also again after a minimum of 24hrs chill at 5°C.

Establishment of the methodology included both the data capture process and the data analysis method. During the course of the trial work a suitable measurement technique was established for capturing the data, and several analysis methods were explored.

Perhaps most notably a difference was observed in the Hounsfield units of the peak for fat between hot and cold primals. Strip loin primals showed an increase of 51.5 HU from -111 (HU) at hot to -59.5 (HU). At a broad level further analysis was conducted using a thresholding analysis method to determine the total number of voxels within the fat range vs. total number of voxels outside the fat range. A significant correlation was observed between the predicted fat content of whole primals between hot and cold measurements (R^2 0.99).

This observation is important as it suggests the ability to determine fat content in either hot or cold primals using CT scanning is the same.

A slight difference in density of muscle was observed between hot and cold scans, evidenced by a shift in the location of the muscle peak on the histogram. Again this trend was significant across all primals types showing an increase of 8 (HU) from hot to cold scans.

Contents

1   Background information

MLA has been investigating whether CT scanning could be used in abattoirs to scan hot and or cold carcases to obtain useful objective measures that will enhance carcase value or reduce operational costs of processing.

Previous cost/benefit studies indicate that acceptable returns on equipment could be obtained if the benefits reported were able to be measured. One hurdle in progressing with development is identifying whether scanning of hot carcases gives measures that correlate with the same measurements in cold carcases.

The key objective of this project is to determine whether the physiology of muscle and fat behaves "similarly" between hot and cold CT scans of the same muscle. If CT scanning can be used to scan cold marbling but cannot be used to scan hot marbling because the density of hot marbling blends with hot muscle and cannot be identified then further research may need to be reconsidered.

This project is not expected to produce the final correlations on converting from Hot to Cold as a large number of variables would still be required to change if development of a commercial system were to proceed. Some of these variables that would impact on final correlations include type of scanner (Cone beam or helical), CT scan power settings, focal settings and pixel size, number of slices taken, length of scan time and so on.

The purpose of the following report is to quantify the risk associated with scanning hot versus scanning cold and whether that risk is large enough to prevent further development of the CT scanning capability.

2   Objectives

1. Methodology a) To establish a methodology for scanning hot and cold bovine muscle with the ability to compare between the same intramuscular region for both scans. To compare the difference in Hounsfield values between hot and cold bovine cuts.
b) To assess several data analysis techniques for predicting content of intra muscular fat and extra muscular fat (subcutaneous fat and seam fat) in bovine muscles.
2. To determine the impact of hot and cold measurement of bovine muscle properties on CT Hounsfield units.

3   Methods

Trial Design
The trial design included two primal types Strip Loin (M. Longissimus Dorsi, Aus-Meat product code 2140) and Cube Roll (M. *longissimus* dorsi Aus-Meat Product code 2244) collected from 5 different carcasses on 3 different kill dates. Carcass information is shown in Table 1.

Table 1: Body information contained

Trial Body #	Kill Date	Carcass weight	Fat Depth	Gender	Teeth	Kill Time	Aus-Meat Marble	Grain Fed
Body 1	15/12/2009							
Body 2	29/01/10	181	6	F	8	09:01:00	0	NO
Body 3	29/01/10	223	24	F	0	08:59:00	0	YES
Body 4	03/02/10	244	6	F	8	10:04:00	0	NO
Body 5	03/02/10	345.6	10	M	4	10:04:00	0	YES

3.1   Sampling Procedure

Primals were removed from the carcass 35 min post mortem (Figure 1) and further divided into between 5 and 10 different measurement slices. Three methods were used to identify measurement units within the primals. For strip loin 1 and cube roll 1 a freehand grid of 100mm x 40mm was marked using bamboo skewers. Measurement units within primals 2 and 3 were marked using a fixed Perspex template to insert bamboo marker points. Primals removed from carcasses 4 and 5 were placed into plastic containers with the bamboo skewers inserted through the top and bottom of the container to stop movement of markers as primals went into rigor (Figure 2). Figure 8 and Figure 9 show CT images of the difference between non-fixed primals during rigor and primals fixed prior rigor.

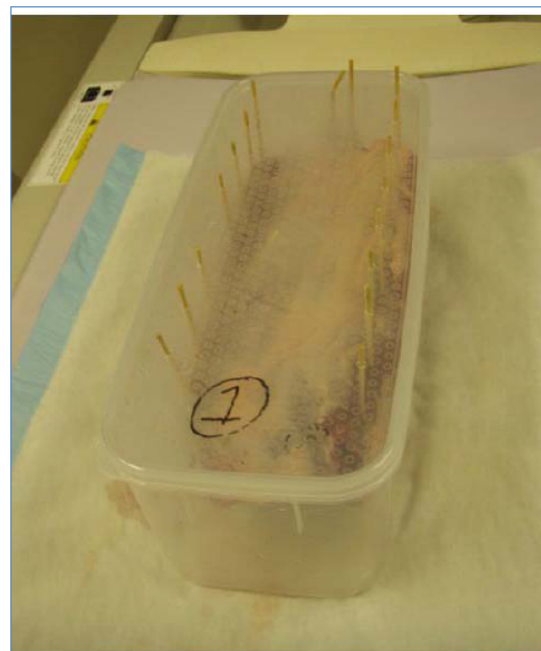Figure 1: Hot Boning Strip Loin and Cube Roll primals from carcass # 4

Figure 2: Cube Roll with 7 measurement slices marked 35mm x 100mm

Table 2 shows a total 71 measurement slices within the experimental data.

Table 2: Trial design

Carcass Number	M Type	# Slices	M Type	# Slices
1	Strip Loin	10	Cube Roll	5
2	Strip Loin	7	Cube Roll	7
3	Strip Loin	7	Cube Roll	7
4	Strip Loin	7	Cube Roll	7
5	Strip Loin	7	Cube Roll	7
Number of slices		38		33
Total number for trial			71	

3.2   CT Measurements

CT scans were conducted by Queensland X-ray at the Beenleigh clinic (1.7 km from processing plant) using a Toshiba Aquilion 16 slice CT system. Table 3 shows the CT settings used for both hot and cold measurements.

