

# Final report

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## Development and validation of Process Modelling - Phase 3 [Pship.150.07E]

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## **Abstract**

Since 2007, Meat and Livestock Australia (MLA) and Meat and Wool New Zealand (MWNZ) had jointly worked on the development, implementation and validation of a process modelling tool, which aptly named Process Modelling. The Process Modelling tool has developed to a point where additional components can be uploaded into the model to provide greater predictive eating quality capabilities. Specifically, the Process Modelling tool consists of two drivers of eating quality outcomes; that being temperature and pH. Previous phases of the development and validation relates to previous projects [Refer to P.PSH.0228 and P.PSH.0264].

In its third iteration of development and evaluation (i.e. third phase), the current phase evaluates the upload of relevant aspects of the Meat Standards Australia (MSA) Eating Quality model. Specifically, the current (Phase 3) evaluates the required modifications to the current prototype Processing Model version to integrate with aspects of the MSA model. This will involve technology transfer and Australian and New Zealand early adopter processors.

The specific objectives of the current Phase 3 were to:

- Determine the underlying putative mechanism for the heat shortening effect (through arbitrary mathematical description of a sufficiently broad dataset of samples exposed to a range of temperatures for a range of durations).
- Develop the necessary matrix of data to allow a predictive model of the heat shortening effect to be constructed, and
- Update on the development of the commercial dataset developed in collaboration with commercial partners in New Zealand.

A model calculating the post-rigor increase in ultimate tenderness under denaturing conditions has been developed in the current project. This shows that the post-rigor effects are significant and needs to be incorporated into development of process specifications for large, slow-cooling carcasses that have a rapid pH decline.

The outcome was to report on MSA data collation and uploading of information into model and to assist MSA to revise current MSA meat science course and other training modules to include the Process Modelling tools. It involved identifying the required modifications to validate the model using MSA data from one plant in Australia to facilitate technology transfer to MSA.

The next step will be to evaluate two plants in much more detail: one will use the conventional high voltage stimulation system and the other the medium voltage system. In both cases, the current performance of the system will be audited in greater detail, particularly by monitoring intermediate levels of pH decline, and end point measurements of tenderness, purge loss and colour will be collected. These data will be used as inputs to the SSF models and will assist in the refinement of the predictions. Longer term, a number of processing options will be put into the model and the quality predictions will be used to identify processing modifications that should be trialled in each plant to further improve and optimise meat quality.

## Executive summary

### 1) Modelling the heat-toughening phenomenon.

The project has extensively evaluated the effects of high pre-rigor temperatures on the final tenderness of beef. However, more recent work has recognised that the heat toughening phenomenon is not limited to the pre-rigor period but continues in the post rigor period.

In 2007, research was conducted to define the kinetics of the post rigor effect by subjecting samples to varying period of high temperature. The results were, however, unexpected: the heat toughening effect was maximal for exposure periods of 6-10 hours at 40°C and 35°C, but longer periods of exposure reversed this effect and the final tenderness became normal. Subsequent effort went into confirming this effect (it was confirmed) and making additional measurements in an effort to understand this unexpected result.

Why the samples should reach a normal tenderness after prolonged exposure to high temperature remains difficult to explain. In the absence of an underlying putative mechanism for the heat shortening effect, it is still possible to model the effect using a purely arbitrary mathematical description of a sufficiently broad dataset of samples exposed to a range of temperatures for a range of durations. This objective will undertake to develop the necessary matrix of data to allow a predictive model of the heat shortening effect to be constructed.

### 2) Development of the commercial dataset

Silver Fern Farms were selected as the commercial partner for this work because they operate a diverse range of processing regimes across each of their lamb plants and also have an in-house quality auditing system which includes regular shear force testing. Therefore, data was available to incorporate into the process model.

Silver Fern Farms supplied Carne Technologies with data for 6 of their lamb processing plants. These data consisted of processing details, temperature regimes and shear force results. Data was put into the model and used to provide predictive outcomes on carcass cooling, shear force and water binding capacity. Taken collectively, the model predictions were very encouraging; while the shear force predictions from one plant did not correlate particularly well to the measured values, this was not surprising as the measured values were far lower than would normally be expected from this process scenario and brings into the question the validity of the measured shear force. In contrast, the predicted result was far more in keeping with expected results from this process.

The process model, with the SFF simulations, was demonstrated to the Company at their head office last month. All staff expressed significant interest in it and all agreed that this process would provide significant value to the Company and should be used as part of their process tailoring and optimisation activities. Procedures to achieve this were discussed and agreed.

### 3) Recommendations

The next step will be to evaluate two plants in much more detail: one will use the conventional high voltage stimulation system and the other the medium voltage system. In both cases, the current performance of the system will be audited in greater detail, particularly by monitoring intermediate levels of pH decline, and end point measurements of tenderness, purge loss and colour will be collected. These data will be used as inputs to the SSF models and will assist in the refinement of the predictions. Longer term, a number of processing options will be put into the model and the quality predictions will be used to identify processing modifications that should be trialled in each plant to further improve and optimise meat quality.

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# 1. Background and scope

## 1.1 Background

The concept of the Central Processing Management System (CPMS) that underpins the MQST program depends on quantifying the interactions of processing variables so that meat quality outcomes can be predicted under a range of processing conditions. The core processing conditions that need to be managed are the rate of change of pH and temperature, and how these interact to affect quality will differ according to which muscle or species of meat animal is being considered. The meat quality attributes that need to be predicted by the model are tenderisation, colour and colour stability and water binding capacity.

Two mathematical models have previously [i.e. Phase 1 & 2; Refer to P.PSH.0228 and P.PSH.0264] been developed at AgResearch: a procedure to calculate the rate of tenderisation under different chilling regimes for electrically stimulated lambs (tenderness model), and a more ambitious pre-rigor model for hot boned meat that calculates the effects of temperature on the rate of pH decline and consequent tenderisation and protein denaturation (Meat Quality Model; MQM). The parameters for the MQM were derived largely from the scientific literature or from experimental data. This model still needs further validation and refinement.

The pre-rigor model was developed for hot boning because hot boning simplifies many of the predictive requirements. Predicting variable temperature gradients within a carcass, or within large muscles in a carcass, could be ignored, as could the need to accommodate differences in the skeletal restraints that either limit the extent of shortening in some muscles, or actually cause stretching. These variables still need to be quantified to produce a pre-rigor model for cold boning.

A second component of the CPMS modelling requirements is to account for the effects of electrical inputs to a carcass. The electrical parameters of these inputs will vary depending on the processing objectives (stunning, immobilisation, rigidity), as will their timing relative to slaughter.

