Client Report

Prepared for DEEResearch

August 2003

DEEResearch

Review of processing

N.J. Simmons, D.M. Broda, M.M. Collinson, M.M. Farouk.

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C O N F I D E N T I A L

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Clyde Daly
Team Leader – Meat Science

Inquiries or requests to:
Nicola Simmons
Meat Science
Email: Nicola.simmons@agresearch.co.nz
Phone: +64 7 838 5258
AgResearch -MIRINZ Centre
Private Bag 3123, Hamilton, New Zealand

DEEResearch – Review of Processing

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Contributors:

Processors from North and South Island Venison plants

Deer Industry New Zealand
Graham Brown
John Tacon
Mat Moyes

Abacus Biotech Ltd
Joanne Stevenson-Barry

Agriquality
Alan Cooke

AgResearch
Clyde Daly
Tracey Cummings
Kevin Gilbert
Nick Penney

August 2003

N.J. Simmons, D.M. Broda, M.M. Collinson, M.M. Farouk

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Summary

The DEEResearch Board requested this review of venison processing as one mechanism to assist in establishing funding opportunities and priorities. The scope of the review was defined following consultation with the Venison Processors Technical Committee and other industry representatives. Having defined the scope, the review was prepared by researchers from Agresearch working in the relevant areas, in addition to input and comment from venison processors and other key industry personnel.

The consultation process identified that this review should cover three main areas; the effects of processing on venison quality, opportunities for value-adding to venison and the causes of and potential mechanisms to overcome blown pack-cloistridia based spoilage.

This review has been broadly split into three main sections covering the topics identified above. Within each of these, the topic is discussed in the light of current knowledge, recent key developments in the area, industry practice and opportunities for improvements and further research.

Processing section

In 1991, the Game Industry Board commissioned MIRINZ to develop a processing specification that could be used by industry to produce consistently acceptable tender venison. Specifically, this work set out to define what shear force values would translate into acceptable venison tenderness as judged by consumers. Having set this target, a processing specification was defined that, if followed, would reliably meet this criterion.

This large study lead to the development of industry standards which detailed stunning, stimulation and chilling specifications that would ensure the attainment of the agreed tenderness standard. These specifications undoubtedly resulted in significant improvements in the tenderness of the product being produced commercially and are still being followed by the industry. However, while this work focused on tenderness, other quality attributes were not taken into account.

More recent studies have shown that the effects of processing, particularly electrical stimulation, on quality aspects such as purge loss and colour do not follow exactly the same pattern as seen in beef and lamb and thus further work is clearly needed to understand the mechanisms of the interaction between stimulation and quality attributes. For example, research has demonstrated that, for reasons that are still largely unclear, venison can lose more water in the form of drip, and is far less colour stable than both beef and lamb.

Following industry consultation, a number of recommendations have emerged, the key one being that the industry should consider benchmarking the current quality to identify fluctuations since the last survey in 1991, and to get some indication of quality parameters in addition to tenderness such as purge from the chilled product and colour stability during retail display.

A number of immediate industry issues came to light during the course of this survey. They are as follows: frozen product may not be reaching the tenderness specification and thus some revision to the current processing regimen may be required. The tenderness testing of all product was considered to be inadequate and increased levels of testing to cover both chilled and frozen product was suggested. Furthermore, it was suggested that testing of chilled product should be representative of the markets for which it is intended. That is if chilled product reaches market at any stage from 3 to 4 days or two weeks after slaughter, then testing should occur at these times also.

Recently, processing of beef and lamb has been tailored to ensure that the quality is optimised for the different markets and has resulted in different processing guidelines for chilled, chilled/frozen and frozen beef and lamb. Similar work could be carried out for venison. For example, there are some clear opportunities that should be explored in the area of faster chilling regimes for chilled product destined for the market at two or three weeks following slaughter. In contrast, product that will be in the market within one week of slaughter and product that is frozen within one or two days following slaughter, may require some revision to the conditioning and ageing specification that is applied in the current standards.

Value-adding section

Very little has been published in the area of value-adding to venison. However, this has been quite an active area of research and development for beef and lamb and much of the information and principles can be used as a guide to add value to venison.

From the literature review, several opportunities have emerged around the areas of new packaging systems. One of these is to re-visit the use of CAP technology to extend the shelf life of chilled venison. Additionally, the USDA has recently allowed the inclusion of carbon monoxide in the pack atmospheres of CAP and MAP meat as this system offers significant improvements in colour based shelf life, although market acceptability of this will need to be assessed.

In the public domain there is literature on methods to add value to venison in the form of whole-tissue products, restructured products, coarse and fine emulsion type products and other by-products such as those from antler and blood, skins, tails, pizzles and edible offal. However, a systematic study to show how each major cut or muscle group can be best utilised to add-value to venison may be of value.

“Blown pack”-causing clostridia/spoilage

“Blown pack”-causing clostridia are very prevalent in New Zealand slaughter stock and meat processing plants and thus their control in a meat plant environment is very difficult.

No single control measure has proven effective for controlling this spoilage, although using a combination of control steps, incremental gains in shelf life of the product can be achieved. Most practical measures for reducing or preventing the onset of “blown pack” spoilage must assure that (1) the lowest possible numbers of spores of “blown pack”-causing clostridia are transferred onto the carcass during dressing, and (2) germination and outgrowth of remaining spores are kept to a minimum by maintaining the cold chain throughout product storage, transport and retailing; and by using the optimal regimen for heat shrinking of vacuum-packed product.

On the basis of current knowledge on “blown pack” spoilage control, it is recommended that venison processors should identify processing plant reservoirs for carcass contamination with “blown pack”-causing clostridia. Enhanced control of “blown pack” spoilage would be achieved by regular audits of processing plants affected by this spoilage condition. Further control would be enhanced if industry standards were developed to optimise processing with respect to control of “blown pack” spoilage.

Contents

1 Introduction

Two of the research goals of DEEResearch are to ensure continued and improved access to international markets for New Zealand deer products and to pioneer world-leading processing research to create new technologies for high margin food. In addition, the supporting industry strategy is to provide a premium product for year round consumption that will satisfy existing and new consumers To achieve these goals, a number of objectives must be met:

- food safety standards must be enhanced
- product attributes must be aligned with consumer requirements in existing and new, emerging markets
- new and improved processing, packaging and value adding technologies need to be developed and transferred to industry.

Thus, after consultation with venison processors, it was agreed that the primary aims of this processing review are to identify work to-date and future opportunities, and to suggest a framework by which these can be developed and successfully utilised by the industry.

The key focus areas are as follows;

- Processing impacts on meat quality
- Impacts of production and processing methods on “blown pack”-causing microbial contamination
- Value adding opportunities

To achieve these goals, literature in the area of venison processing and in relevant beef and lamb processing has been reviewed. The initial work to develop the current venison processing standard has been described, along with its uptake by the industry. Current industry best practices were ascertained by the use of a survey from a sample population of processors: this undertaking has provided a critical component of this review by identifying areas of processing where knowledge gaps exist, where market and consumer expectations are now shifting and where quality problems are being perceived.

With these survey data forming a focal point, future areas for research and development have been identified and the discussion has drawn upon similar recent work in beef and lamb processing.

In an effort to identify strategies by which these emerging research and development initiatives may be met, research providers with a track record in venison work, both internationally and within New Zealand, have been identified along with their respective areas of expertise.

2 Processing Effects on Quality

2.1 Current processing based knowledge

This section provides basic explanations on the principles of key meat quality attributes, how they are defined, how they can be modified and some of the sources of variability. Information provided has been drawn from scientific and industry publications and recent research from within Agresearch. The information comes largely from work into red meat quality although the principles are equally valid for venison.

2.1.1 Definition of eating quality

There are many attributes that contribute to the quality of meat and can be defined, most commonly identified as;

- Tenderness
- Texture
- Fat levels
- Drip or purge
- Colour
- Microbiology

These elements of meat quality are based on the visual quality or eating quality or a combination of the two. In other words, meat quality can be understood in terms of effects on appearance, and thus influence the purchase decision, and those that become obvious upon eating. These principles are outlined in Table 1.

Table 1. Attributes that affect product purchase and eating quality	
Appearance (purchase decisions)	**Eating quality (repeat purchase decision)**
Fat-to-lean	Tenderness
Lean colour & fat colour	Juciness/succulence
Purge in pack	Flavour
Price	Texture

The stages of the farm to fork continuum that can influence these quality attributes have been categorized below in Table 2. Clearly, many of these attributes are affected by both production and processing practices. Thus, for any quality initiative to be truly successful, there has to 'buy-in' from farmer, processor, transporter and retailer.

Table 2. The key influences that affect quality as defined above are as follows:				
	Production	Processing	Cold-chain/Ageing	Packaging
Lean colour	√	√	√	√
Fat-to-lean ratio	√			
Purge		√	√	√
Tenderness/texture	√	√	√	
Juiciness/succulence		√	√	√
Flavour	√		√	√

Eating quality is also frequently examined in terms of flavour and overall acceptability of eating quality. However, flavour in particular is largely unaffected by processing *per se* (although prolonged vacuum packed storage has been shown to introduce flavour modifications in most species), so is not covered within this review.

2.1.2 What defines the key quality attributes

This section of the review will focus principally on the impact of the muscle environment during the pre-rigor and subsequent conditioning period. This is the period defined as meat or carcass processing: it is critical in determining the behaviour of the muscle proteins and their subsequent effect on key quality attributes such as tenderness, texture, succulence, drip loss, colour and colour stability. This pre-rigor environment determines the extent of muscle contraction (shortening) during processing, the activity of the meat tenderizing enzymes (proteases), and the degree to which the muscle proteins are disrupted (denatured), an effect that later becomes apparent as the levels of purge (drip loss), and aspects of colour and colour stability during retail display.

Two key variables can be controlled by the processor to manipulate the muscle environment and influence meat quality: these are the rate of chilling and the rate of pH decline through the use of electrical inputs to the carcass to stimulate muscle contraction. A third variable that can contribute to processing specifications is direct physical manipulation of muscle length (stretching muscles). This can be accomplished by different methods of carcass suspension for example tenderstretch or aitch-bone hanging or by stretching muscles mechanically after hot boning. Tenderstretch has had relatively little application in New Zealand and does not appear to have been evaluated for venison.

Although defining specifications on the basis of two processing variables might appear superficially as a relatively simple proposition, in practice there can be considerable complexities to manage: first, the specification needs to be defined for the specific product type. For example, the optimum processing specification for a frozen product is very different from the requirements of a chilled product. Second, the specification needs to accommodate the particular operational requirements for any particular plant, such as chiller storage space or throughput rates. Third, the implementation of the specification needs to be consistent and reliable to ensure some consistency in the quality of the end product.

Processing to produce a high quality product such as venison, and identifying opportunities to create quality advantages and marketing opportunities, means finding the optimum balance between a number of, sometimes conflicting, requirements. These include identifying product quality requirements in specific markets, optimising cost and benefits during processing and understanding the impact of processing events on meat quality attributes. It is particularly this last aspect that this review will

analyse in the context of identifying opportunities for improved quality and value for the venison industry.

Tenderness

The eating quality of meat is still largely dominated by tenderness, an attribute that remains a major concern for all sectors of the red meat industry. Unacceptably tough meat is particularly a feature of frozen product, while tenderness variability between carcasses and cuts continues to be a feature across all species.

As shown in Table 2, a number of factors influence tenderness and they can be split broadly into two categories; direct animal contribution and processing factors. The most important features of the direct animal contribution are the ultimate pH, determined by the level of glycogen in the muscle at the time of slaughter, and by preslaughter growth rates, which influence the behaviour of the tenderizing enzymes after slaughter. However, neither of these factors can be influenced by the processor.

It is generally agreed that processing accounts for between 60 to 70% of tenderness variability (Thompson *pers comm*), while contributions from the animal and cooking make up the remaining 30 to 40% in approximately equal amounts. Generally, the management of tenderness by processing is defined by two features; the degree of muscle shortening and the degree of enzyme-induced tenderisation, commonly referred to as ageing or maturation.