Table 3: CT settings used for generation of images

Parameter description	Setting
Scan Type	Helical
Energy or penetrating power (strength of the photons in the x-ray)	120kVp (photon (kilo Volts peak) none should be over 120
Amount of Photons (dose) amount of x-rays produced	300 mA
Rotation time	3 second
Number of slices per rotation	16
Slice Width	0.5mm
Window width*	200 HU centred on 40HU

*(selection of Hounsfield units used to make up the image, given that there are 4000 densities recognized by the CT need to set a range) (this means the measurement window ranged from (-60 to +160) with white being applied to anything above this range, and black to anything below, and 16 level grey scale applied to everything in-between.

Figure 3 shows a cube roll primal ready for CT measurement, and Figure 4 shows a CT image of a strip loin primal, the container used to hold the primal, and the marker points in 120 mm x 35 mm grid used to identify measurement slices.

Figure 3: Toshiba Aquilion 16 slice CT system with cold cube roll ready for measurement
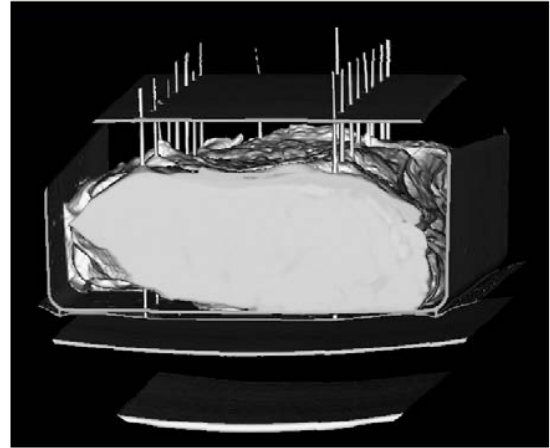

Figure 4: CT image of primal, container and marker points

Hot Carcass CT measurements were obtained immediately post slaughter (within 2 hrs). Primals were then placed in the chiller for 24 – 48 hrs and "Cold" CT measurements were taken.

3.3   Physical Sample Measurements

After cold CT measurements were taken primals were sliced according to the markers for each measurement unit. The area outside of the bamboo markers was removed and also any extra muscular fat occurring on the surface of the muscle (Figure 5 and Figure 6). Individual muscle samples were then sent to Symbio Alliance for chemical analysis.

Figure 5: Slice removed from primal
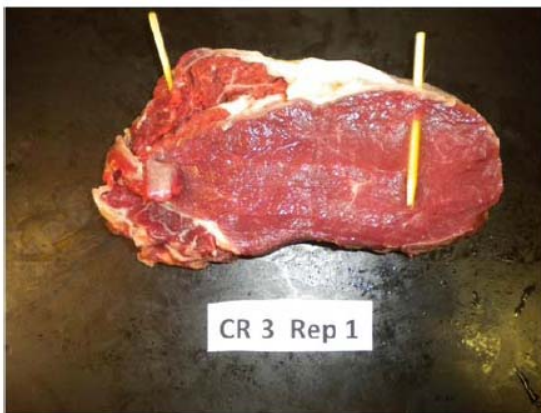

Figure 6: Muscle section from slice used for chemical fat analysis

Chiller assessments were conducted on both cube roll and strip loin from carcases 4 and 5. Parameters assessed included meat colour, fat colour, and marbling grade. Primals were sliced and allowed to bloom for a minimum of 30 minutes and assessments were conducted on each of 7 replicates in each primal (see Figure 7).

Figure 7: Strip Loins 4 & 5, and Cube Roll 4 & 5 cut and exposed to oxygen for 40min prior to certified Aus-meat Chiller assessment of meat colour, fat colour and visual marbling grade.

4   Data Analysis

Images were analysed using the software Analyze Direct 9.0. Dicom images from the CT scan were loaded into the software, rescaled to a cubic structure and rendered into a virtual 3D representation.
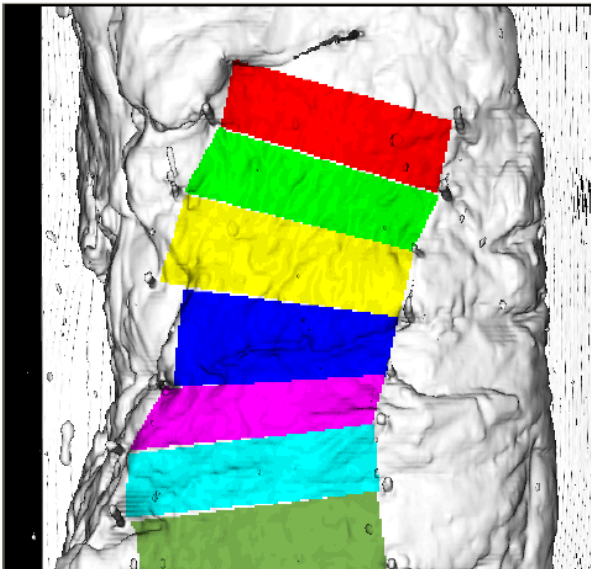

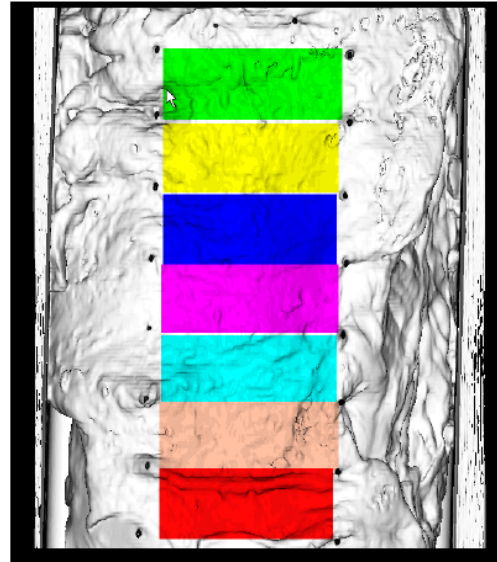Figure 8: Strip Loin 1 cold measurement (prior to primals being fix before rigor)