This project will undertake two principal objectives: the first will be to continue the development of the pre-rigor model to accommodate the requirements of cold boning; the second will develop the electrical stimulation component to meet the requirements of the CPMS. Wherever possible, other projects within the Meat Quality Science & Technology (MQST) program will contribute the data to develop the models and will be used to validate aspects of the model as they are developed.

The Meat quality model has a number of potential applications. In its simplest form, it can provide a decision support tool for the meat industry to help identify improved processing conditions. Beyond this, it can integrate with the CPMS framework to integrate on-line measurements and processing controls with meat quality outcomes. Last, the modelling framework provides a useful environment to identify research requirements and to test meat science principles. At this stage in the development of the model, a structure for most of the key meat quality considerations has been constructed.

## 1.2 Previous research (Phases 1 & 2)

Previous research has extensively studied the effects of high pre-rigor temperatures on the final tenderness of beef. However, more recent work has recognised that the heat toughening phenomenon is not limited to the pre-rigor period but continues in the post rigor period.

In 2007, research [Phases 1 and 2; to P.PSH.0228 and P.PSH.0264] was undertaken to define the kinetics of the post-rigor effect by subjecting samples to varying periods of high temperature. The results were, however, unexpected: the heat toughening effect was maximal for exposure periods of 6-10 hours at 40°C and 35°C, but longer periods of exposure reversed this effect and the final

tenderness was equivalent to controls. Subsequent effort went into confirming this effect (it was confirmed) and making additional measurements in an effort to understand this unexpected result.

Why the samples should reach a normal tenderness after prolonged exposure to high temperature remains difficult to explain. In the absence of an underlying putative mechanism for the heat shortening effect, it is still possible to model the effect using a mathematical description of a sufficiently broad dataset of samples exposed to a range of temperatures for a range of durations. This objective therefore undertakes to develop the necessary matrix of data to allow a predictive model of the heat shortening effect to be constructed.

The effects of pre-rigor denaturing temperatures on the ultimate tenderness of meat has previously been quantified and incorporated into the MQM. More recently, the post rigor effects have been recognised but the behaviour of the system proved to be unexpectedly complex: Short periods of exposure to denaturing temperatures produced the expected increase in ultimate tenderness, but prolonged exposure (roughly, more than 12 hours) reversed this effect until a normal ultimate tenderness was attained.

From a commercial standpoint, this information has two implications: first, the contribution of the post-rigor tenderness effects needs to be quantified as this could have significant implications for the optimisation of pH/temperature conditions in a carcass. Second, the reversal of the toughening produced by denaturation by more prolonged exposures to high temperatures may, under the appropriate circumstances, be exploited to contribute to improving eating quality.

### **1.3 Scope of the current project (Phase 3)**

Since 2007, Meat and Livestock Australia (MLA) and Meat and Wool New Zealand (MWNZ) had jointly worked on the development, implementation and validation of a process modelling tool, which aptly named Process Modelling. The Process Modelling tool has developed to a point where additional components can be uploaded into the model to provide greater predictive eating quality capabilities. Specifically, the Process Modelling tool consists of two drivers of eating quality outcomes; that being temperature and pH. In its third iteration of development (i.e. third phase), the current phase evaluates the upload of relevant aspects of the Meat Standards Australia (MSA) Eating Quality model.

Specifically, the current (Phase 3) evaluates the required modifications to the current prototype Processing Model version to integrate with aspects of the MSA model. This will involve technology transfer and Australian and New Zealand early adopter processors.

## **2. Objectives**

The overall aims of the current (Phase 3) development and validation of the Process Modelling tool by:

- Incorporate MSA data
- Assist MSA to revise current MSA meat science course and other training modules to include process model
- Upload NZ commercial data into model.

The specific objectives were:

- Determine the underlying putative mechanism for the heat shortening effect (through arbitrary mathematical description of a sufficiently broad dataset of samples exposed to a range of temperatures for a range of durations).
- Develop the necessary matrix of data to allow a predictive model of the heat shortening effect to be constructed, and
- Update on the development of the commercial dataset developed in collaboration with commercial partners in New Zealand.

The expected outcome was to report on MSA data collation and uploading of information into model and to assist MSA to revise current MSA meat science course and other training modules to include the Process Modelling tools. It involved identifying the required modifications to validate the model using MSA data from one plant in Australia to facilitate technology transfer to MSA.

### 3. Methodology

The process steps used were:

- 1) Determine the heat toughening mechanism (Milestone 1)
- 2) Develop a predictive model for heat shortening (Milestone 2)
- 3) Develop commercial dataset

#### 3.1 Determine the heat toughening mechanism (Milestone 1)

This involved determining the underlying putative mechanism for the heat shortening effect (through arbitrary mathematical description of a sufficiently broad dataset of samples exposed to a range of temperatures for a range of durations).

*M. longissimus dorsi* muscles from both sides of eight prime beef carcasses were collected pre-rigor on 2 separate occasions (16 carcasses). The muscles were immediately cooled in a 15°C water bath and transported back to the laboratory. pH declines were continuously monitored in all the samples until rigor mortis was attained.

At rigor mortis, a sample from each animal was cooked for shear force measurement. A portion of each animal was distributed into water baths held at 33, 36, 39 & 42°C, where they remained for periods of 2, 5, 8 & 12 hours. After each time period, half the portion was immediately cooked for shear force measurement and the remainder vacuum packed and stored at 15°C for 5 days to age to completion.

#### 3.2 Develop a predictive model for heat shortening (Milestone 2)

Develop the necessary matrix of data to allow a predictive model of the heat shortening effect to be constructed (Milestone 2).

This required a modification to the model as follows:

- A post-rigor denaturation module to calculate a biphasic increase in ultimate pH needs to be integrated with the existing pre-rigor denaturation module. This latter component is based on integrating time, temperature and pH to calculate the denaturation of myofibrillar proteins (used to derive water binding capacity and colour effects) and enzymatic reductive capacity (used to derive oxygen consumption rate). The pre-rigor effects on tenderness are derived from the calculated denaturation of myofibrillar proteins.
- To integrate the pre and post rigor tenderness changes, the model was modified to allow the denaturation effects to persist beyond rigor mortis (myofibrillar proteins are protected by rigor mortis and the calculation normally ends at this point). Since pH decline has stopped at rigor mortis, the calculation has only a temperature and time component.
- Since there is not, at this stage, any mechanistic explanation for the biphasic nature of the effects of denaturing conditions on ultimate tenderness, the second phase of the tenderness changes (reducing ultimate kgf) is calculated as a reversal of the denaturation effects, using the same rate calculation.
- The kinetics of the tenderness changes produced by denaturation are calculated from the equations derived in the previous milestone. This allows calculation of the peak value of the final tenderness, the time at which the denaturation effects are 'reversed', and the temperature effects of the rate of denaturation.