Muscle shortening

The classical studies of Locker and Hagyard (1963) first demonstrated that very rapid chilling causes muscles to shorten and meat to toughen dramatically. Cold shortening occurs when meat is cooled to less than 10°C while the pH of the meat remains above 6.0. This typically causes the muscle to shorten by 20-40% and the meat remains tough even if it is aged for long periods of time. (Davey & Gilbert, 1973).

Very slow cooling combined with high levels of electrical stimulation produce a low muscle pH combined with high muscle temperatures. Although these conditions are exactly opposite to those required of cold shortening, they can produce similar levels of shortening. Termed heat or rigor shortening, this condition produces effects on toughening that are more complex and far less understood than cold shortening: when assessed instrumentally (Tenderometer), heat shortened meat does not appear to toughen and, in fact, can often reach moderately acceptable shear force levels. However, taste panels can describe such meat as tough (Hertzman *et al.*, 1993). Presumably, taste panels are able to detect some aspects of texture associated with

heat shortening that are described as toughness. Or perhaps, because the meat also suffers high levels of drip loss, the dryness and lack of succulence is interpreted as toughness. In any case, even after prolonged ageing, heat shortened meat fails to reach expected levels of tenderness, an effect attributed to the premature loss of proteolytic enzyme activity (see following section).

Ageing

Temperature also plays a major role in determining the level of proteolytic activity or ageing during both the pre- and post-rigor period. Dransfield (1993) developed a theoretical model to describe the activities of the major meat tenderising enzymes, calpains and calpastatin, in response to temperature and used the model to predict meat tenderness. He proposed that high temperatures resulted in an early loss of enzyme activity, so that, although proteolysis is initially rapid, the extent of proteolysis is limited.

In an effort to examine this model empirically and, more specifically, to consider the temperature effects independent of the rate of pH fall, the calpain and calpastatin activity was compared at equivalent pH values in muscles that had been held at three different but constant pre-rigor temperatures (Simmons *et al*, 1996). This work showed that, at the completion of rigor, μ-calpain, the enzyme that has been most closely linked with meat tenderisation, was substantially depleted in muscle held at 35°C, but levels in muscle held at 15°C, and to a lesser extent the 25°C, were largely unchanged from the levels found at slaughter. In this work, Tenderometer measurements at rigor showed that the 35°C samples had lower shear force scores than the 15 and 25°C maintained muscles. In contrast, samples held at 15 and 25°C had lower Tenderometer values than the 35°C samples after 4 days. These effects are characteristic of the heat shortening phenomenon and show how the enzymes involved in tenderisation are affected by temperature: at high temperatures (35°C+), the enzymes are very active and produce tender meat very quickly, but they also lose their activity very quickly. In effect, they ‘burn out’. Hence meat subjected to these processing conditions do not attain the very low shear force values required for high value, high quality markets. At lower temperatures, the balance is shifted towards a longer but slower period of enzyme activity; because the enzymes remain active for longer, due to less burn out, the meat reaches much lower shear force values as long as enough time is given to age.

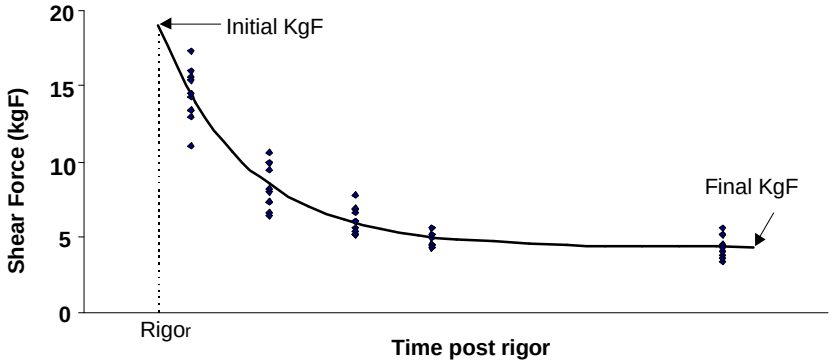


Figure 1. Typical pattern of ageing or tenderisation

The effects of temperature on tenderization are represented at a practical level in Figure 1. This Figure shows the typical pattern of tenderization as measured by cooked shear force testing using a MIRINZ Tenderometer. While this pattern of tenderization is representative of most species, the time scale along the x-axis (time post rigor) to reach the desired end-point of tender meat (typically less than 8 kgf) can be achieved in anything from 5 to 28 days in beef depending upon the pre-rigor temperature and pH environment of the muscle. This demonstrates the enormous impact of processing conditions on the development of tenderness.

Texture

Obviously, if meat is unacceptably tough, then texture is relatively unimportant, but once meat is acceptably tender, other textural attributes relating to juiciness, fragmentation and other characteristics commonly referred to as mouth-feel, become important. Currently, AgResearch, under contract to Meat New Zealand, are developing objective methods for measuring these alternative textural attributes that will enable descriptors such as cohesiveness, fibrousness, softness, succulence and juiciness, which have previously only been described by trained sensory panellists, to be measured instrumentally.

Previous trials using trained texture panels found that meat held at high temperatures pre-rigor often suffers deterioration in specific textural attributes or may develop a different set of dominant textural attributes. Meat that undergoes rigor between 30-35°C was softer and less fibrous compared to samples that had undergone rigor at 15-20°C. Scores for both fibrousness and cohesiveness were significantly different, although the shear force for these samples was not statistically different (Simmons *et al*, 2003). This work was a useful demonstration of how the normally reasonable correlation between shear force and taste panel assessments of tenderness is lost in

meat that has been exposed to high pre-rigor temperatures. Subsequent trials using untrained consumer panels showed that consumers expect a certain textural experience when eating meat, and exposing carcasses to high temperature rigor treatments produces meat that lacks fibrous and cohesive qualities. These characteristics were deemed less acceptable.

These results may also explain, at least in part, why past research has reported that meat processed at high rigor temperatures has shear forces comparable to meat processed at lower rigor temperatures but are perceived as tougher when measured by panellists. Untrained panellists may simply mark such samples as tougher because they find them less acceptable.

Fluid loss

The amount of water in a piece of meat is not fixed, but changes depending on time and how the meat is treated. Fluid losses can occur by evaporation, drip or purge and during cooking.

Evaporation

Water loss by evaporation from the surface of a carcass occurs during chilling, because the water vapour pressure at the warm surface of the meat is far higher than that in the air flowing over it, even if saturated. Typically, a carcass can lose up to 2% of its weight via evaporative losses during chilling. These evaporative losses continue in chilled storage if the atmosphere is not saturated. In addition to the monetary value, evaporation adversely affects the appearance of meat, lowering its acceptability to the consumer. Lovatt *et al* (1992) demonstrated that evaporation dehydrates the surface layers of the meat, although this tends to occur through just a few millimetres at the surface and, therefore, once trimmed, a hydrated surface can be displayed.

Drip or purge

The water holding capacity of meat, or its converse, drip, or purge as it is sometimes known, is of considerable financial importance as well as affecting both the appearance and eating quality of the meat. In conventional processing, drip loss during carcass chilling is usually negligible, although the weight of a carcass can be significantly reduced by evaporative losses. Once the carcass is cut however, drip losses can increase to significant proportions. The drip is commercially significant due to the loss in weight, the poor appearance in the retail pack and loss of eating quality through reduced juiciness.

Drip is affected largely by post mortem processing conditions. Drip is caused primarily by shrinkage of the lattice space that separates the protein fibre responsible for the

contractions of muscle tissue. This shrinkage of the lattice spaces expels water out of the muscle cells and into the space between cells, and ultimately out of the muscle/cut altogether. The level of shrinkage of the fibres is largely determined by the extent to which the contractile proteins denature, a process by which proteins lose their structural integrity. The interaction of pH and temperature in the pre-rigor muscle is a key factor: conditions of low pH and high temperatures are particularly hostile to protein integrity and accelerate the denaturation process. Some muscle protein denaturation will invariably occur during the post mortem period because of the pH fall associated with the development of rigor. However, the extent of denaturation during the pre-rigor pH fall increases with temperature, and becomes particularly severe when high temperatures (>35°C) persist as the muscles near rigor mortis.

While the pre-rigor period is the critical phase in determining how much water will be lost from the meat, it is the post-rigor loosening of the microstructure that allows the water to become mobilised and to manifest itself as drip. This movement of the water through the meat to collect as drip takes time (Honikel *et al*, 1981). In beef, the full expression of water lost as drip occurs somewhere between 30 to 60 days post mortem, while drip loss from lambs can increase up to 12 weeks of chilled storage. Drip lost during ageing tends to correlate negatively with drip lost during retail display and cooking (Simmons *et al,* 1998); in other words, if fluid is lost during storage, it is not available as display or cook loss. Moore (1990) reported a similar relationship between the amount of drip lost during thawing and the drip lost during retail display. Taken collectively, these findings suggest that the extent of losses are determined by processing conditions and will eventually manifest as water loss during ageing and storing, distribution, retail display or cooking. At which of these stages this loss occurs will depend upon processing and additional factors, such as the size of the meat piece and the number of cut surfaces, the packaging conditions, temperature of storage and cooking temperature.

The amount of drip will also increase if meat is frozen and then thawed. This is largely due to the effect of ice crystal formation, which both denatures cell proteins and physically damages the cell, with the effects of both decreasing water binding and accelerating the loss of purge.

Cooking losses

When meat is cooked, an unavoidable and usually substantial shrinkage occurs due to contraction of the collagen component of meat. Such shrinkage physically expels fluid, the magnitude of which can reach 40% by weight. In addition, the shrinking process also results in some toughening of the product, a process which shows three phases

as the cooking temperature increases: an initial phase beginning at about 40°C and levelling off at approximately 55°C, a second phase between 60 and 80°C and a third phase above 80°C (Davey & Gilbert, 1974). The increases in toughness correlate with changes in the distribution of water in the meat and its shrinkage firstly laterally and then longitudinally (Bendall & Restall, 1983).

While some degree of shrinkage is inevitable during cooking, processing conditions can have a significant effect on the extent of the fluid loss and, hence, on eating quality. The key is to ensure that the waterbinding capacity of the meat remains high: by avoiding protein denaturation, water remains tightly bound and resists cook-induced shrinkage and the expulsion of fluid. This effect is particularly evident when meat is cooked at low temperatures (rare to medium), but cooking to high temperatures largely negates the benefits of high waterbinding capacity.

Meat colour

The colour of meat is an important component of meat quality. Although colour will not affect eating quality, consumers often use meat colour at point of purchase to assure themselves that the meat is not spoiled and will be of good eating quality. Meat retailers therefore look for two aspects of meat colour: meat needs to have a bright, fresh appearance and this appearance should be maintained for the required period of retail display.

Making sure that meat looks attractive at retail depends on contributions from both the producer and the processor. By the choice of breed, age at slaughter and nutritional status, the producer will influence meat colour and colour stability by influencing the amount of pigmentation (myoglobin), the composition of fiber type and the amount of antioxidants in a muscle. In turn, through control of the chilling rate and the use of electrical stimulation, the processor can influence the initial ‘bloom’ and colour stability.

The effect of the myoglobin state on meat colour

In living tissue, the normal colour is a bright red because the pigment myoglobin is oxygenated due to the presence of oxygen carried by the blood.

To accomplish its physiological role in oxygen transfer, myoglobin is able to exist in a variety of states, each of which exhibits a different colour.