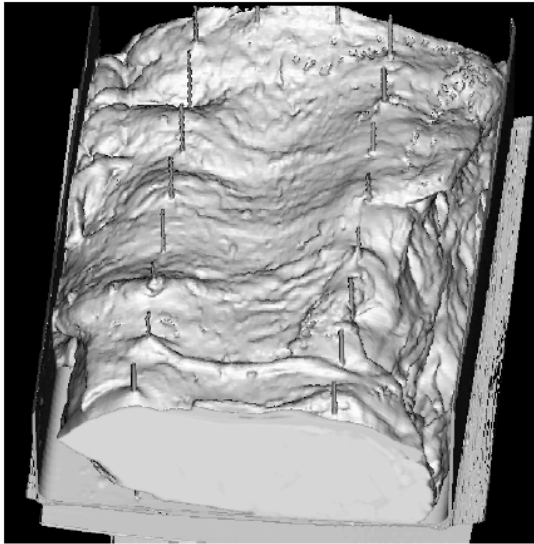Figure 9: Strip loin 5 cold CT measurement (primals fixed during rigor)

Figure 10: CT image of whole primal with showing regions marked for intramuscular analysis
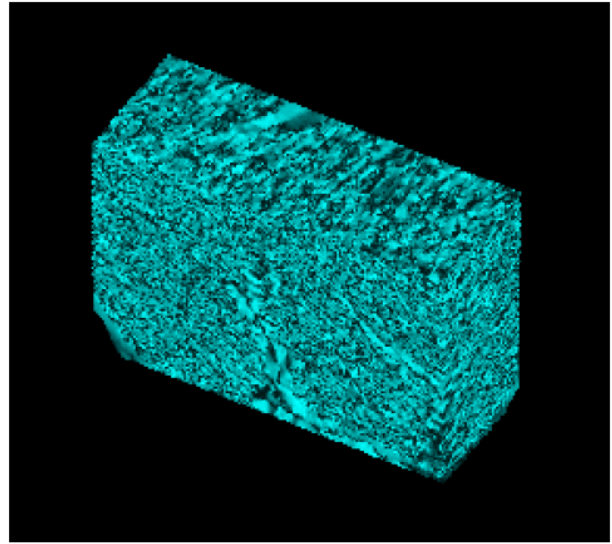
Figure 11: Region of muscle excised for chemical analysis and corresponding intramuscular fat isolated using analyse direct software

Using the software, slices were cut out of the 3D image of the whole primal (marked by bamboo pegs in Figure 9) that matched the physical slices cut from the cold primal (see Figure 6). CT estimation of fat was compared to chemical methods (see Figure 11). Care was taken to cover the same locations for the two slice methods (virtual and real), using toothpicks as guides for the slice boundaries. However, as our focus was on intramuscular fat, there were some differences between the location and shape of the virtual and real slices, which was perhaps the greatest source of error in the analysis.

Histograms were then constructed for all the voxels in the slice, and the two peaks for fat and muscle were usually observed. The location of the peak depended on whether the CT scan was conducted in a hot or cold condition. For hot scans, the fat peak was around -120 Hounsfield Units, while muscle was at around 60 Hounsfield Units. In cold scans, the fat peak shifted to around -75 Hounsfield Units and the muscle peak shifted to around 70 Hounsfield Units.

The percentage fat in the images was determined by one of three approximation methods – binary threshold, complex threshold and mixture modelling. The binary threshold value assumed all voxels contributed to either the fat or muscle content, allowing the threshold value to be adjusted to optimize the difference between the image analysis method and the chemical measurements. The complex threshold method put threshold boundaries around the known Hounsfield Units for fat, muscle and water, allowing each threshold to be varied to best optimize the difference.
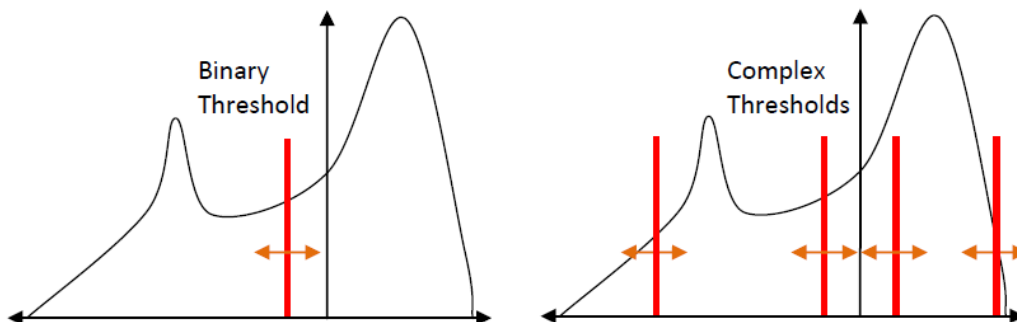
The mixture method considers the fact that each voxel between the muscle and fat peaks is likely to contain a mixture of muscle and fat as illustrated in Figure 12. Therefore both contribute to the voxel intensity according to the law of mixtures.
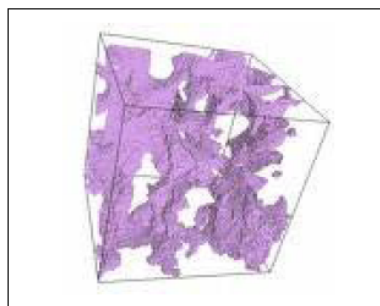

Figure 12: Illustration of CT voxel size (0.5mmx0.5mm) where the CT measurement reports the content of the voxel as an "average" density (Hounsfield Unit). Analysis finer than this CT's capabilities would show a combination of both muscle and fat within the voxel.

The equations to determine the fractions of fat, v_{Fat}, and muscle, v_{Muscle}, are given by

$$v_{fat} = \left(\frac{H - H_{Fat}}{H_{Muscle} - H_{Fat}} \right)^n$$

$$v_{Fat} + v_{Muscle} = 1$$

Where H is the Hounsfield Unit associated to the histogram bin, H_{Fat} and H_{Muscle} are the Hounsfield Units at the centre of the fat and muscle peaks, and n is an empirical factor that can allow some non-linearity into the approximation. The muscle peak was sufficiently large to enable H_{Muscle} to be determined for every histogram. However, the fat peak was sometimes so small that it could not be distinguished from the noise, so a fixed value of H_{Fat} was used. Fixed values like this are often referred to as the "Threshold" value. In medical CT different tissues have known densities and subsequently have universally agreed threshold values.
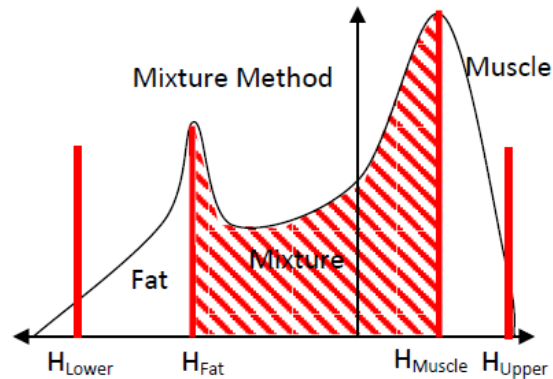
The equation for the percentage fat therefore was given by

$$Fat\% = 100 \frac{\left(\int_{H_{Lower}}^{H_{Fat}} F_{voxels} \, dH + \int_{H_{Fat}}^{H_{Muscle}} v_{Fat} F_{voxels} \, dH \right)}{\int_{H_{Lower}}^{H_{Upper}} F_{voxels} \, dH}$$

Where F_{voxels} is the frequency of the voxels for any Hounsfield Unit in the histogram.