### 3.3 Develop commercial dataset (Milestone 3)

Update on the development of the commercial dataset developed in collaboration with commercial partners in New Zealand (Milestone 3).

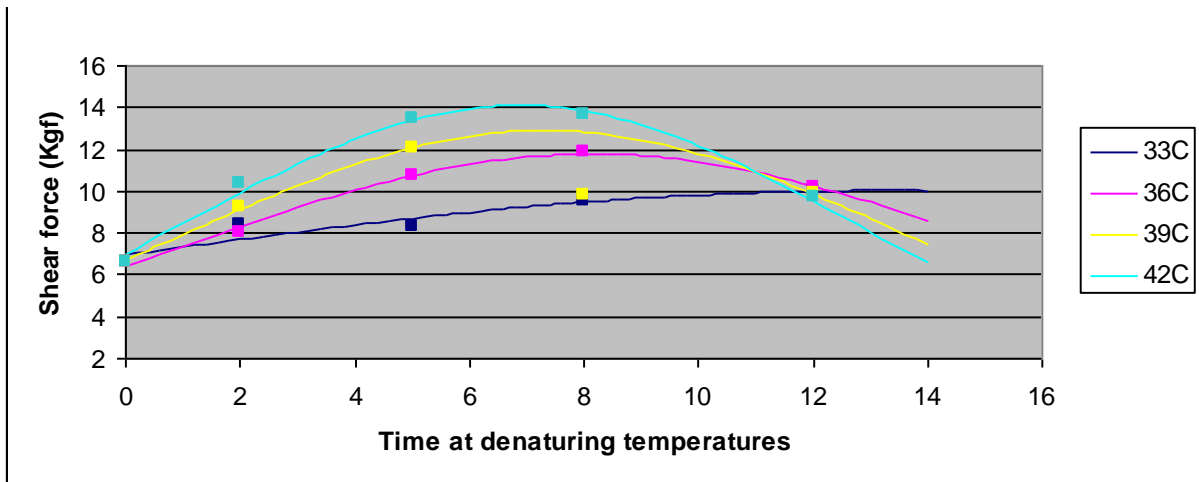
## 4. Results and discussion

### 4.1 Determine the heat toughening mechanism (Milestone 1)

The effect of post-rigor denaturing temperatures showed a similar pattern to those seen in previous comparable trials. A gradual temperature-dependent increase in the ultimate tenderness occurred, a process that is then reversed with continued exposure to denaturing temperatures. The lowest test temperature, 33°C, did not appear to show the reversal, but this is assumed to be due to not allowing sufficient time in the experiment to allow this effect to manifest.

The behaviour of the tenderness effect could be effectively fitted to a Gaussian curve [ $y = a \exp(-1/2(x-b/c)^2)$ ], which allowed independent expression of the temperature effect on the maximum toughness (a) and the time needed to reach the maximum (b). The  $r^2$  of the fit were 0.82, 0.97, 0.99 (8 hour time point omitted) and 0.94.

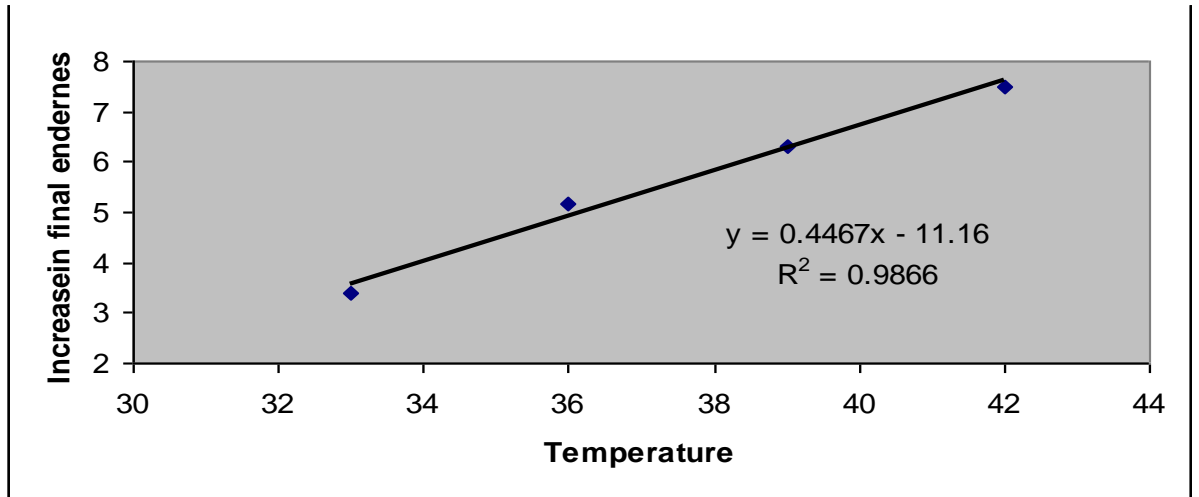




**Figure 1:** Curve fit of denaturation effect on tenderness of beef.

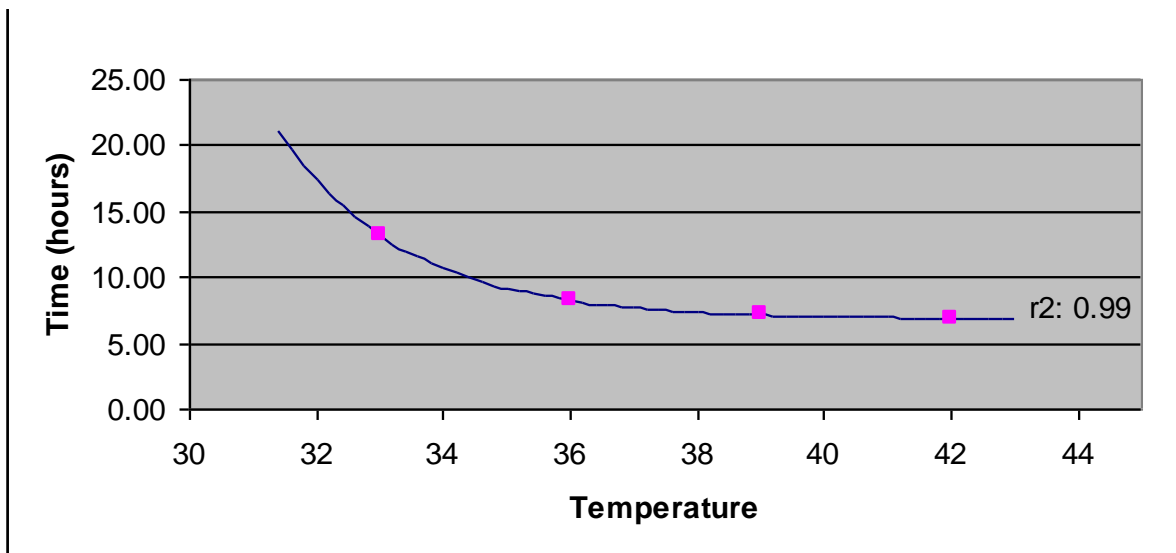
The key step in incorporating these tenderness effects into the model is converting the tenderness effects at a single, constant temperature into a calculation that accommodates a continuously varying chilling curve. This is done by defining the relationship between temperature and the 3 parameters that define the final tenderness at each temperature.