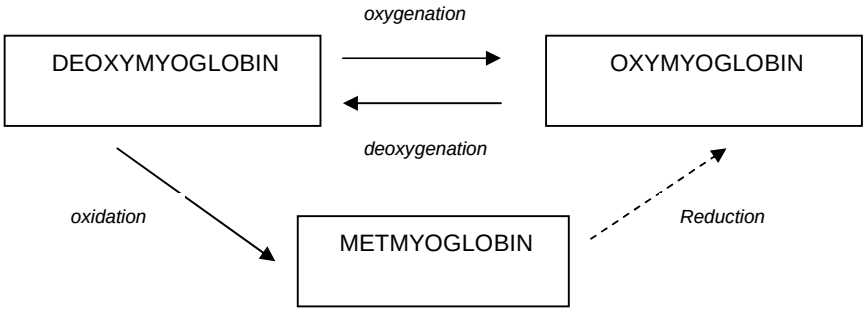


Figure 2: Principles of meat colour

All three versions of myoglobin are reversible and the concentrations of each in a piece of meat are determined mainly by the concentration of oxygen. At moderate oxygen concentrations, above about 10%, bright red oxymyoglobin will dominate, while at low oxygen concentrations at less than 1%, purple deoxymyglobin dominates. The deoxymyoglobin/oxymyoglobin reaction is fully reversible, while the conversion of brown metmyoglobin back to purple deoxymyoglobin is a process that requires an enzyme, referred to as a reductase (Figure 2).

The rate at which meat turns brown during retail display is a critical attribute for meat retailers, and this depends on the balance between how quickly the brown metmyoglobin forms and how quickly it is reverted back to the myoglobin form by the reductase enzyme.

The rate at which metmyoglobin forms depends on two main factors. The first is the level of antioxidants in the meat: increasing antioxidant levels reduces the rate of myoglobin oxidation. Hence, animal supplementation with, for example, vitamin E can improve retail colour stability.

The second contributing factor is the extent of oxygenation of the surface layers, which determines the proportion of myoglobin in the oxymyoglobin form. When this proportion is high, the development of the brown metmyoglobin is inhibited and colour stability is increased. This is the principle of the modified atmosphere packaging systems, where 80% oxygen atmospheres are used to produce the complete conversion of myoglobin on the surface layer of meat to the oxymyoglobin form.

The second aspect of metmyoglobin formation is the survival of the reductase enzyme responsible for converting the brown metmyoglobin back to myoglobin. Unfortunately, the activity of this enzyme is gradually lost with increasing time post-mortem. Therefore, increasing storage times, beneficial for tenderness, also has the effect of reducing retail display life.

Processing conditions also affect the survival of the reductase enzyme. Processing conditions that result in low muscle pH while the muscle temperature is still high (denaturing conditions – see the drip loss section) damage the enzyme and reduce the ability of meat to return the brown metmyoglobin back to myoglobin. Retail colour stability is therefore reduced.

The survival of the reductase enzyme during storage and retail is highly temperature dependent and effective temperature control in the cold chain is vital to ensuring good retail display life. An increase in retail display temperature from 0 to 10°C increases the rate of browning by 2-5 fold.

Surface reflectance

In the earlier section discussing fluid loss, the mechanism of muscle lattice shrinkage as it affects drip was briefly described. In essence, when the pH falls quickly while the muscle is still at a high temperature in the period soon after slaughter, the protein filaments that produce contraction are disrupted or denatured and 'shrink'. This results in a reduced amount of space within the protein lattice and the water is expelled as drip. This mechanism also results in an increase in the paleness of the meat. This is because the reduced lattice spacing within the muscle causes more light to be reflected from the meat surface, imparting a whiter/paler appearance. Overall therefore, inappropriate processing conditions that involve a rapid pH fall while temperatures are still high can give rise to a pale muscle appearance that also loses large amounts of drip and, because of less reductase enzyme, has reduced shelf life.

Electrical stimulation to modify meat quality

Originally, electrical stimulation of lamb carcasses was developed to overcome cold-shortening (Chrystall & Hagyard, 1976). The contraction of the muscles in response to the electrical current caused the pH to drop quickly during the period of stimulation, and the extent of this drop depended to some extent upon the parameters of the stimulation current, such as duration, current levels and frequency. In the original specification for rapid processing of lamb, stimulation was used to drop the pH to 6.2, which would then allow the carcass to be exposed to a rapid chill without the risk of cold-shortening (Davey & Chrystall, 1980).

Later, it was shown that electrical stimulation accelerated post mortem tenderisation (Chrystall *et al,* 1982). This effect is based on two principles: first, ageing starts when muscle reaches rigor mortis; and, second, the rate of ageing increases with increasing temperature. Because stimulation accelerates the onset of rigor mortis, ageing starts sooner and, because the carcass will not have cooled as much, ageing will progress more quickly. The overall effect of this is that the rate of tenderisation is quicker in the early post mortem period when compared to non-stimulated counterparts.

Although most studies report a positive effect of electrical stimulation, some have shown that stimulation, when combined with slow chilling, may have an adverse effect on tenderness (Geesink et al, 2001; Marsh et al, 1987; Unruh et al, 1986), water holding capacity (Hertog-Mieschke et al, 1996) and colour (Ledward et al, 1986). Unfortunately, processors often enthusiastically stimulate carcasses in the belief that if a little stimulation is good, a lot must be better, and this has given rise to many quality problems, particularly in the overseas chilled meat trade.

The negative effects of electrical stimulation on tenderness are particularly complex. The rapid pH fall while the carcass temperature is still high can result in either tougher carcasses (Pike et al, 1993) or significant initial tenderisation which is followed by a reduced ability to age subsequently, so that the ultimate tenderness of the meat is compromised (Simmons et al, 1996).

These same conditions also result in high levels of protein denaturation and reduce the meat's ability to bind water, giving rise to increased drip (Hertog-Meischke et al, 1996, Eikelenboom & Smulders, 1986) and cook loss. These in turn affect both the appearance of the meat in the retail display and succulence and juiciness of the product upon consumption. The pre-rigor phase is the period when the proteins are most susceptible to denaturation and is the most significant period in 'setting the scene' for subsequent meat quality. Thus, if proteins are exposed to unfavourable conditions (low pH/high temperature) during the pre-rigor period, drip losses are likely to be significant at some stage during storage.

There is still some debate on the effect of electrical stimulation on the colour of beef and lamb. It is generally agreed that stimulation improves the initial colour, giving it a lighter redder colour when compared to unstimulated counterparts (Hector et al, 1992). However, after extended retail display, the brown metmyoglobin levels are more marked in the stimulated muscles (Ledward, 1985). These effects tend to become more significant if the meat is well aged prior to retail display.

Recent studies at AgResearch, funded largely by Meat New Zealand and Foundation of Research, Science and Technology, have shown that electrical stimulation should remain an important component of processing for many markets. However, avoiding deleterious effects on quality and, taken a step further, optimising quality for different markets, involves judicious tailoring of stimulation parameters (some combination of duration, current magnitude, waveform, electrode position and time of application of the stimulation current post mortem) in combination with chilling. These opportunities are discussed later in this review.

2.1.3 Principle sources of meat quality variability

Tenderness variability independent of ultimate pH effects

Animal Effects

Tenderness variability associated with different ageing rates was identified in section 2.1.2. In section 2.2.2, the generation of different rates of ageing or tenderisation are clearly linked to different processing scenarios and the principles outlining how processing can be used to modify ageing rates allowing 80% of tenderisation to occur either within the first week of slaughter or to reduce the rates to a level where it takes almost 4 weeks to get to the same level, will be described. However, in addition to a processing driven ageing rate, there are generally large differences between animals. There are a myriad of papers that have reported on tenderness and its variability between carcasses for all species. Some of this work has identified differences between breed, age, sex, production and processing systems, but the reality remains that the high levels of variability between animals remains to be properly understood.

A major problem with many of these studies has been the use of unstandardised processing regimes: unless differences in the pH response to stimulation, individual carcass cooling rates (affected, as it is, by carcass weight) and ultimate pH are properly accounted for, distinguishing animal effects from post mortem processing effects becomes impossible. A highly standardised procedure has been developed at AgResearch to address these issues and was used in a large scale genetic study in beef (Daly, 2000) utilizing 273 limousin X jersey cattle that formed the subjects of a QTL trial (Morris *et al,* 2000). First, mechanical stunning was used instead of electrical stunning and electrical stimulation was avoided as both these electrical inputs can generate quite different pH responses between animals. Second, the samples were maintained at constant temperatures within 30 minutes of slaughter while the muscle was still in the early pre-rigor period. Third, the pH was monitored through the pre-

rigor period to define when the ultimate pH was reached and thus the start of tenderisation. Fourth, all samples with an ultimate pH greater than 5.7 were excluded. Samples were then tenderness tested at rigor and subsequently at intervals during ageing under controlled temperature. This procedure allows an ageing rate to be calculated for each animal, as demonstrated in Figure 1 (Section 2.1.2).

Despite these highly controlled conditions, this trial illustrates the wide variation in tenderness: The initial shear force ranged from 8 to 25 kgf and, after complete ageing, the range was still large.

Immediate pre-slaughter stress

Pre-slaughter stress is recognized as having an important effect on meat quality, although these effects are usually attributed to an increase in the ultimate pH (see later section). However, recent research results and some commercial data with lambs have shown that acute physical stress immediately before slaughter can produce toughness without a change in the ultimate pH of the meat (Daly et al, 1995, Morton et al, 1997, Simmons & Collinson, 2003, in preparation). These effects only become apparent when the pre-slaughter stress is combined with some level of electrical stimulation, and, in the experimental studies cited above, these effects were more pronounced when low voltage electrical stimulation was used. While the pH fall during and after stimulation was comparable between the stressed and unstressed animals, the shear force values when measured during the ageing period, was always higher in the pre-slaughter stressed and stimulated group.

Effect of freezing on tenderness variability

Freezing meat generally results in an improvement in tenderness when compared to meat that has been aged for an equivalent period but has not been frozen (Whipple & Koohmaraie, 1992). However, while freezing with and without subsequent thawing does have a tenderising effect on cooked shear force, these effects are significant when the meat has had limited ageing, but become less marked with increases in ageing (Collinson, 2003). This suggests that the tenderisation effect of freezing, may be due in part, to the physical disruption imparted by the inter- and intra-cellular ice-crystal formation. This may also explain the relatively large impact of freezing on the partially aged muscles compared to the reduced effect in well-aged muscle where proteolysis has already caused myofibrillar weakness and thus ice-crystal disruption has little additive effect.

Therefore, while freezing can improve tenderness per se, tenderness variability will still be a feature of frozen product due to differences in ageing rates between carcasses prior to freezing.

Effect of increased ultimate pH on tenderness variability

The pH fall that occurs post slaughter is due to the conversion of muscle glycogen to lactate. If sufficient glycogen is present in the muscle at slaughter, the ultimate pH (pHu) should fall to approximately 5.5 to 5.6. pHu will be higher if the initial preslaughter stores of muscle glycogen are low. Undoubtedly, pHu is due to a multiplicity of causes and, to compound the matter further, the responses to individual stressors are different for each species. For example, dogs are identified as a predator by sheep, but are a mere annoyance to cattle. Mixing of cattle and pigs before slaughter leads to high levels of physical and psychological stress, but mixing does not appear to affect sheep (Lacourt and Tarrant, 1985). Thus eliminating pHu requires a highly coordinated effort from the producer right through to the processor. While this is not an easy undertaking, many processors are now recognising the various quality problems associated with high pH meat and are thus measuring and identifying high pH carcasses in the chiller the day following slaughter.

Quality problems associated with high pHu can be split into two categories: the first is the phenomenon of intermediate pHu-associated toughness, which occurs typically in product with a pHu within the range of 5.8 to 6.0; the second problem is the poor storage life, evident when the pHu exceeds approximately 6.0.

Intermediate pH toughness

A relationship has been shown to exist between pHu and cooked meat tenderness in both beef (Purchase & Aungsupakorn, 1993), lamb (Watanabe et al, 1995) and more recently venison (Stevenson-Barry et al, 1999). These studies have shown that toughness can be markedly increased in meat when pHu lies between 5.8 and 6.0 when compared with normal and high pH meat.

In lamb, toughness related to intermediate pH has been shown to reduce during ageing, so that it eventually reaches an acceptable tenderness level albeit at a slower rate than normal pH lamb (Watanabe et al, 1995). However in venison and beef, this toughness has been shown to persist despite ageing of up to 3 and 6 weeks respectively (Stevenson-Barry et al 1999; Simmons & Cairney, 1997). Benchmarking studies of prime beef quality from export plants has shown that, at certain times of the year, the incidence of intermediate pH can rise to up to 17% of which 7% suffer from the enduring toughness associated with this pH range (Simmons & Gilbert, in press). The incidence of intermediate pH in venison is not currently known.