In general, all three methods produced similar results. For a given muscle and temperature, the mixture method produced slightly better correlations than the other two methods, but this difference was small and did not offset randomness introduced by experimental sources of error.

5 Results and Discussion

Differences in tissue density between hot and cold CT for whole primal data
Hounsfield units are a measure of the density of the tissue as measured by the CT machine. Significant differences were observed between hot and cold Hounsfield units for both fat and muscle in both primal types.

Hot vs. Cold Fat Tissue
Table 4 shows that cold Hounsfield units for fat were similar for both Strip loin and cube roll, however the peak for fat in hot primals was slightly lower in the Strip Loin than in the Cube Roll (CR-118 SL -111). This generated a bigger difference between hot and cold fat peaks in cube roll primal as opposed to Strip Loin (60.5 CR & 51.5 SL). It is also noted that there was greater variation in the histogram peak for fat in cold cube roll as compared to other fat measurements. As the data shown is for the entire primal (no filtering of extra-muscular fat) this is consistent with the fact the strip loin samples had a much greater percent of extra-muscular fat than the cube roll samples as demonstrated in Table 6.

Hot vs. Cold Muscle Tissue
Differences in density of muscle were observed between hot and cold scans. However Table 4 shows a greater consistency was seen between primal types for the change in Hounsfield units for the primal peak between hot and cold. The peak for hot muscle on average occurred at 60 Hounsfield units, and the average peak for cold muscle occurred at approximately 68. This

produced a consistent increase of 8 Hounsfield units in the peak for muscle between hot and cold. Further consideration for reasons causing the change in muscle peak is given on Page 18.

Table 4: Raw data showing difference Hounsfield units from both hot and cold CT scans for fat and muscle. (Data is for whole primal, no filtering of extra muscular fat has been applied to this data)

	Fat			Muscle		
	HOT	COLD	Difference	HOT	COLD	Difference
Strip Loin 2	-113	-61	-52	61	69	-8
Strip Loin 3	-109	-59	-50	61	69	-8
Strip Loin 4	-113	-63	-50	59	65	-6
Strip Loin 5	-109	-55	-54	59	67	-8
Average	-111	-59.5	-51.5	60	67.5	-7.5
Standard deviation	2.31	3.42	1.91	1.15	1.91	1.00
Cube Roll 1		-73			67	
Cube Roll 2	-117	-61	-56	61	69	-8
Cube Roll 3	-119	-55	-64	61	69	-8
Cube Roll 4	-115	-49	-66	61	69	-8
Cube Roll 5	-121	-65	-56	63	71	-8
Average	-118	-60.6	-60.5	61.5	69	-8
Standard deviation	2.58	9.21	5.26	1.00	1.41	0.00

Figure 13 is used to illustrate the general trend that was observed in Hounsfield units between hot and cold CT scans. Basically the general trend observed is a shift in Hounsfield units from left to right between hot and cold scans with fat tissues shifting more than muscle tissues.
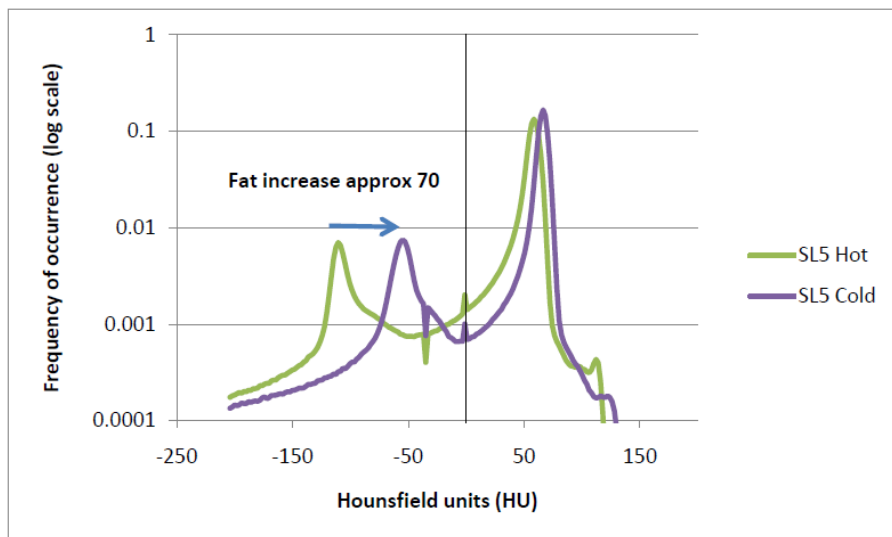
Figure 13: Histogram of both fat and muscle for whole hot and whole cold primal.

Table 5 estimates total percent fat of the whole primal using the threshold method outlined in Section 4 under data analysis. Similar to data shown in Table 4 the results shown in Table 5 are based on the CT data from the whole muscle primal, as opposed to intramuscular CT data only. The percentages reported are therefore a predicted fat % (or lean meat yield) of the whole primal. The purpose of this table is firstly to observe the differences between hot and cold predictions, and secondly to compare consistencies between predicted values for hot and cold across different muscles.