The effect on temperature on the final tenderness of the samples was linear (Figure 2).



**Figure 2:** The effect of temperature on maximum toughening of beef.

The exposure time needed to reach maximum toughening is accurately represented by an exponential curve (Figure 3).



**Figure 3:** The effect of temperature on time to maximum toughening of beef.

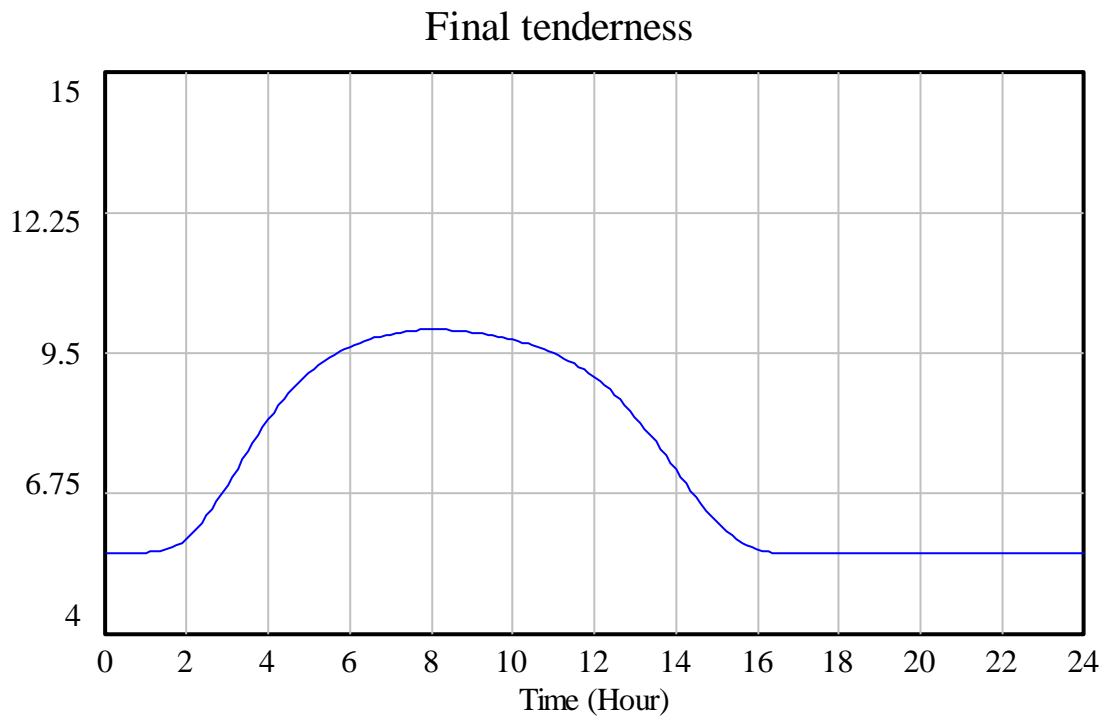
The last parameter relates to Full Width at Half Maximum (FWHA) and, again, can be accurately represented by an exponential curve ( $r^2:0.99$ ).

The data generated in these experiments generated the key formulae that will allow the effect of post-rigor denaturing temperatures on final tenderness to be calculated. The next milestone will undertake to transfer the relationships to the Meat Quality model and evaluate the implications in terms of the heat toughening phenomenon.

## 4.2 Develop a predictive model for heat shortening (Milestone 2)

To develop a predictive model for heat shortening required developing the necessary matrix of data to allow a predictive model of the heat shortening effect to be constructed (Milestone 2). Changes to the existing Process Modelling tool were required to accommodate for the effect of heat toughening.

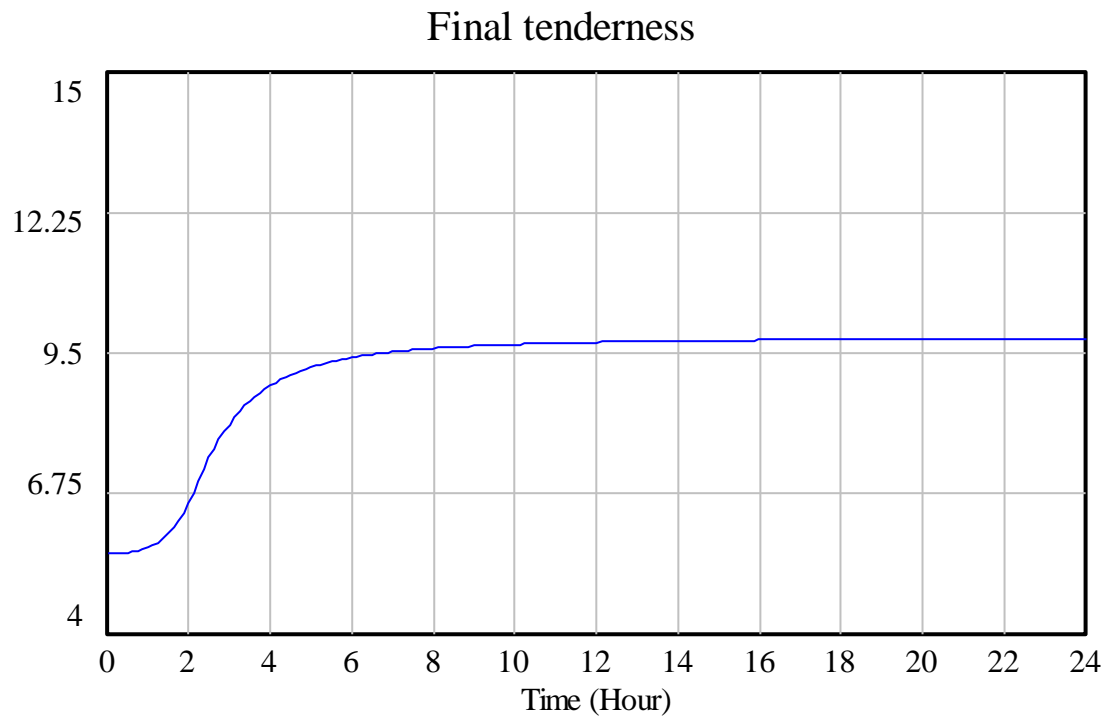
An example of the modelled changes in the ultimate tenderness caused by denaturation is shown in Figure 4. This represents the effects of continuous exposure to 39°C, which closely approximates the measured data.