Ultimate pH above 6.0

In general terms, meat with a high ultimate pH appears darker. This is because the light scattering ability of the meat diminishes once the pH is greater than 6.0

(MacDougall, 1982); the muscle filaments in meat remain more open at high pH, as it is in pre-rigor meat, resulting in a greater translucency and thus darker appearance. The associated physical conditions of firmness and dryness are also associated with this more open spacing of the muscle filaments.

In addition to the poor appearance of high pH meat, it also suffers from much higher spoilage rates. Under anaerobic conditions - for example, vacuum packed product - meat that has a pHu of 5.8 or more typically has much higher numbers of both the microorganisms *brochrothix thermosphacta* and *enterobacteriaceae*. As there is no glycogen in the meat, the microorganisms are forced into the early utilisation of amino acids (the building blocks of protein) because of less carbohydrate substrate, with consequent production of malodorous by-products.

2.2 Recent key developments in red meat processing

This stage of the review outlines recent work on process modifications to improve quality in red meat. The work in this next section was carried out at MIRINZ (now AgResearch), and used revenue from Foundation of Research Science and Technology (FRST) to benchmark meat quality from lamb and beef processing plants throughout New Zealand. While this was occurring, experimental work back in the laboratory was providing a greater level of understanding regarding the impacts of the pre-rigor muscle environment on the development of meat quality attributes, in particular tenderness development and the muscles' water holding capacity. This information, in combination with work from other laboratories was used to underpin new processing guidelines for both beef and lamb processors. The funds for this work were provided by Meat New Zealand and over a 3 year period experimental and commercial trials were conducted utilising standard processing practices such as stunning, immobilisation, stimulation and chilling, culminating in a complete revision of the AC & A process for lamb (accelerated conditioning and ageing) and the provision of multi-faceted processing guidelines for beef and lamb.

2.2.1 Benchmarking lamb tenderness

Two years ago, AgResearch undertook a study to measure tenderness from lambs processed in both North and South Island export plants. All plants were operating an audited AC & A process (accelerated ageing and conditioning), consisting of halal stun followed by low voltage immobilisation and spinal discharge, followed by high voltage stimulation applied to the dressed carcass and finally leading to a conditioning period on a cooling floor operating under tight chilling specifications. The original AC & A specification was designed for frozen lamb and was intended to ensure that, when lamb was frozen at 24 hours post mortem, the shear force was within the defined specification of 95% of all bites to be less than 11 kgf with a mean of 8 kgf.

During this benchmarking exercise, the shear force of these samples was measured at 48 hours post slaughter, so the meat had an additional 24 hours of ageing above and beyond the amount required before freezing according to the AC&A specification. This survey found that the tenderness requirements of the AC&A specification were not being met consistently. Although the mean shear force for all samples over the course of a year was 8.1, almost meeting the mean tenderness specification (mean of 8 kgf or less), the percentage of samples whose tenderness value was less than 11 kgf was only 82%, falling well short of the specification. These findings were particularly worrying since all samples had undergone more aging than specified, and therefore should have been more tender than the specification.

This survey and other commercial and experimental studies conducted at the same time found that a number of factors affect whether meat will conform to quality specifications. The most important of these factors are as follows:

- Genetics
- Seasonal effects
- Stress (on-farm, during transportation)
- Electrical inputs during processing
- Chilling regime

Out of these factors, only the last two can be fully controlled by the processor.

To accommodate the change in animals over the last 10 years (due to genetics and production systems), as well as new requirements in processing and shifts in product quality requirements, a thorough re-evaluation of the AC&A process specifications became appropriate.

2.2.2 Process optimisation

New Zealand work - Processing Guidelines produced by Meat New Zealand

AgResearch used underpinning understanding of the impacts of the pre-rigor muscle environment on the resultant meat quality as a basic starting point (some of these are outlined in the earlier section). The next step was then to utilise some of the features of processing such as stunning, immobilisation, stimulation and chilling procedures to control the pre-rigor environment of the muscle to generate different meat quality outcomes. This was defined in terms of rates of rigor onset.

To illustrate this, two contrasting processing regimes were applied to beef carcasses in an effort to generate meat quality attributes suitable for a chilled overseas product and a chilled product destined for the local market. The aim of these processes was to generate two very different rates of rigor development, and measure the differences in the meat quality of some of the major muscles after different periods of chilled storage. The slow rigor process consisted of a captive bolt stun with no subsequent electrical inputs followed by chilling at 4°C for 24 hours. In contrast, the fast rigor process consisted of head-only electrical stun (Halal) with 15 seconds of low voltage immobilisation (180mA) and high voltage stimulation (1130 Volts for 90 seconds) applied to the dressed carcass at 30 minutes post-mortem; chilling was then controlled at 10°C for 15 hours then 4°C until 24 hours. These two processes were selected to ensure that the temperature of the m. *longissimus dorsi* (LD) from 6.2 to rigor (5.6)

was between 10 and 6 °C for the slow rigor process (chill export product) and 30 and 24°C for the fast rigor process (chill local product).

The fast rigor process resulted in significantly lower shear force values in the LD muscles than the slow rigor process. From the fast rigor process, the LD was considered as a highly acceptable product (as judged by consumer panels) after 7 days of ageing while the slow rigor samples took 21 days to reach equivalent scores. However, after 60 days of storage the tenderisation process in the slow rigor samples had progressed further and generated lower shear force scores than the fast rigor samples. For two other muscles, the *M*. *Biceps Femoris* and *Semimembranosus* the shear force was unaffected by the rate of rigor onset, but, compared with the slow rigor treatment, fast rigor resulted in significantly higher purge and cooking losses, and colour stability was reduced during simulated retail display compared to the slow rigor treatment.

This work illustrated some of the complexities associated with process tailoring: The effects of pH/temperature profiles on both proteolytic activity and protein denaturation are reasonably well understood and this has now been taken further to develop a series of temperature/pH profile targets for different markets.

For example, a processor preparing meat destined for retail within 7 days of slaughter requires tender meat within a few days. Frozen product also needs to tenderise quickly. In contrast, a chilled meat exporter has the advantage of anything from two to six weeks of chilled storage during transit in which ageing can take place and needs product that will remain stable for prolonged periods. The knowledge of how processing can be manipulated to generate an array of pre-rigor pH/temperature profiles in the muscle creates the opportunity to tailor processing for specific market requirements.

In Figure 3 we have tried to represent this concept diagrammatically. Essentially, any process tailoring should firstly be avoiding either cold-shortened meat or PSE (pale soft and exudative meat).

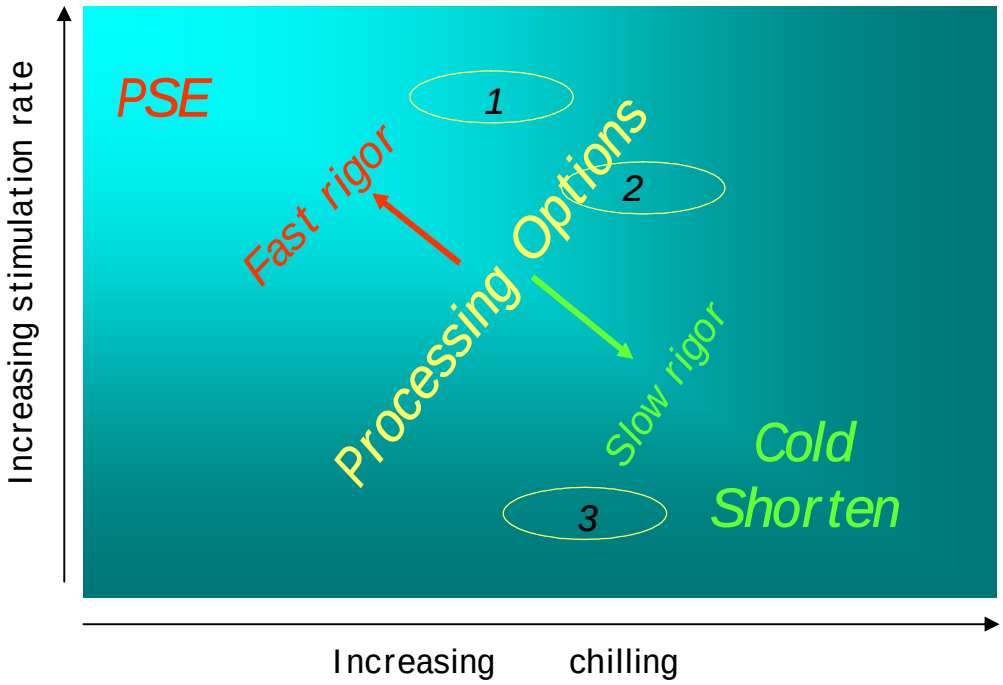


Figure 3. Process optimization principles

1 = Target area for frozen product

2 = Target area for chilledl/frozen product

3 = Target area for chilled/chilled product

PSE or pale soft and exudative meat is a term borrowed from the pig industry and describes meat that is very pale and loses a lot of drip. While this term is most closely associated with stress sensitive pigs and the manner in which they respond to slaughter, it has also been reported in both beef (Simmons et al, 1998) and venison (J. Stevenson-Barry, *pers comm*.).

Clearly, processes that generate either cold-shortened or PSE are to be avoided, but, between these two extremes, the scope for process tailoring is still vast.

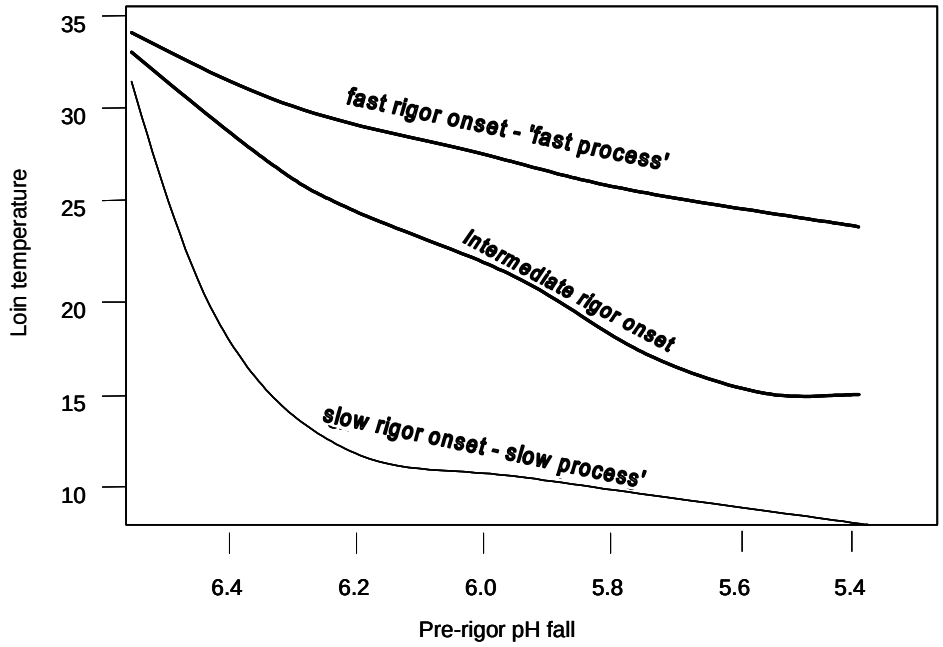


Figure 4. pH/temperature profile for contrasting rates of rigor onset

In very broad terms, a process tailored for a frozen product requires a fast rigor onset. Whereas, a process tailored for a 6-week chilled product will require a slow rigor onset. The pH/temperature profile of the LD of such carcasses has been shown in Figure 4 above.

Release to industry

This work has culminated in new processing guidelines which were released by Meat New Zealand to the red meat industry last year. These guidelines cover processes for chilled/chilled, chilled/frozen and local trade beef and lamb, a summary of the likely meat quality outcomes for the processes and quality critical control points to enable plants to internally audit the new procedures.