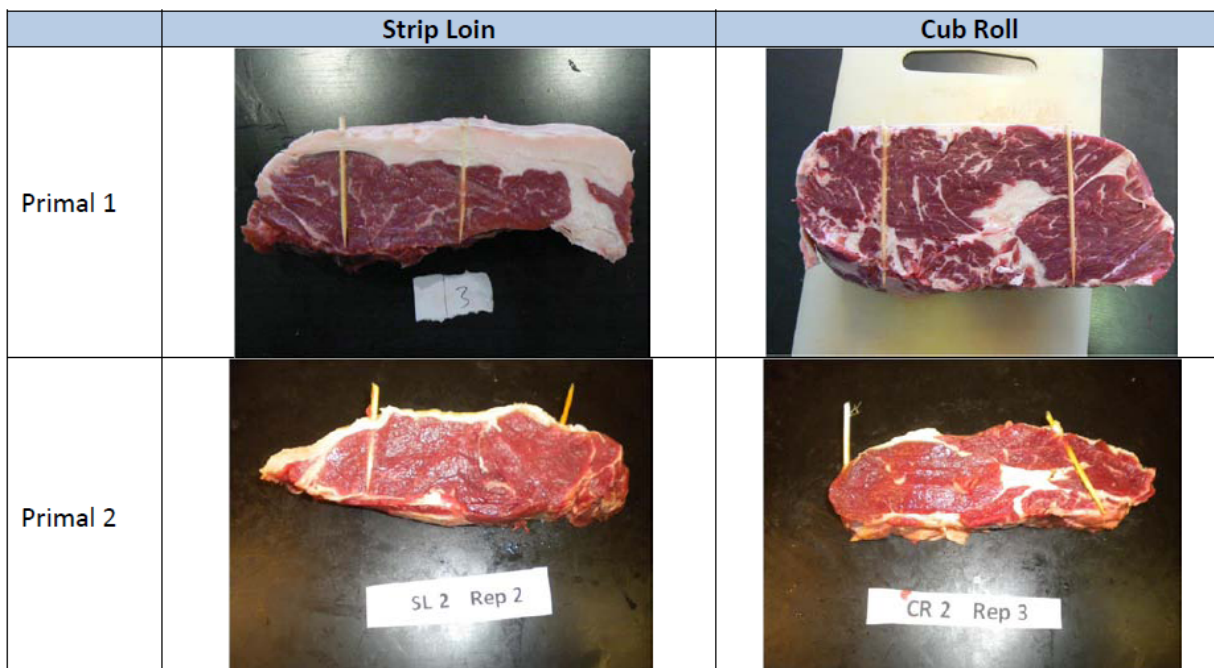A greater percent fat is predicted for the whole primal in strip loin as compared to cube roll, this is expected because of the higher amount of extra-muscular fat that was on the strip loin primals as compared to the cube roll primals.

The main point to note from this table is the standard deviation for the difference between hot and cold predictions is less than 1% for both Strip Loin and Cube roll. This would suggest that while a difference may be seen between hot and cold data, this difference remains consistent with primals that had different overall fat contents. Correlation between hot and cold percentages in this case are very high (R2=0.99) indicating comparison between hot and cold CT scans is achievable if a standard conversion factor was developed. However, many more specimens would be required to test and optimize the model's parameters used in this analysis.
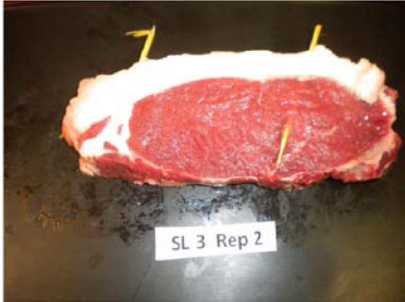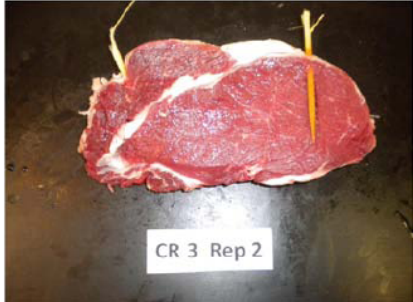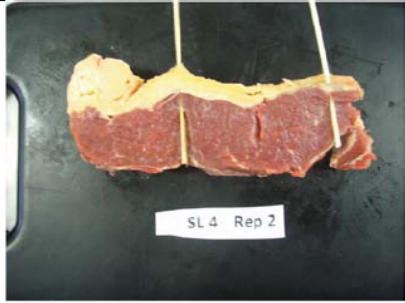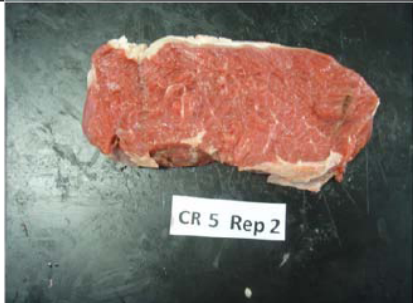
Table 5: Predicted fat % of whole primal (including extra muscular fat) using the threshold Method

		Hot	Cold	Difference
Strip Loin	Muscle 2	7.30%	8.50%	-1.19%
	Muscle 3	25.82%	27.65%	-1.83%
	Muscle 4	16.14%	17.97%	-1.83%
	Muscle 5	11.57%	11.29%	0.28%
Average		15.21%	16.35%	-1.14%
Standard Deviation		7.94%	8.52%	0.99%
Cube Roll	Muscle 2	8.13%	7.58%	0.56%
	Muscle 3	5.81%	4.91%	0.90%
	Muscle 4	8.90%	8.34%	0.55%
	Muscle 5	3.00%	2.55%	0.45%
Average		6.46%	5.84%	0.62%
Standard Deviation		2.65%	2.64%	0.20%

Table 6: Cross section image of one slice from muscle primal

	Strip Loin	Cub Roll
Primal 1		
Primal 2	SL 2 Rep 2	CR 2 Rep 3

	Strip Loin	Cub Roll
Primal 3	SL 3 Rep 2	CR 3 Rep 2
Primal 4	SL 4 Rep 2	CR 4 Rep 3
Primal 5	SL 5 Rep 1	CR 5 Rep 2

There are indications that cooling may increase the volume of fat slightly (as measured using CT analysis). However there is insufficient volume of data to prove this observation and any change in volume could be accounted for in development of a conversion factor between analysis of hot and cold scans.