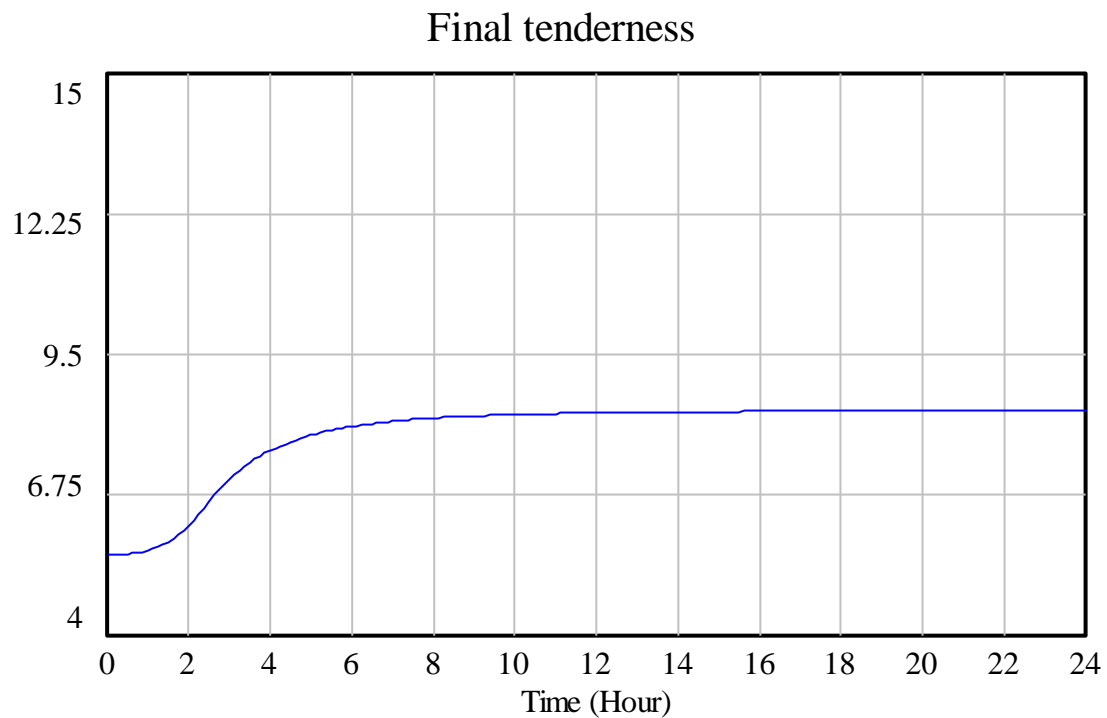
**Figure 4:** Effect of sustained incubation at 39°C for 24 hours on calculated ultimate tenderness

To evaluate the effects of different cooling curves on the calculated changes in ultimate tenderness, three calculated effects on final tenderness are shown below: in each case, the initial temperature is 42°C, rigor mortis (ultimate pH) is attained in 2.5 hours and the three cooling rates are defined by temperature at 24 hours post mortem.

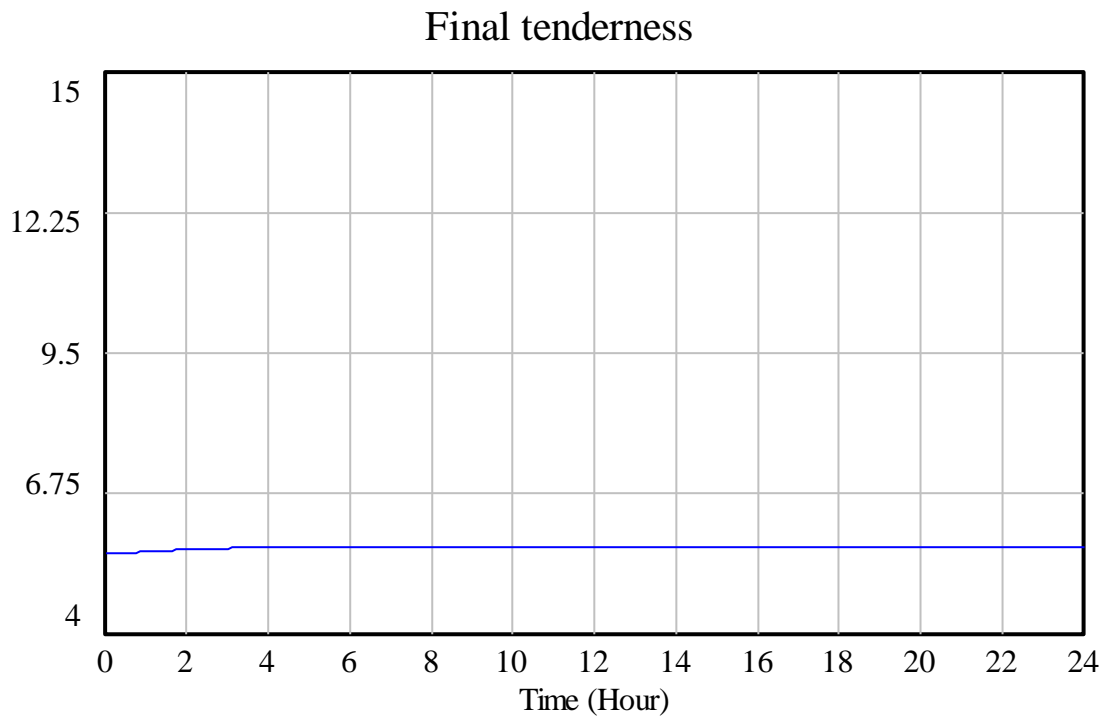
Under these conditions, cooling to either 20 or 10°C in 24 hours produced a significant increase in the ultimate tenderness: to approximately 10 and 8 kgf respectively. In both cases, approximately half this increase occurred post rigor. By cooling to 0°C in 24 hours, denaturation-induced toughening was avoided.