This work has never been carried out anywhere else in the world and is now forming the basis of process optimisations which are carried out by AgResearch in the UK, Ireland and the USA.

Australian work- Meat Standards Australia

The Meat Standards Australia (MSA) beef quality scheme is based on identifying possible pathways leading to defined quality standards. The pathways include both production and processing options. Examples of production pathways include animal age and the presence of *Bos indicus* genetics and growth rates, while

processing pathways include the use of electrical stimulation, carcass suspension techniques and ageing rates. The interaction of a number of different production and processing variables has been combined to produce a model that allows quality to be predicted.

The quality standards are based on 3, 4 and 5 stars, representing everyday, above average and excellent eating experiences, and the grading has been developed using consumer tests. The scoring system has been subdivided to include different meal types – for example frying, roasting, stir fry – and different muscles have been graded, at the end of each pathway, for their quality with respect to each meat type.

This remarkable database allows rational decisions to be made on how to link eating quality with production processing systems, and how to quantify the quality attribute, and therefore, potential value, through production and processing methods. By comparison to the research programme in New Zealand (see above), there has been considerably more emphasis on the contribution of production to quality and less on the contribution of processing. In part, the wide range of environmental conditions, which are also often very difficult, and the presence of *Bos indicus* genetics, which are recognised to produce tougher meat, may be contributing to the different philosophy. However, the extensive use of consumer panels, the wide range of muscle analysed and the shear scale of the undertaking has resulted in a remarkably comprehensive database. The cost has also been commensurately great and probably outside the scope of smaller sectors of the meat industry; the MSA development may however provide the model for a more targeted approach to understanding and predicting the relationships between production and processing. More information on this can be obtained from Meat & Livestock Australia (www.mla.com.au)

Techniques to reduce the variability associated with low voltage stimulation

Low voltage stimulation has formed a pivotal part of the process in the majority of the new processing guidelines discussed above. However, in all species, low voltage stimulation tends to generate a far more variable rate of pH fall both within and between carcasses, when compared to high voltage stimulation. The variable rate of pH fall obviously results in different times to rigor onset and thus variable tenderness levels between carcasses during the first week of maturation. While these effects tend to reduce less once ageing has increased to 2 or 3 weeks, it is obviously an issue for

frozen or chilled/frozen product that has only a limited period of ageing before freezing.

To overcome these problems, AgResearch under contract to Meat New Zealand and Meat & Livestock Australia, have developed a new low voltage system that measures the carcass response to the stimulation and then automatically modifies the subsequent stimulation parameters to produce the desired end-point. In this way, the stimulation process is tailored on a carcass-by-carcass basis. This not only generates a highly flexible process and less variable product, but can also be used to predict ultimate pH and tenderness.

This process is currently undergoing commercial testing in both beef and lamb plants and will be available for industry uptake during 2004.

2.3 Current venison processing knowledge

The next section of this review focuses on current knowledge of venison processing. It identifies all the key publications in the area from immediate pre-slaughter handling to processing and the effects of these procedures on venison quality. This work has been evaluated in light of the principles defined in section 2.1 and their application to beef and lamb process optimisation as covered in section 2.2. Thus, wherever possible, we have attempted to evaluate the publications in this section based upon the muscle pH and temperature environments that have been generated by the treatments and focus on the impacts of these on venison quality outcomes.

2.3.1 Effects of immediate pre-slaughter procedures on venison quality

Activity in lairage

Very little is known about the extent of bruising in commercially slaughtered deer and even less about the cause of this damage. Most research (Jago *et al.*, 1996; Pollard *et al.*, 1998; Pollard *et al.*, 1999) has studied the effect of transportation on behaviour and venison quality. Research by Pollard (1999) has incorporated some observations of behaviour in lairage. The deer slaughter premises (DSPs) where the research was carried out, Otago venison, have facilities specifically designed for deer lairage. In contrast, some other DSPs are currently using cattle, sheep or pig facilities, and the absence of specialist facilities may aggravate stress and bruising. Lairage involves a range of potential stressors, including close confinement with unknown deer, human presence, unfamiliar environment and movement through small areas. Even minor

handling of deer can result in increase in physiological stress indicators (Ingram *et al.*, 1997 cited by Pollard *et al.*(2000).

Selwyn and Hathaway (1990) surveyed three DSPs and found that between 1.3% and 9.8% of carcasses were wounded or bruised, with the consequences of trauma as the most common reason for downgrading. McCausland and Miller (1982; cited by Jago *et al*., 1996) found that at least 43% of all bruises occurred after the animals had arrived at the DSP. However Jago *et al*. (1996) found that the level of bruising on animals held overnight was not significantly different from that of deer slaughtered on the day of arrival.

Stevenson-Barry (2000) recorded the behaviour of deer at a DSP and observed an average of 14 antagonistic encounters/hour with a minimum of 3/hour and maximum of 34/hour. The level of encounters tended to increase during the night. In the lead-in race rearing, jumping and lying down were observed (Pollard *et al.*, 1999; Stevenson Barry, 2000). Deer that showed these behaviours were more likely to have been held in the race for a long time rather than for short periods. They concluded that there was no indication of a relationship between ultimate pH and fighting or agitation in lairage or the lead-in race. The results of Wiklund *et al.* (2001) support these findings.

A recommended design layout for deer lairage is needed. Processors have tended to base designs on experience, precedence and/or word of mouth. However, research is needed to define the implications of using lairage designed for other species, especially smaller animals, on stress and venison quality.

To our knowledge, there have been no similar studies to those reported in section 2.1.3. on the effects of pre-slaughter stress, independent of increased ultimate pH, conducted with venison. However, as even minor handling of deer can result in an increase in physiological stress indicators (Ingram *et al.*, 1997 cited by Pollard *et al.*, 2000), it is highly likely that some sources of tenderness variability in venison may lie in the interaction between pre-slaughter stress and subsequent electrical stimulation. In addition to stress-induced variability in venison quality, differences in tenderness due to breed (Shaw, 2000), age and sex (Stevenson-Barry et al, 1992 & 1999; Mojto et al, 1992) and production systems (Forss et al, 1979; Manley & Forss, 1982; Volpelli, et al, 2002) have also been demonstrated.

Pre-slaughter stunning

Halal electrical stunning has been used in the slaughter of sheep and cattle for many years in New Zealand. This method of pre-slaughter stunning was accepted by the Muslim community in 1982 when it was demonstrated that the animals could be rendered unconscious by head-only electrical stunning techniques but that the animal

would recover and behave quite normally if given sufficient time after the period of the stun. Because the animals recover fully if they are not bled, carcasses can be certified Halal and head-only stunning has been adopted in most of the export killing plants in New Zealand. Because the Halal meat can be exported into any Muslim area of the world, Halal slaughter is an important market initiative.

The majority of venison plants in New Zealand use captive bolt stunning. This method invariably results in severe damage to the brain and animals will not recover. The technique is therefore not allowed under Muslim law. However, as with beef and lamb, there is some potential in adopting head-only electrical stunning for deer and thus allowing new markets for venison to develop in Muslim dominated countries.

There is limited information on the effects of head-only electrical stunning in deer. In 1994, Cook et al demonstrated that currents of 1 to 1.3 amps for 4 seconds produced an effective stun as judged by the presence of an epileptic seizure. The seizure, and therefore unconsciousness, lasted from 58 to 68 seconds, enough time to allow the animals to be bled and die humanely.

The animals' reactions were similar to those seen in cattle and sheep following head-only electrical sunning, with typical tonic limbs, neck and head extension and body rigidity followed by a period of clonic activity. The clonic kicking movements developed between 20 and 30 seconds after the stun. They concluded that, as long as the current levels were as defined above and the animals were stuck within 20 seconds of the end of the stun, the act of head-only electrical stunning in deer was humane and could be used in a commercial environment. (Cook *et al.*, 1994a; Cook *et al*, 1994b).

However, the same authors recommended that if carried out in a commercial environment, there must be adequate head restraint to ensure accurate positioning of the stunning electrodes and ensure that adequate current passes through the brain. Also, the neck cut must be monitored to ensure severance of the carotid artery and thus a quick and efficient loss of blood, thereby ensuring death of the animal occurs while the effects of the stun endured (Cook *et al.*, 1994b).

Unfortunately, head-only stunning of cattle and deer can cause high levels of blood splash. This can range from discrete blood spots in the muscle to areas of widespread bruising (Gilbert, 1993). This problem only becomes severe if sticking is delayed and if the Halal cut is not quickly followed by a thoracic stick (Gilbert, 1993). Similar principles have been shown to exist in venison; Mulley (1999) studied blood splash in deer and found that, when the interval between stunning and sticking was greater than 5 seconds, a thoracic stick significantly reduced the incidence of blood splash for electrical and captive bolt stunning. The thoracic stick method was also more effective

at preventing blood splash than the gash cut method, regardless of the stunning method used.

Thus, from the work to date, it seems feasible for head-only electrical Halal stunning to be introduced. However, the corollary to this is that following the stun, sticking should be carried out within 5 to 10 seconds of the stun end followed by a thoracic stick before subsequent work-up.

While in principle Halal stunning can be introduced into venison processing, it should be based on the realisation that subsequent processing may be affected by this electrical input and thus some downstream processing modification may be required. A salutary reminder of this can be found in the export lamb process; the original AC & A process was developed based upon a gash cut of the lambs with no further electrical intervention until the high voltage tunnel after dressing. Electrical stunning was introduced to address new standards of animal welfare. In order to control post-slaughter convulsive activity and allow faster line speeds and to avoid worker injury, electrical currents were applied after sticking to immobilise the carcass and then sometime later, an additional burst of high voltage current (spinal discharge) was applied for the same reason. These procedures were grafted onto the AC & A process without considering the impacts on product quality. It was not until the quality was benchmarked some years later that the carcass response to these electrical interactions was measured and it was found that meat quality had actually deteriorated.

2.3.2 Post-slaughter treatments – effect on quality

Venison processing literature available in the public domain has been summarised in Table 3. While other literature on venison quality exists, these have generally concentrated on comparing different production-based issues on quality (breeds, sex, age, production system etc). While some of these references have been identified in this review, the primary purpose of this review is to identify the effects of processing on venison quality. It is clear from Table 3 that literature relevant to venison processing is limited.

The literature has been selected on the basis of addressing different processing options to identify effects on meat quality. The table identifies the scope of each paper in terms of electrical stimulation, chilling and ageing parameters, packaging technologies and then the meat quality outcomes that have been measured. It is intended that this table can serve as a reference source for anyone wishing to look at specific processing topics in detail.