Exclusion of extra muscular fat from data
Images shown in Figure 14 and Figure 15 show the results of Analyze Direct software used to filter out certain components of the primal. Figure 14 shows chemical lean, or myofibrillar components of the primal removed. Further filtering is used in Figure 15 to differentiate between fat (light green), and intramuscular fat. The data for these images was captured from hot primals (less than 1 hour post mortem).
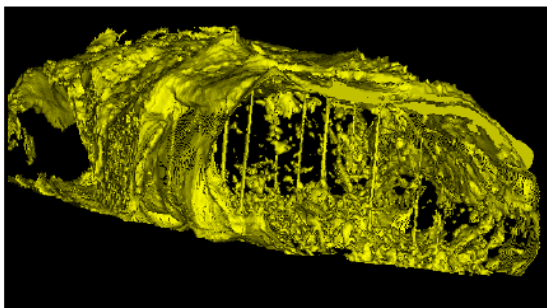
Figure 14: Strip Loin (5) Hot, Intra & Extra muscular Fat
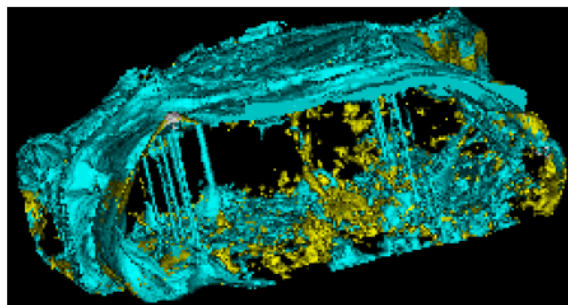
Figure 15: Strip Loin (5) Hot, Image rendered with filter to differentiate between intra and extra muscular fat

Figure 16 Shows a Hounsfield histogram of whole primal overlaid with the histogram of whole primal that had the extra muscular fat removed. The obvious point to note is that once the extra muscular fat has been removed from the data the fat peak seen at -110 is basically non-existent reflective of a reduction in volume of fat in the sample . This would suggest that while the fat peak may be a useful measurement parameter for a whole primal section, it is very difficult to identify a specific peak for fat once the when only looking at intramuscular data. The data displayed in the histogram for Figure 16 was from strip loin 5 which had an average chemical fat content of 2.92 %.
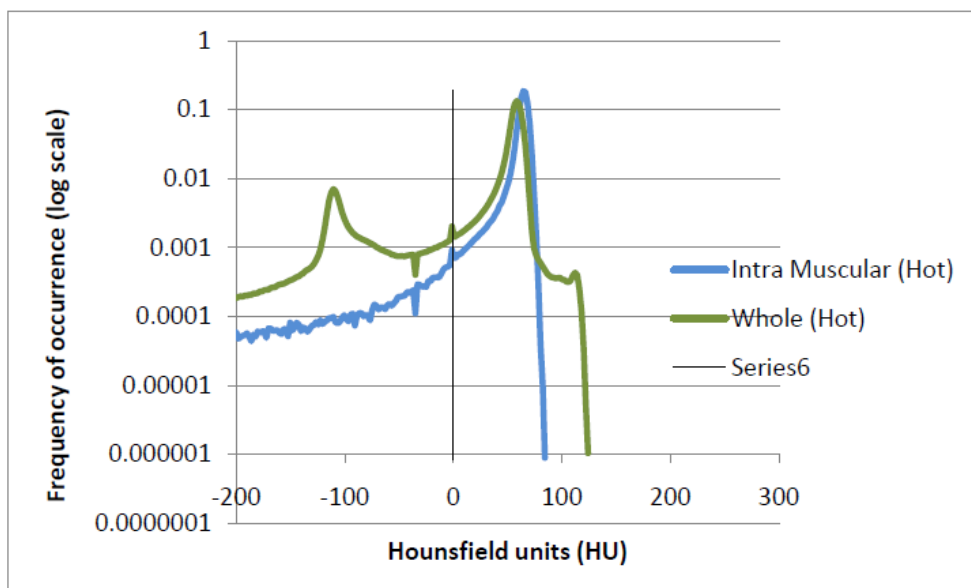
Figure 16: Histogram for total primal, overlaid with histogram of only intramuscular CT data.

Figure 17 displays a histogram of intramuscular CT data from a hot and cold strip loin primal. It is important to note that while a difference of 8 (HU) is observed in the peak for muscle between hot and cold intramuscular data, other data observed between the two trends is very similar. Two small peaks are observed at -40 HU and also at 0 HU for both hot and cold measurements.
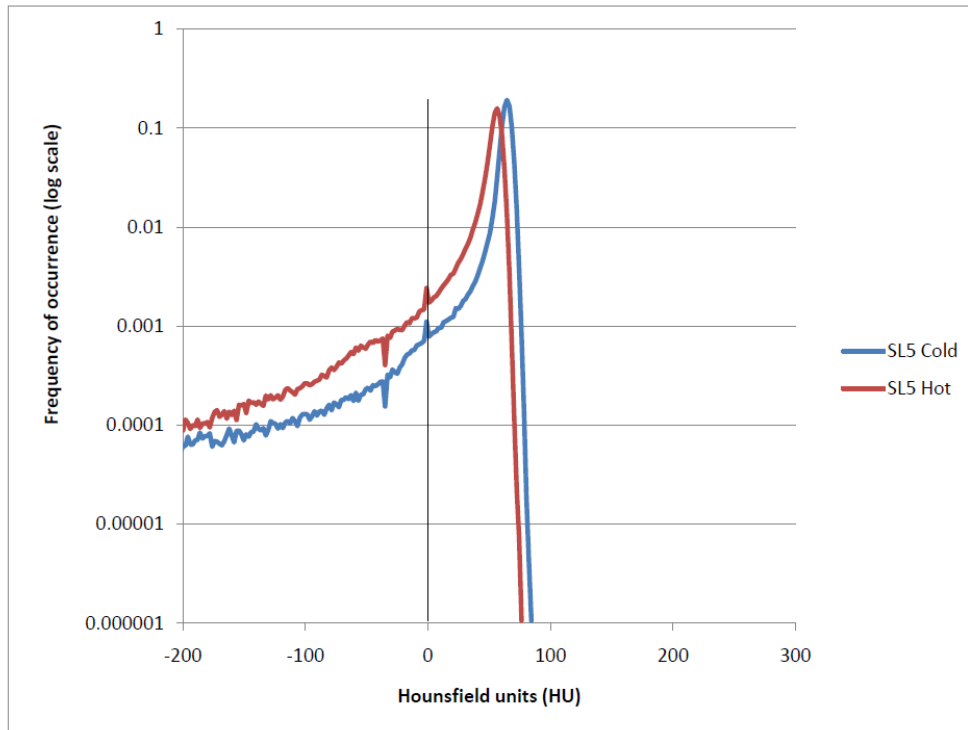
Figure 17: Overlay of hot and cold intramuscular fat peaks

Comparison of CT scans Hot v's Cold for muscle
As noted in Table 4 the change observed in Hounsfield units where the peak for muscle occurred between hot and cold data was very consistent at 8 (HU). Possible factors driving this change between hot and cold CT measurements include

1. Increase in the density of the primal due to cold shortening.
2. The physical dimensions of the voxels being such that any given voxel contains a certain amount of intramuscular fat, and this increase in the HU of fat the fat peak between hot and cold (50 – 60 HU) is also influencing the fat peak by shifting to the right 8 HU
3. Possibility of water loss.