**Figure 5:** Cooling to 20°C in 24 hours.



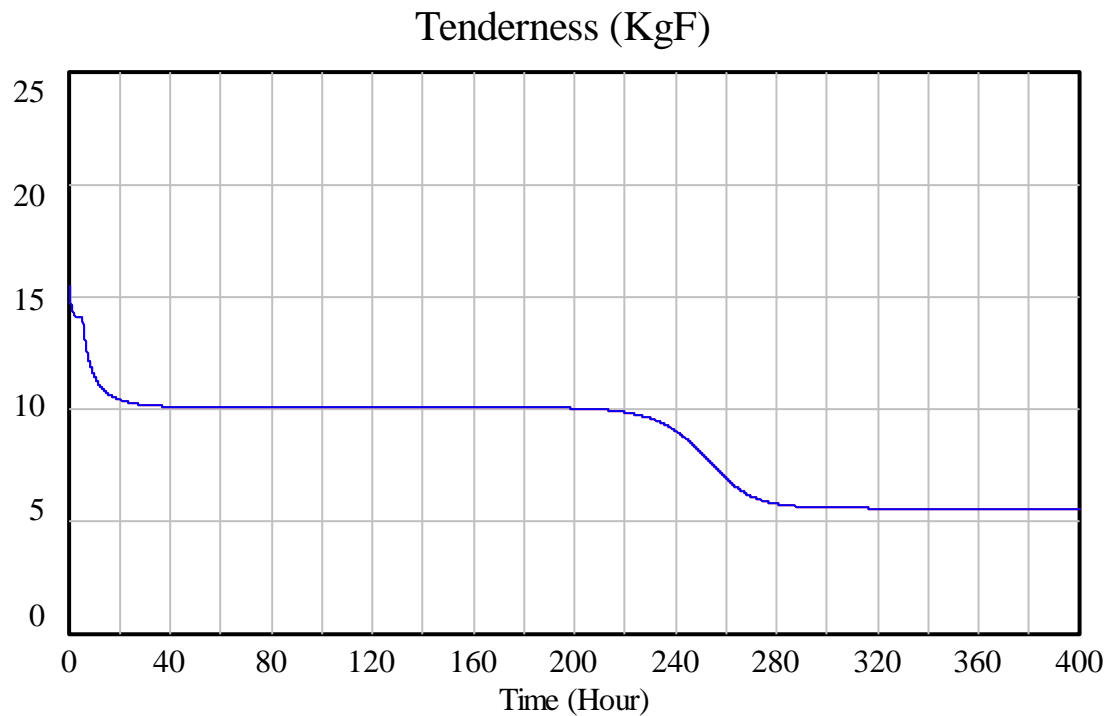
**Figure 6:** Cooling to 10°C in 24 hours



**Figure 7:** Cooling to 0°C in 24 hours.

The present model structure creates a potential anomaly in the tenderness curves in meat processed under denaturing conditions. Because of the ‘reversed’ denaturation – the improved ultimate tenderness produced by sustained exposure to denaturing conditions – relatively slow cooling curves show a two phase tenderness curve, as shown in Figure 5. This is seen, under the same conditions as used in the previous Figures, in the temperature range between 28 and 25°C at 24 hours: after approximately 180 hours of ageing, there is a second phase of tenderisation caused by the change in the ultimate tenderness.

There is obviously some doubt about whether or not this will ever have any commercial significance since carcasses must be cooled much faster than this under normal circumstances. However, this could represent an interesting test of the validity of this model structure. It is interesting that a similar biphasic ageing curve has been seen in semi-commercial trials: our own work with  $\beta$ -agonist treated lamb, a treatment that is normally viewed as toughening but which we have found will reach a normal ultimate tenderness (at least in lamb) if the ageing time is sufficient to overcome a plateau of partial tenderisation. This produces a similar ageing curve to that seen in Figure 8.



**Figure 8:** Two phase tenderness curve at cooling rates which produced 30°C in 24 hours.

A model calculating the post-rigor increase in ultimate tenderness under denaturing conditions has been developed. This shows that the post-rigor effects are significant and needs to be incorporated into development of process specifications for large, slow-cooling carcasses that have a rapid pH decline.

The phenomenon of improving ultimate tenderness in response to sustained exposure to denaturing conditions is unlikely to offer any benefit under normal cooling conditions.

### 4.3 Develop commercial dataset (Milestone 3)

A collaboration was instituted with Silver Fern Farm. This company was selected because of their multiple sheep plants and the differing processing specifications used in each.

Processing information was developed from 6 plants, based on auditing their stunning, stimulation and chilling regimes (Table 1). In addition, regular tenderness measurements carried out by the company at each of the 6 plants provided the endpoint measurements against which to assess the performance of the model (Table 2).

**Table 1:** Plant specific processing specifications.

Plant	Spinal discharge (400 V, 50 Hz; sec)	LV immobilisation (90 V, 15 Hz; sec)	HV stimulation (sec)	Carcass conditioning (hours / temp)	Chilling temperature (°C)
1	4	30	90	5 / 14.5	0.5
2	Not used	Not used	90	4 / 15	1.5
3	Not used	Not used	90	4 / 8	0
4	3	Not used	62	12 / 9.5	0
5	4	30	90	15 / 15	0
6	7	27	96	6 / 20	3