Table 3 Processing for Quality References

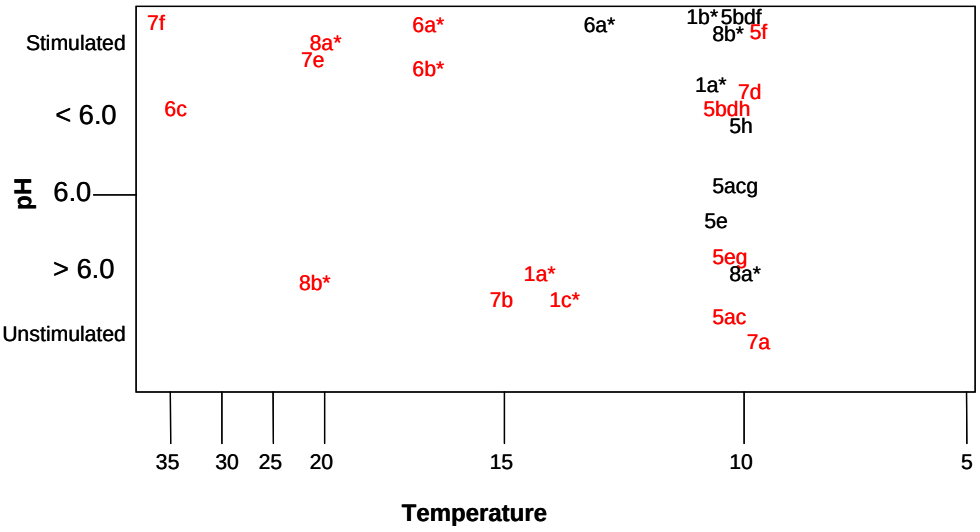
Ref	Ref type	Client	ES	Chilling	Ageing	Freeze Thaw	VP	CO2	MAP	Micro	pH	Kgf	WHC	Colour	Sensory	Comments
1	pub		+/- lvs	10 or 2°C	48 or 72	-	-	-	-	-	pr	+	-	-	-	
2	pub		lvs/hvs	-	-	-	-	-	-	-	-	-	-	-	-	overview
3	Pub		lvs	Combinations of 10 and 0°C	1, 2 & 4 days	+	-	-	-	-	-	+	-	-	-	
4	pub		+ ?	10°C 2hrs 0 22hrs	-	+	-	-	-	-	-	+	-	-	-	
5	pub		lvs	0 or 10°C	0,1 & 3 days	-18°C	-	-	-		pr	+	-	-	-	
6*	cri	NZGIB	hvs/lvs	3°C 4 hrs/10°C 4 hrs	1 d, 3 weeks	-	+	+	-	-	-	+	-	-	+	Several trials carried out
7	cri	NZGIB	+/- lvs	Variable	1 d, 3 weeks	-	+	-	-	-	pHu	+	+	+	+	Chilling rates assessed at 5 hours pm
8	pub		+/- lvs	commercial	1,3,6,12 weeks	-	+	-	-	-	pr/pHu	+	+	+	-	
9	cri	NZGIB	lvs	10°C 10 hrs then 2°C 14 hrs, 2°C 24 hrs , 0°C 24 hrs	13 weeks		+	-	-	+	pHu	+	+	+		Air and spray wash chills

6* - original cervena process work (1991)

1. Electrical stimulation of deer carcasses (1981). Chrystall et al. NZ Journal of Agr. Res. Vol 26, 89.
2. Tenderness by stimulation (1982). Aylward. Food Tech. in NZ, May, 25.
3. Matching tenderness with a price-tag (1984). Drew, K.R., Crosbie, S.F., Forss, D.A. The Deer Farmer. Summer, 1984
4. Farmed venison slaughter and processing (1985/86). Drew et al. Agricultural research division annual report
5. Electrical stimulation and ageing of carcasses from red, fallow and NZ Wapiti-type male deer (1988). Drew et al, Meat Science, 43, 245
6. Venison processing for optimum tenderness (1991). Gilbert, K.V., Fraser, E.M., Devine, C., Chrystall, B.B. Confidential report for the GIB (CR 279)
7. Influence of post-mortem chilling rate and pH on water binding and meat quality in venison (1997). Daly, C.C., Cairney, J. M. Confidential report for the GIB (#CR 603)
8. Electrical stimulation of red deer carcasses – Effects on pH decline, meat tenderness, colour stability and water-holding capacity (2001). Wiklund et al. Meat Science, 59, 211.
9. Chilling regimes to minimise moisture loss (1998). Merts, I. Hill, H.K, Chadderton, T. Confidential report for the GIB (CR 687)

While other references have discussed stimulation in a general sense, those cited in this table are the only ones that detail the processing conditions and their effects on quality outcomes. This section therefore compares and summarises the effects of stimulation, chilling and ageing on venison quality, and will compare how these responses in venison may differ from those reported in beef and lamb.

Figure 5 analyses the publications in respect of the two major process variables, cooling and the rate of change in the muscle pH. Each suffix in this figure is a reference as identified in the table above. By reducing the analysis to these two components it is possible to describe in a graphical format (with some approximation) the various processing conditions used in the research reports. The two axes of the figure therefore describe the rate of pH and temperature decline post-mortem and, in theory, any venison processing specification can be positioned accordingly. The units used for the axes are somewhat arbitrary and are based on the temperature of the loin at either 5 or 8 hours after slaughter and the pH in the loin muscle at 1 hour. Unfortunately, the majority of papers do not report either pre-rigor pH's or actual muscle temperature, choosing instead to report chiller setting temperatures. Therefore, Figure 5 is based on data from just 5 papers. Refer to table 3 for details of each reference.



• pH in the loin muscle at 1 hour after slaughter
• Temperature of the loin 5 or 8 hours after slaughter

* - loin temp and pH decline. Others – pH fall modelled and predicted values given

Figure 5. Venison references evaluated in terms of pH fall and temperature

Up until quite recently, all work examining the effect of processing on venison quality has focused on the use of low voltage stimulation to improve tenderness. The first major study of this kind was conducted in 1981 by Chrystall and co-workers at MIRINZ. In this study, low voltage stimulation was applied to deer carcasses for 90 seconds within 30 seconds of slaughter. This treatment was combined with three chilling regimes utilising different combinations of 10 and 2°C for periods of 1 to 3 days post mortem. Electrical stimulation resulted in a significant improvement in tenderness compared to the non-stimulated counterparts for all chilling treatments, producing a shear force of 10.7 kgf after 48 hours at 10°C followed by a further two days conditioning at 2°C compared to the unstimulated counterparts at 17.6 kgf. However, the authors did comment that the shear force values were more variable from the stimulated samples.

Drew et al (1984) conducted a similar study and found that the improvements from increased levels of conditioning and ageing were more marked in stimulated carcasses. The same group produced similar results in 1988 and, even after extending the total ageing period out to a total of 4 days using a combination of 10 and 4°C conditioning and ageing temperatures, the carcasses that had received low voltage stimulation were more tender.

2.3.3 Developing industry processing standards

In 1991, the Game Industry Board commissioned MIRINZ to develop a processing specification that could be used by industry to produce consistent tender venison. Specifically, this work set out to define what shear force values would translate into acceptable venison tenderness as judged by consumers. Having set this target, a processing specification was defined that, if followed, would reliably meet this criterion.

Using the MIRINZ Tenderometer, tenderness standards were agreed and set for frozen product (mean 7 with 95% < 10 kgf) and for chilled aged (mean 5.0 with 95% < 8gf).

The process to achieve this specification required the use of low voltage electrical stimulation at a frequency of 12.5 to 17.5 pulse per second, delivering a minimum of 125mA peak current, to be applied for 60 seconds via electrode clips on the lip and anus, and to be delivered within 5 minutes of sticking.

The recommended chilling regime was for carcasses to be cooled with air temperatures of 10°C for the first four hours, the chilling rate to be increased thereafter to ensure that a deep shoulder temperature of 2 to 5°C was achieved overnight.

Frozen product was to go into the freezers after boning and packing, whereas the chilled product was aged for at least 21 days at -1°C. The caveat to this was that alternative processes could be approved on the basis of achieving the equivalent tenderness level. However, low voltage stimulation was considered central to the process. A copy of the industry standards can be found in Appendix 1.

2.3.4 Recent experimental and industry based developments

While this large study led to the development of industry standards which are still in use today and undoubtedly resulted in significant improvements in the tenderness of the product being produced commercially, other quality attributes other than tenderness were not taken into account.

In 1996, Chadderton and Merts, under contract to the GIB, audited three commercial venison plants. The aim of this work was to demonstrate the plants were processing to the Industry Agreed Standard, that this was hygienically acceptable to MAF and to ascertain the magnitude of evaporative weight loss from venison carcasses. The results demonstrated that the industry has little quantitative data on evaporative weight lost or of subsequent drip in vacuum packs. However, these authors proposed a number of possible solutions, including improvement of existing chilling systems and/or the adoption of new chilling technologies, and calculated that the overall gains from better control of chilling and storage could add up to improved returns worth more than $2 million per annum.

As a result of this, Daly & Cairney were commission by the GIB in 1997 to conduct an experimental trial to accurately demonstrate the effects of chilling and stimulation on moisture losses and other quality parameters in venison. Using a large range of temperature gradients, they found that the ability of the venison to retain its water was increased with faster chilling rates and that this effect was improved further if the carcasses were not stimulated beforehand. After 3 weeks of chilled ageing, stimulated samples generally lost more drip than the unstimulated counterparts. Similarly, taste panel assessments of 3-week-aged venison found that the tenderness and juiciness was slightly better in the samples that had been fast chilled, although this was not affected by stimulation. In contrast, the colour stability (as measured using a colour meter) was increased in unstimulated unaged samples although, after 3 weeks of ageing, the colour stability was generally superior in the stimulated samples over the 3-day display period.

In an effort to repeat this trial in a commercial environment, Wiklund et al (2001), looked at the effects of low voltage electrical stimulation followed by a commercial

chilling regime on venison quality. This trial accurately reported loin pre-rigor pH and temperature profiles and results clearly show that the low voltage stimulation generated, on average, a 0.5 unit reduction in pH which persisted until at least 10 hours after slaughter. While the response to stimulation was extremely effective in this trial, the chilling regime was the same for both treatments and so two different pre-rigor/temperature profiles were generated (see Figure 4, ref number 8). The experiment demonstrated that the electrically stimulated samples were more tender after 1 day, 1 and 3 weeks of ageing, but differences disappeared thereafter. Drip losses in the vacuum packed product were unaffected by electrical stimulation, irrespective of the ageing period. In contrast, the colour stability was significantly reduced by electrical stimulation in product that had been aged for 1 week, but after 3, 6 and 12 weeks of ageing, these differences were no longer evident. In summary, this study demonstrated that electrical stimulation was beneficial in product destined to be aged for 1-2 weeks, but the effects did not persist thereafter. These results are broadly consistent with the results of Daly & Cairney, and differences between the studies can probably be attributed to the extent to which pH and temperature were manipulated.

Taken collectively, the limited amount of robust literature in the area of processing effects on quality would indicate that venison appears to be somewhat less sensitive to processing conditions when compared to either beef or lamb. The effects of stimulation on protein denaturation, and thus drip loss and colour, are perhaps not following exactly the same pattern seen in beef and lamb, and further work is needed to understand the mechanisms of the interaction between stimulation and quality attributes. Clearly, tailoring processing to optimise quality for the different markets offers as much potential for venison as for beef and lamb, but the underlying principles may need to be modified for venison.

Furthermore, in a study where the processing conditions were highly controlled, a wide variation in venison tenderness was illustrated. Using the same procedures as outlined in section 2.1.3. (see Daly et al, 2000), venison shear force was measured at rigor mortis and at regular intervals thereafter. In this study, the initial shear force ranged from 19.7 to 9.8 kgf and after 2 weeks of ageing at 0°C, the range was still large, from 6.2 to 9.4 kgf (Stevenson-Barry *et al*, unpublished). Clearly, by the end of the 2 week ageing period, many samples did not appear to have aged to their ultimate tenderness values, and a longer period of ageing may well have reduced the variability of the shear force values. Such long periods would be available for chilled produced destined for sea transport where potentially 6 or more weeks of ageing time are available, but would have serious implications for air freight chilled product (in

market within a few days of slaughter), or product that is frozen within 1 or 2 days of slaughter.

2.3.5 Conclusions from literature

- Little difference in pH response between high and low voltage stimulation.
- Low voltage stimulation provides tenderness advantages in product that has been aged for periods of ≤ 3 weeks.
- Tenderness variability between carcasses can be large and ageing periods longer than 2 weeks are required to overcome this.
- Over longer periods of storage, venison that has been through a rapid chill, losses less moisture as purge or drip.
- Low voltage stimulation does not seem to have an effect on colour stability if product has been aged for ≥ 3 weeks.
- Current literature provides conflicting results on the effects of low voltage stimulation on drip or purge.

2.3.6 Current Industry practice and quality outcomes

Current industry practice - Processing to industry agreed standard

During discussions with processors in the course of compiling this review, it is evident that the industry standards for the processing of venison are largely being followed. This is consistent with the authors' understanding of results of audits of DSPs against industry Agreed Standards. More detailed comments from those interviewed are provided as follows:

- **Stunning**: All processors surveyed were using captive bolt stunning. However, many expressed some interest in Halal stunning (electrical head-only stunning – see section 2.3.1).
- **Stimulation**: All processors surveyed were using low voltage electrical stimulation. The duration of stimulation ranged from 45 to 65 seconds with an average current of 125 – 150mA. The stimulation was applied between 30 and 70 seconds following sticking. Many of the plants monitored the stimulation and all calibrate on a regular basis.