Difference between hot and cold CT scans for predicting % intramuscular fat
Table 7 shows the difference between the predicted hot and cold intramuscular fat content. The data is similar to that previously displayed in Table 4, however it's important to make the distinction that Table 4 applies data from whole primal which includes extra muscular fat, where as Table 7 applies to a different data set which only includes the intramuscular sections that were used for chemical analysis.

It is important to note that the differences observed in the predictive ability of the percent intramuscular fat between hot and cold are quite small for both strip loin and cube roll primal. Similar to trends observed with the whole primal, the difference in the prediction of fat content was slightly higher for cold measurements in the strip loin, and slightly higher in the hot CT measurements for cube roll. These differences seen between hot and cold data and primal types can only be made at the observational level and are not statistically significant.
The main point to make from Table 7 is that minimal differences were observed in the predicted intramuscular fat values between hot and cold CT scans.

Table 7: CT Prediction of intramuscular fat using complex threshold analysis method

		Hot	Cold	Difference
Strip Loin	Muscle 1	NA	2.32%	NA
	Muscle 2	1.31%	1.48%	0.17%
	Muscle 3	0.13%	0.37%	0.23%
	Muscle 4	1.86%	1.94%	0.08%
	Muscle 5	1.49%	1.20%	-0.29%
Average		1.20%	1.46%	0.05%
StDev		0.75%	0.75%	0.24
Cube Roll	Muscle 1	NA	12.56%	NA
	Muscle 2	0.90%	0.90%	0.00%
	Muscle 3	0.22%	0.47%	0.25%
	Muscle 4	0.81%	0.76%	-0.05%
	Muscle 5	0.79%	0.36%	-0.43%
Average		0.68%	0.62%	-0.06%
StDev		0.31%	0.25%	0.28

Due to the narrow range of intramuscular fat content contained within the dataset, correlations are not included for this analysis.

5.1 Prediction of marbling using CT scans

The small size of the dataset and limited range of marbling scores do not allow meaningful correlations between CT marbling score and chemical fat content. However, the CT scans certainly show three dimensional marbling indicating that if cold CT scans can measure marbling accurately, hot scans have the same capability. Particularly for fine marbling, further work should consider the use of both three dimensional analysis (voxels) and two dimensional analysis (pixels). The sampling methodology established during the project set a baseline that would allow existing data to be included as part of a larger study if that were to occur.

5.2 Intramuscular chemical fat

Chemical fat results are displayed in Table 8. For both Strip loin and Cube roll the first primals collected had the highest chemical fat content. This can also be observed visually in from the cross section images provided in Table 6.

Table 8: Results from Chemical fat analysis

Muscle Type	REP #	M1 % Fat	M2 % Fat	M3 % Fat	M4 % Fat	M5 % Fat
Strip Loin	1	13.5	2.2	3.2	2.4	2.2
Strip Loin	2	6	2.8	3.5	2.8	4.3
Strip Loin	3	8.2	2.2	1.8	2.4	5.1
Strip Loin	4	6.4	2.7	4.4	1.6	2
Strip Loin	5	5.1	2.8	2	2.6	1.8
Strip Loin	6	5.2	3.2	2.3	2.8	2.6
Strip Loin	7	4.9	3.4	3.1	1.9	2.5
Strip Loin	8	7.5	-	-	-	-
Strip Loin	9	6.2	-	-	-	-
Strip Loin	10	11	-	-	-	-
Average		7.40	2.76	2.90	2.36	2.93
St Dev		2.82	0.45	0.92	0.45	1.26
Cube Roll	1	25.2	3.8	2.4	1.5	1.4
Cube Roll	2	19.1	4.5	2.9	2.8	2.5
Cube Roll	3	19.8	4.3	6	3.3	1.8
Cube Roll	4	11.4	3.4	4.9	5.5	2.5
Cube Roll	5	14.3	2.9	7.6	3.1	2.6
Cube Roll	6	NA	3.2	5.6	3.6	3
Cube Roll	7	NA	4.4	7.6	3.5	4.4
Average		17.96	3.79	5.29	3.33	2.60
St Dev		5.33	0.64	2.06	1.19	0.96

5.3 Potential considerations for Aus-Meat and the Language and Standards Committee; assessment of marbling

Current industry practice for the determination of marbling is achieved by a 2 dimensional visual assessment of the intramuscular marbling on the primal surface within the eye muscle. Any intramuscular fat which touches or joins the extra muscular fat is referred to as "exclusion fat" and is not considered in the assessment for marbling. If this same rule was to be applied to a 3 dimensional CT image of the primal it is important to note that while many of intramuscular seams seen on any given 2 dimensional surface may not touch or join the outside surface of the primal for a given plane, a 3 dimensional assessment of primals show a large portion of these seams may actually touch the external boundary of the eye muscle at a different point along the axis of the primal. This is seen in Figure 18 where using the Analyze Direct software the CT image is rendered to only show intramuscular fat. Although it is difficult to see in this 2 dimensional still image, A rotatable 3D image shows many of the intramuscular fat seams seen in this image actually touch the external boundary of the eye muscle at some point. This would suggest that if the same current marbling assessment rule was applied for the 3 dimensional assessment of marbling, predicted scores may be significantly lower than the current method. Alternatively if these fat seams were to be included in the analysis, predicted marbling scores would actually increase relative to the current assessment technique.