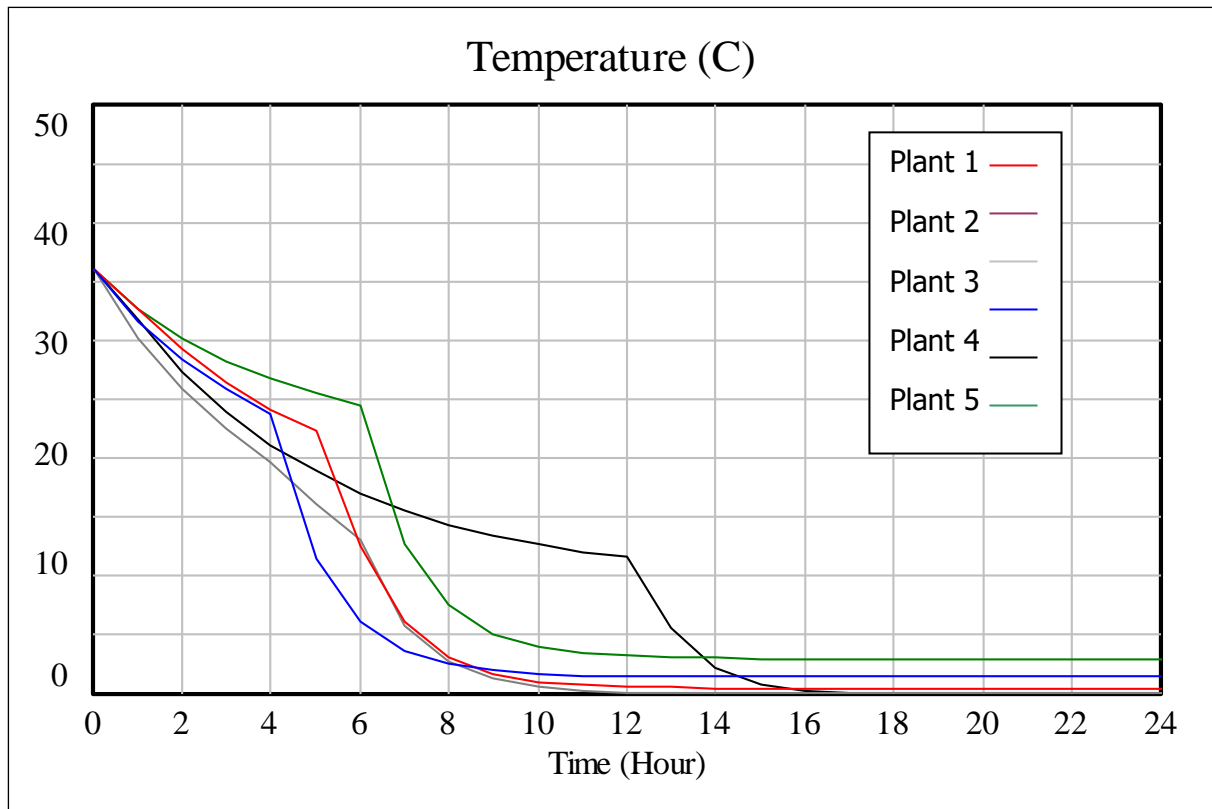
**Table 2:** Shear force results from each plant (in house tenderometer),

Plant	Tenderness (Mean kgf)
1	6.4
2	6.8
3	3.1
4	8.4
5	4.3
6	6.9

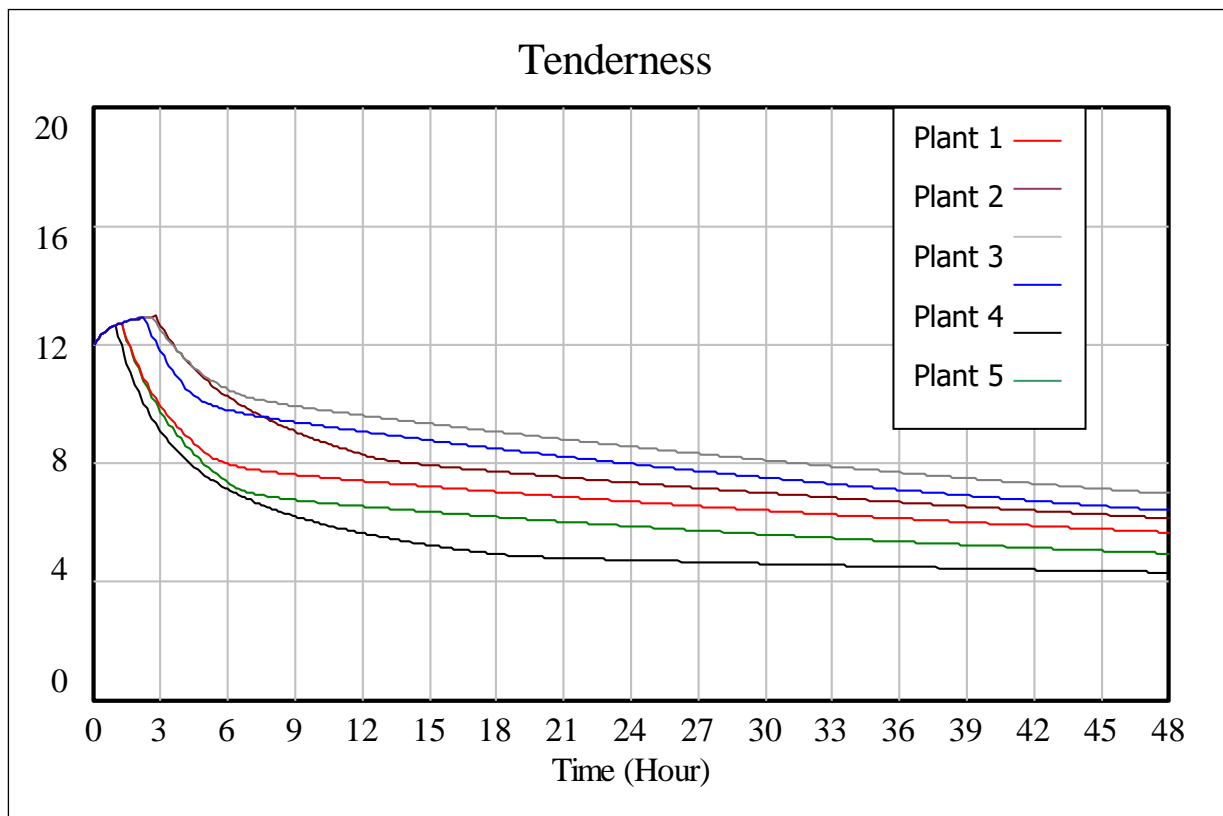
Temperature curves were calculated using the chilling conditions provided by SFF. The curves are based on predicted deep loin temperatures in 17 kg carcasses (Figure 9).

The pH declines are calculated from a combination of the electrical inputs to the carcass during processing and the subsequent temperature curve. The dominant variable is the use of LV electrical immobilisation, whereas the use of spinal discharge produces a much smaller effect. Since all the plants in this dataset used high voltage stimulation, all pH declines are relatively rapid.

Given the temperature and pH declines, the tenderness curves over the first 48 hours could be calculated (Figure 10).