- **Chilling**: There has been some modification to the cooling procedures, but in essence, the basic specification remains the normal process. The majority of plants surveyed commented that they load the chillers between 5 and 12°C and then some chill straight down to 0°C while others hold for a period at approximately 8°C ± 2°C, before pulling down to 0°C.
- **Boning**: The majority bone and pack at 24 hours post slaughter.
- **Freezing**: Generally, freezing is started after boning and packing using a 48-hour freezing cycle, although some plants commented that on some occasions they may hold for a few days at chiller temperatures before freezing down.
- **Tenderness testing**: All plants surveyed undertake routine tenderness monitoring as required under the industry standards.
- **Time to Market** (extremes). Much of the transport to market is via sea freight taking 4 to 6 weeks. This is for both chilled and frozen product. Some venison is air-freighted chilled and can sometimes be in the market within 2 to 3 days from slaughter.

Industry-identified issues:

Cold-chain: There are occasional claims associated with air-freight, but the temperatures are logged with both sea and air transport, so disputes are easily rectified and control is generally good. In contrast, it was generally felt that there were significant issues with cold-chain integrity in the local market.

Tenderness: Discussions with venison processors and others involved in the supply of quality venison have identified that chilled venison that takes several weeks to get to market rarely suffers from tenderness problems. In contrast, there are some issues with the tenderness of product that is frozen within 1 or 2 days of slaughter or chilled air-freighted product that is in the market within 2 to 3 days of slaughter.

In the tenderness survey conducted by MIRINZ in 1991, loins and topsides were collected from 10 plants and tenderness tested at 2 days and 3 weeks after slaughter: while the average tenderness ranged from 6.7 to 3.9 kgf after 3 weeks of ageing, after two days it ranged from 9.6 to 3.2 kgf. This work clearly identified the tenderness variability between plants while the variation between samples from within a plant was also generally high. No further surveys have been conducted since this.

Many contributors to this document commented that they felt the frequency and level of tenderness testing was not representative and should be revised to ensure that

testing frequency and sample numbers were relevant. It is also evident that, although most processors are sending samples for tenderness testing on a routine basis, there is no standardisation of the ageing time (time post-slaughter of samples being cooked prior to shear force testing), although, generally, they tend to have been aged for 2 to 3 weeks prior to testing. Also, only one plant was testing the tenderness of frozen or short tem chilled (air freight) product. This is an issue that needs addressing as it is likely that any tenderness issues reside with these products rather than chilled product that has been aged for several weeks. Several processors recognised that they have tenderness problems with such product and commented that they would like to develop strategies to overcome this.

Clearly, there is a large variability associated with ageing rates between venison carcasses and strategies are required to ensure that there is a greater level of tenderisation before freezing, first to overcome some of this variability and second to ensure that all product is of an acceptable tenderness level.

Other quality measures: While some processors conduct evaluations of purge and shelf life stability on chilled product, this tended to be on an ad-hoc basis and the methods of testing were inconsistent. However, some processors have measured purge levels in chilled product and claim it can be as high as 7% from loins, and tends to be a particular problem when the silver-skin is removed. A comment was made that in general, the market tends to be realistic about venison shelf life as it relates to colour. However, it was felt that venison quality in the market was not well understood and that regular objective quality measurements in-market would be of great benefit.

Recommendations based upon industry identified issues:

- Processing of frozen product requires revision to improve tenderness.
- Processing of chilled product that is destined for markets within 1 week of slaughter requires revision to improve tenderness.
- Ultimate pH should be measured routinely during the chilled season, and carcasses with a pH of greater than 5.8 should be removed from chilled consignment and frozen or put into further manufacturing.
- Tenderness testing procedures need to be revised to ensure that:
    - Sample selection numbers are statistically valid;
    - Increased frequency of testing;

  - Testing of product should be representative of the markets for which it is intended ie. If chilled to market within 1 week then testing should occur after 1 week of chilled storage;
  - Frozen product should be tested in addition to chilled product;
  - Cooking for shear force evaluation should be carried out from the frozen state thereby representing the worse case scenario;
  - Testing of chilled product should be carried out using tight specifications relating to conditions of sample transport; and

- Other objective measures of quality should be carried out in New Zealand and in the market.

2.3.7 Potential research opportunities identified for the venison industry arising from work reported in previous sections

Process optimisation

There are some clear opportunities that should be explored in the area of faster chilling regimes and the effect on long-term quality. Processing for chilled product destined to be in the market within 1 week of slaughter and for frozen product, may require some revision to the conditioning and ageing principle explored originally by Drew et al (1988) and then given commercial application by Gilbert et al in 1991. The seasonality of venison, with processing for the chilled markets tending to be limited to the 4-month window of August to November and the remainder of the year dedicated to frozen, allows different processes to be easily applied.

In general, some stimulation combined with different chilling regimes ranging from the use of high temperature conditions to rapid chilling, would clearly provide some opportunities to optimise specific product with regard to all quality attributes apart from just tenderness. Thus, the process optimisation work that was previously carried out for beef and lamb as discussed in section 3.2, could be readily applied to venison. Processing options for long-term chilled, short-term chilled and frozen product should be included.

Furthermore, low voltage tailored to the requirements of an individual carcass, as outlined in section 2.2.2, could be readily applied to venison. This technique would overcome some of the tenderness variability in both frozen and short-term chilled product in addition to having the potential to predict the ultimate pH.

Hot boning opportunities

Although hot boning has not been specifically addressed in previous sections of this review, it clearly has some potential benefits to quality if applied to venison processing.

While the temperature profile of superficial muscles such as the loin are easy to control and manipulate, it is much harder to control the temperature profiles of the deeper muscles when using traditional cold-boning techniques: significant temperature gradients will form within big leg muscles during the chilling process, invariably limiting the consistency of the process specification. Techniques such as hot-boning, combined with some of the newer technologies such as immersion chilling, create the opportunity to control and define the pre-rigor temperature/pH decline on an individual muscle/cut basis.

The label of 'slash and pack' is one that has been applied to hot-boning of beef. This has evolved due to the typical practices associated with hot boning bull and boner cow: Since hot boning was developed originally as a method of increasing processing efficiencies, the effects on quality, a relatively minor consideration in manufacturing beef, was ignored.

However, many hot-boning beef plants in New Zealand are now achieving comparable quality and in some cases, superior quality in the processing of prime beef compared to the cold-boning counterparts. They have achieved this due to the realisation that, when carried out carefully, hot boning allows the pre-rigor environment to be tailored very accurately. Additionally, processing of individual muscles can be tailored to the specific size and fibre type of the muscle, which has the potential to improve both the quality and consistency of the product. Precise processing conditions also offer the possibility of upgrading the qualities of some of these muscles.

Admittedly, a move from a cold to a hot-boning operation involves large capital outlay and a complete revision of virtually all cutting operations. However, there are opportunities to develop hybrid systems, where specific deep and slow cooling hind-quarter muscles are hot boned while the remainder of the carcass is cold boned in a conventional manner. The deep cuts are invariably the ones that produce high levels of drip and reduced colour stability, a consequence of their slow cooling rates. By cooling these cuts individually, heat transfer rates would be improved significantly and benefits in quality realised.

Summary of potential opportunities identified

- Benchmark existing quality to identify fluctuations since the last survey in 1991, and to get some indication of quality variability between carcasses.

- Revision of the current processing standard with process tailoring based upon frozen, chill/frozen and short-term chilled, long term chill/chilled.
- Application of advanced low voltage stimulation techniques to overcome some of the product variability issues.
- Hot boning and rapid chilling of some of the deep, slower cooling hind quarter muscles to improve purge loss, colour stability and texture.

3 Adding value to venison

This section of the review moves away from slaughter floor practices and procedures and considers the area of adding value to venison. Adding value can be applied to further cut preparation over and above basic primal cutting, or, at the other extreme, can include elaborate meal solutions tailored to different cultures and themes. However, to ensure that this section is relevant to the current industry focus and to realistic future opportunities, feedback on the potential topics was sought from the contributing processors. From this process, it was clear that some more information on packaging in general and upgrading lower value cuts would be of interest and thus these topics now form the focus of this next section.

3.1 Packaging

This section outlines current knowledge on packaging systems using information from key publications that cover both red meat and venison.

At chiller temperatures, meat spoils most rapidly through the development of high spoilage potential aerobic microflora dominated by Pseudomonas. The term ‘high spoilage potential’ is used to describe microorganisms whose growth on meat produces malodorous or otherwise offensive by-products.

Growth of such organisms can be effectively controlled by manipulating the oxygen levels within meat packs to create an oxygen limited storage atmosphere. Packaging systems that modify or restrict the growth of spoilage microorganisms are collectively described as preservative packing. Preservative packaging can be divided into primary and secondary packing systems.

Primary packaging refers to the long-term storage or transport packs while secondary packaging refers to retail packs used for short-term display.

3.1.1 Primary packaging systems

Vacuum packaging

The major preservative packaging utilised for chilled lamb, beef and venison is vacuum packaging, where an oxygen deficient environment is established around the meat by applying a vacuum to remove the ambient air from a meat pack and using a flexible oxygen barrier packaging film sealed around the meat. Product life of vacuum packed meat is inversely related to the oxygen permeability of the film. Today, most of

the films used for the export of chilled meat from New Zealand have very low oxygen transmission rates. If the film cannot conform closely to the product surface, the residual air occupying these spaces results in an evacuated rather than a vacuum pack. The oxygen in these voids will permeate into the pack during storage and bacterial growth will accelerate and the product will brown due to the formation of metmyoglobin. It is claimed that small quantities of residual oxygen that may be trapped will be converted into carbon dioxide by the respiratory activity of the meat, but how effective this mechanism is remains in some doubt.

Spoilage will normally become apparent several weeks after the lactobacillus microflora have reached numbers of $10^8/cm^2$, at which concentration both the odour and flavour of the meat is affected. With good temperature control, storage lives in excess of 8 weeks have been reported for lamb, and 12 weeks for beef (Devine & Moore, 1993). These storage lives for beef and lamb are relatively conservative estimates but are compatible with meat having the expected sensory attributes and a display life at of at least 2 days at 2 to 4°C when repackaged into oxygen permeable overwraps, or 4 days when packed into MAP packs.

A report by Devine & Moore 1983, showed that vacuum packed venison lasted up to 15 weeks and even after this period, there was very low microbial growth. However, the product lasted just a matter of hours in retail display as overwrapped steaks before it become visually unacceptable.

The reason for the very long vacuum packed storage of venison, when compared with beef and lamb, is not understood and requires further investigation.

Controlled atmosphere packaging

In the early 1980's it became apparent that the vacuum packing technology did not provide the storage life, particularly for lamb, to enable New Zealand to service distant markets with chilled product transported by sea. Despite strict attention to process hygiene and temperature control, which improved product performance in retail display, product could not be guaranteed a storage life beyond 10 to 12 weeks. The limiting factor affecting the storage life was found to be the ingress of oxygen into the packs, and a resolution of this problem was found in packaging in a carbon dioxide-enriched atmosphere, and replacing the plastic barrier film with an oxygen impermeable laminate. This new technology further extended the storage life of chilled lamb and beef.

Carbon dioxide is soluble in meat. Therefore, to ensure the pack atmosphere is saturated with carbon dioxide the gas must be sealed in the pack in the proportion of 1.5 litres per kg of meat. Initially, at pack sealing, the foil bag is distended like a pillow

but, after 24 hours, most of the carbon dioxide will be absorbed by the meat. However, the pack still contains a residual carbon dioxide level and so, unlike a vacuum pack, the packaging material is not tightly applied to the meat surface. Because of the tendency of myoglobin to oxidise to the brown metmyoglobin at very low oxygen concentrations, the packaging atmosphere must be less than 0.1% residual oxygen, a very low concentration of oxygen that requires carefully designed flushing procedures to accomplish consistently.