The same may also apply when collecting muscles samples for intramuscular chemical lean analysis. While fat on a sample may be included in chemical analysis, using current rules which remove exclusion fat from the CT data may actually result in inaccurate compassions.
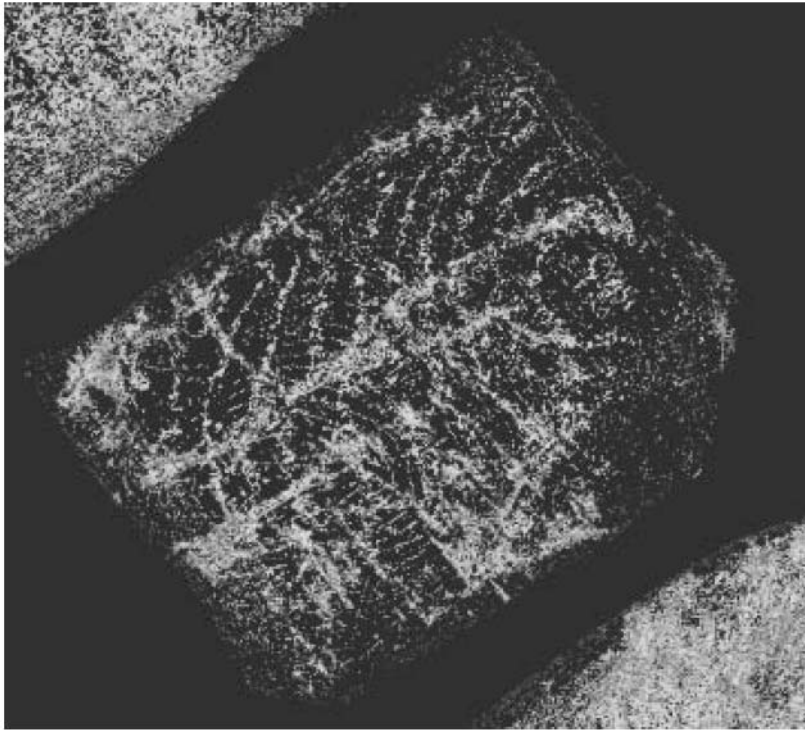

Figure 18: CT imaging of intramuscular fat seams.

6 Success in achieving objectives

Objective 1: Methodology

1. Great care needs to be taken to ensure that slices analysed in the images are nearly the same as the slices used for chemical fat analysis. The variability in the distribution of fat within a muscle, coupled with differences in the analysed volumes (CT vs. Chemical volumes), was likely the greatest source of error in the study.

2. Constraints during cooling are required when comparing hot and cold slices. Otherwise cold shortening added significant error into the comparative slice locations and to the image analysis methods.

3. Consistent voxel sizes, pixel sizes and slice depths are required for each scan.

4. Smaller voxel/pixel sizes and more slices per cm are better, as this enables finer fat seams to be observed.

5. Images that exhibit striations reduce the clarity of the image analysis.

6. Simple threshold, complex threshold and mixture methods were trialled. Mixture methods did exhibit a slight advantage over the other methods, however, the improvement was not large. The mixture method may be most useful for fine fat seams.

7. Large fat seams are readily measurable. Fine fat seams are not as easy to measure or distinguish from the muscle.

Objective 2: Data analysis results

1. Significant differences were observed in the Hounsfield units for hot and cold bovine fat, see Table 9.

Table 9: Averaged differences observed in hot and cold CT values for Strip loin and Cube roll

	Hot (HU)	Cold (HU)	Difference
Strip Loin (Muscles 2- 5)	-111	-59.5	-51.5
Cube Roll (Muscles 2- 5)	-118	-60.6	-60.5

2. Significant differences were also observed in the Hounsfield unit peak for muscle when comparing between hot and cold CT scans. Changes in CT values between hot and cold scans for muscles peak were more consistent between primal type when compared to changes observed in fat between hot and cold (Table 10)

Table 10: Differences observed in Hounsfield units for the muscle peak between hot and cold CT scans

	Hot (HU)	Cold (HU)	Difference
Strip Loin (Muscles 2- 5)	60	67.5	7.5
Cube Roll (Muscles 2- 5)	62	69	8

3. Significant trends were observed between the prediction of fat content of whole primals for hot and cold CT measurements with the inclusion of extra muscular fat (R_2 0.99) (Table 11).

Table 11: Average predicted fat content of whole primal (including extra muscular fat)

	Hot % voxel in fat range	Cold % voxel in fat range	Difference
Strip Loin (Muscles 2- 5)	15.21%	16.35%	-1.14 %
Cube Roll (Muscles 2- 5)	6.46%	5.84%	0.62 %

4. Small differences were also observed in the predicted intramuscular fat content of strip loin and cube roll primals when comparing between hot and cold CT primals measurements.

	Intramuscular fat HOT CT Scan	Intramuscular fat COLD CT Scan	Difference
Strip Loin (Muscles 2- 5)	3.34%	3.31%	.22%
Cube Roll (Muscles 2- 5)	1.53%	1.40%	-.13%

5. Observations on 3 dimensional CT images showed a large portion of intramuscular fat seams do come in contact with the external surface of the muscle. Consideration would need to be given to in future work as to how this fat should be included for analysis of marbling grades.

7 Recommendations to MLA for future work

A number of risks were previously identified for the commercialisation of online CT scanning of carcases. The outcomes of this research mitigate the risk of measurement inconsistencies between hot and cold scans.

A number of other risks have not yet been mitigated. The following recommendations take a step by step approach to mitigating risk as they progressively develop on the outcomes from each previous recommendation:

1. Having identified that hot and cold CT measurements on Bovine muscle have little influence on the predictive capability to segregate fat and muscle tissue at a preliminary level, further work should quantifying CT's ability to measure marbling across the full range of marbling scores. This would involve increasing the existing data set to include a wider range of marbling scores. Logistical constraints and trial costs could be greatly reduced by measuring cold bovine muscle only.

2. Further work needs to be conducted into various statistical and image analysis methods that can accurately correlate CT marbling measurements with chemical lean values, and to identify the advantages and disadvantages of each. a. This will be possible with a full data set achieved in recommendation 1

b. The results of recommendation 2 could form the basis of a submission to Aus-Meat for CT measurement of marbling.

c. Image analysis techniques developed during this work could be applied directly to automatic measurement of other parameters identified in the original cost/benefit analysis as worthwhile for plants to measure commercially.

3. Once the technical capability of the equipment is confirmed for the measurement of intramuscular fat, a specific plant should be identified where CT equipment can be installed, the trial work should continue on hot bovine muscles and carcass sections. a. Automatic identification of defects such as cysts, lesions and abcesses could be identified on plant which has not been done to date due to lack of access to defect samples.

b. Image analysis techniques developed in recommendation 2 would be applied directly in this work.