**Figure 9.** Predicted temperature curves.



**Figure 10.** Predicted tenderness curves.



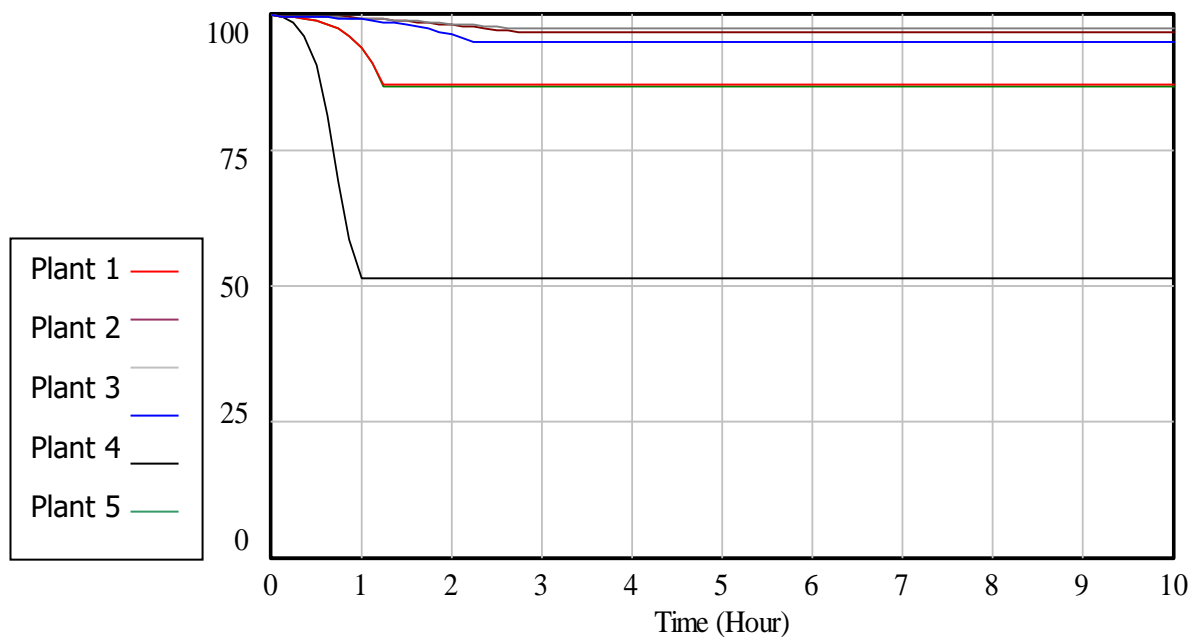
**Table 3:** Calculated tenderness curves for each plant.

Plant	Measured Tenderness (Mean kgf – as in Table 1)	Predicted tenderness from the model
1	6.4	6.7
2	6.8	7.5
3	3.1	8.5
4	8.4	7.8
5	4.3	4.7
6	6.9	5.8

In broad terms, the results appear very encouraging. The result for Plant 3 was particularly different but the measured tenderness values appear exceptionally low for this plant, without any particular reason for this (Table 3). The validity of the shear force measurement is therefore questionable.

In contrast, the low shear force value for Plant 5 is expected from the prolonged period of high temperature conditioning used in that particular process and the accurate prediction of the accelerated tenderisation produced by the model is encouraging.

The potential disadvantage of the extended period of high temperature conditioning in Plant 5 is predicted to result in decreased water binding capacity drip loss (See Figure 11). The extended high temperature conditioning is normally used in conjunction with producing a frozen product, so this disadvantage is not necessarily significant commercially. Nevertheless, two additional plants show some increase in potential drip loss, and both can be attributed to the effect of combining low voltage and high voltage stimulation.

**Figure 11:** Predicted Water binding capacity (% of maximum).

## 5 Conclusion and recommendations

### 5.1 Conclusions

The data generated in these experiments generated the key formulae that will allow the effect of post-rigor denaturing temperatures on final tenderness to be calculated. The next milestone will undertake to transfer the relationships to the Meat Quality model and evaluate the implications in terms of the heat toughening phenomenon.

A model calculating the post-rigor increase in ultimate tenderness under denaturing conditions has been developed. This shows that the post-rigor effects are significant and needs to be incorporated into development of process specifications for large, slow-cooling carcasses that have a rapid pH decline.

The phenomenon of improving ultimate tenderness in response to sustained exposure to denaturing conditions is unlikely to offer any benefit under normal cooling conditions.

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Silver Fern Farms supplied Carne Technologies with data for 6 of their lamb processing plants. These data consisted of processing details, temperature regimes and shear force results. Data was put into the model and used to provide predictive outcomes on carcass cooling, shear force and water binding capacity.

Taken collectively, the model predictions were very encouraging; while the shear force predictions from one plant did not correlate particularly well to the measured values, this was not surprising as the measured values were far lower than would normally be expected from this process scenario and brings into the question the validity of the measured shear force. In contrast, the predicted result was far more in keeping with expected results from this process.

The process model, with the SFF simulations, was demonstrated to the Company at their head office last month. All staff expressed significant interest in it and all agreed that this process would provide significant value to the Company and should be used as part of their process tailoring and optimisation activities. Procedures to achieve this were discussed and agreed.

### 5.2 Future research and recommendations

A presentation of the model and the results of these analyses were made to all the technical staff of the SFF group video link. There was significant interest expressed, as well as discussion about how a modelling tool such as the process model can be used by the company.

A further change since this dataset was provided is that two SFF plants have switched from high voltage stimulation to medium voltage stimulation (in one case, as part of the Smart Stimulation validation) combined with high frequency immobilisation, and this therefore offers the opportunity to compare the effect of the different stimulation systems.

The next step will be to evaluate two plants in much more detail: one will use the conventional high voltage stimulation system and the other the medium voltage system. In both cases, the current performance of the system will be audited in greater detail, particularly by monitoring intermediate levels of pH decline, and end point measurements of tenderness, purge loss and colour will be collected. These data will be used as inputs to the SSF models and will assist in the refinement of the predictions. Longer term, a number of processing options will be put into the model and the quality predictions will be used to identify processing modifications that should be trialled in each plant to further improve and optimise meat quality.

## **6 Acknowledgements**

Meat & Livestock Australia acknowledges the matching funds provided by the Australian Government to support the research and development detailed in this publication. MLA partnered with Meat and Wool New Zealand and wishes to acknowledge their contribution to the project.

Embedded in computer modelling and shared between M&WNZ and MLA.

IP is shared between MLA and M&WNZ, on the condition that MIRINZ Inc will be acknowledged in any media release or public statement concerning the results of the MLA / M&WNZ collaborative research program.

For each project, the relevant parties will contribute the intellectual property that they own, or are otherwise entitled to provide, that directly relates to the objectives and proposed outcomes of the Program. At the date of this Agreement the parties intend to provide the Background IP arising from the projects listed below:

- P.PSH.0228 [Carcass modelling – Phase 1 (PSHIP.150.07)]
- P.PSH.0264 [Carcass modelling – Phase 2 (PSHIP.150.07B)]