Essentially, carbon dioxide packaged meat can be reliably guaranteed for 13 weeks in lamb and 18 weeks in beef. However, the shelf life of CO_2 packed product either over-wrapped or packed in modified atmosphere (MAP) packs.is no different to product that has been previously stored under vacuum.

In contrast, work with carbon dioxide packaging of venison is limited, The early investigations done with prototype technology by Seman *et al.* (1989) showed no control of spoilage microflora and no storage life extension. These results can be questioned as the oxygen levels in the packs increased during storage, demonstrating that the packs were not secure and a lack of atmosphere control may have allowed microbial growth.

In contrast, trials carried out by AgResearch MIRINZ Centre using modern industrial packaging machines and materials, showed that venison primal cuts had a storage life of 30+ weeks at -1ºC using CO_2 packaging, and at 2ºC, the storage life was 15 weeks (Gill, 1990). Gill concluded that although vacuum packaging obviously works well for venison, use of packaging would allow greater diversification of product type and give the product a longer and more reliable storage life. Clearly there is a need to re-evaluate the use of CAP technology for high quality chilled venison.

3.1.2 Secondary packaging

Overwrap

This is the simplest cheapest form of retail packaging; the pack will prevent contamination but does very little to enhance the appearance of the product. However, this form of retail display dominates in New Zealand and is still evident in some areas of Europe and the USA.

Modified atmosphere packaging

Modified Atmosphere Packaging (MAP) replaces the ambient atmosphere with a gas mixture rich in oxygen, to enhance the bright red bloom of the meat, and carbon dioxide, to help control the spoilage bacteria. This technology significantly increases retail display life compared with the overwrap packaging, typically doubling the time to

unacceptable browning of the meat surface. This retail packaging form is now used extensively throughout Europe, particularly the U.K., for beef and lamb, despite being very expensive.

Currently Most chilled venison exported from New Zealand is destined for the hotel, restaurant and institution market, where display colour is not an issue. A very major limitation of venison when supplied for retail sale is its extremely short and variable shelf life particularly after chilled storage. Trials carried out by MIRINZ (1993) for the GIB showed that MAP increased display life compared with conventional overwrapped venison, an effect that would enhance chilled venison's retail appeal. This has increasing importance as retail market development is a critical part of the industry's strategy. However, even with MAP packaging, the retail display life of venison can be as little as 3-4 days.

In the last year, the USDA has allowed the inclusion of carbon monoxide (CO) in the pack atmospheres of CAP and MAP meat. CO binds to the myoglobin in meat and prevents its conversion to the brown metmyoglobin, effectively enhancing the bright red display colour and probably extending the very short display life of venison. Preliminary trial with ground beef packed in an atmosphere containing traces of CO showed major improvements in colour stability (unpublished), and may offer a revolutionary development for the retail display of venison.

3.2 Product opportunities from venison

The deer industry is efficient in producing and marketing table cuts, which already command premium prices, but is not doing much to add value to the manufacturing grades, which make up about 45% of the boneless meat produced (Swan, 1995). As a result, the high returns from the superior table cuts are strongly offset by low returns from the rest of the carcass. To improve revenue from deer processing, the value of the cheaper manufacturing cuts must be enhanced. To do this, the industry must know which of the muscles in the lower value regions of the carcass are suitable for producing value added products for various outlets. The industry must make sure that the values of the by-products especially the edible offals are also enhanced.

Thus, this section of the review firstly outlines strategies that have been applied to add value to the red meat industry and suggests methods by which these can be transferred to venison.

3.2.1 Recent work in adding value to beef and lamb

Recently, the meat industry worldwide has increased its focus on adding value to meat from species important to their respective countries/economies. For instance, Meat New Zealand recently funded studies to add value to beef and lamb (Farouk, 2000; 2001; 2002). Opportunities to add value to beef have been studied in the USA (NCBA, 2000) and the same is being done for Pork in the same country. Australia recently catalogued the best recipes for value-added beef and lamb products from butchers all over Australia (MLA, 2001). We are not aware of a similar work done on adding value to venison in New Zealand or overseas.

The strategies suggested for adding value to beef and lamb in the Meat New Zealand studies include:

- Creating new cuts;
- Further processing of new and existing cuts;
- Creating opportunities to use cuts in higher-value products; and
- Creating convenience products for ready-meal, functional foods and other niche markets.

These strategies were followed to suggest added-value opportunities for beef and lamb (Farouk, 2000, 2001, 2002). The added-value opportunities identified by Farouk in the Meat New Zealand studies have been summarised pictorially (Appendix 1 beef and 2 lamb).

Because of the difference in the physical, chemical and sensory attributes of individual muscles in cattle, sheep and deer, the suggested product opportunities for adding value to beef or lamb should not be directly adopted for venison. However, the strategy followed in adding value and the rationale for the choice of added value products could be used as a guide to add value to venison.

The following summarises the work done by Farouk (2000, 2001, 2002) on adding value to beef and lamb and provided the rationale for the 16 and 24 added-value products identified for the two species:

Adding value to beef

For beef forequarter, 14 of the forequarter muscles of commercial significance were characterised and used to develop 12 added-value products for various outlets. The 14 muscles were *brachialis, complexus,* extensors, flexors*, infraspinatus, longissimus cervicus, longissimus thoracis, rhomboideus, serratus ventralis, splenius, subscapularis, superficial pectoral, supraspinatus* and *triceps brachii; and* the 12

products were breakfast beef, pastrami, smoky beef, free-flow beef, microwavable beef, breaded/breaded and crumbed beef, soft jerky, beef floss, potroast, oven roast, gelled beef and restructured beef steaks.

For beef hindquarter, 17 of the hindquarter muscles of commercial significance were characterised and all but two of these muscles were used to prepare 15 added-value products for various outlets. The 17 muscles, sourced from steers and heifers, were the a*dductor femoris, biceps femoris*, *extensors* and *flexors* (as a group), *gastrocnemius, gluteus medius, gracillis, longissimus dorsi, obliquus externus abdominis, psoas major, rectus abdominis, rectus femoris, semimembranosus, semitendinosus, tensor fasciae latae, tranversus abdominis, vastus intermedius* and *vastus lateralis;* and the 15 products were beef floss, beef spread/dip, breaded/breaded and crumbed beef, breakfast beef, dried beef strips, free-flow beef, marinated steaks, oven roast, pastrami, potroast, ready meals, restructured beef steaks, restructured beef jerky/whole-tissue jerky, rotisserie beef and smoked beef strips.

The added-value products developed from beef fore- and hindquarter fell within the following categories: whole tissue products, intermediate meats, restructured products and novel meats. Whole tissue products included conventional steaks for traditional presentation and as an ingredient in convenience type meals, microwavable steaks, breaded beef, potroast and oven roast products for food service, dehydrated (beef floss) or intermediate moisture (soft jerky) products for snacks, and products for delicatessens including pastrami, smoky beef and breakfast beef. Intermediate meats included frozen free-flow chunks of various shapes and sizes. These products were targeted mainly at the convenience sector (home meal replacement and food service). Restructured steaks were made from heat- or cold-set whole muscle pieces or coarse beef particles. Novel meats that imitate other premium cuts or that target specific demographic groups were developed with the help of a professional chef. Consumer panels liked the products moderately to extremely.

Whole-tissue meat products

The products included in this category are marinated steaks, rotisserie beef, breaded beef, potroast, oven roast, pastrami and breakfast beef.

Currently, the consumption of steaks is increasing (Sloan, 2000). Value can be added to cheaper hindquarter cuts by developing acceptable steaks from these cuts to exploit this current trend. Adding marinade to cuts is another way of adding value, particularly when steaks are sold by weight. This also will exploit the trend toward

increased purchases of marinated products in supermarkets, particularly in the USA (Sloan 2001).

Rotisserie whole chicken is popular in many outlets. Rotisserie and other forms of direct flame cooking or roasting impart a unique flavour that consumers like. Currently, little or no rotisserie cooking of whole-tissue beef cut is done. Identifying cuts that could be used for rotisserie cooking, particularly in the food service sector, will result in higher value for these cuts both in the raw and cooked state.

Breaded products are popular finger foods (Sloan, 2001). Many breaded or breaded and crumbed poultry and marine products exist, but there are hardly any such products from red meats, including beef. One of the biggest problems with breading beef is the shrinkage of the meat portion during cooking, resulting in the crumbs or breadings falling off. Selecting the right meat cuts and developing batters or processing techniques can solve this problem. Successful development of breaded beef products has good potential to add value to cheaper cuts from beef.

Pot-roast and oven roast are popular foodservice items. The slow cooking involved in pot-roasting allows cuts that are tough due to a high collagen content to be used, making this process favourable for some of the high-collagen fore- and hindquarter cuts, especially the *M. gastrocnemius* and *rectus femoris*. Foodservice is a fast-growing sector in the USA, and opportunities exist in that sector for precooked, pre-seasoned, pre-sliced and portion-control products (Salvage, 2000). Suitable cuts for oven- or pot-roasting could be developed from cheaper cuts as a way of adding value.

Delicatessen products such as pastrami and breakfast beef are a good way to add value because such products currently are sold at ~ NZ$3 – 4 per 100 g of product. The yield of these products is about 95% of raw weight if our procedure is followed. Also, smoke flavour – used in both pastrami and breakfast beef – is one of the most popular flavours in the North American market. Similar price and yield as for pastrami could be obtained from the breakfast beef.

Intermediate products

The products included in this category are chunked meats of various sizes and shapes that were frozen and made to free-flow. These products are intended for the fast-growing home meal replacement or meals solution sectors and other similar sectors of the food industry. A diversity of production/distribution solutions is being offered to consumers, with the aim of partially or fully replacing homemade meals (Costa *et al*., 2001). In this sector, processors appreciate uniform, discrete meat pieces with flow characteristics that allow mechanisation of production. For instance, a home meal producer of curried beef on rice or beef stroganoff could incorporate an automatic

metering device to deliver a certain quantity of meat chunks for either batch or continuous product manufacture. Chilled meat pieces tend to stick together, which limits their use where flow is required.

Restructured Products

The products in this category include cold-set restructured steaks from either whole muscle or coarsely sliced beef pieces. Restructuring enhances the use of variably shaped and variably tender cuts of meat. Another advantage of restructuring is portion control, which is particularly important in the food service sector. With restructuring, low-value cuts of beef can be made to look like and have the eating quality of expensive cuts. Recent work has found restructured steaks from beef and venison forequarter muscles to be equally acceptable or better than non-restructured striploins and tenderloins from the same species (Farouk, unpublished data).

Snack Products

Snack bars are expected to be one of the fastest-growing eating-place segments (Sloan, 2001). Beef is the most widely used meat source for meat snacks (Brandt, 1999), and the fastest-growing snack item in the USA – our biggest beef market – is jerky. Therefore, developing snack products to capture the trend will add value.

Adding value to lamb

Fifteen lamb cuts were characterised and used plus six other underutilised cuts/muscles that were not characterised to suggest 24 added-value product opportunities for various outlets. The cuts/muscles used include blade roast, bolar, bones, boneless whole shoulder, boneless flap, brisket, cartlet, cube roll tail-on, eye of the round, flat, full loin, heel meat, inside, knuckle, neck, rib bones, rump, shank, tenderloin, tunnel-boned legs and trim*;* and the 24 products were appetizers/nibbles, battered & crumbed lamb, BBQ-ready lamb, boneless breast of lamb, burgers/patties, breakfast lamb/bacon, corn-on-the-cob-neck, deep fried lamb, dried lamb, free-flow lamb, jerky, kebabs, lamb bone soup/stock, meat balls, oven roasts, precooked roasts, steaks, ready meals, restructured lamb, ring sausages, rotisserie lamb and toppings.

The added-value product opportunities presented for lamb were chosen to create convenience and variety – two of the desirable attributes that consumer surveys identified as lacking in lamb (Heany, 2001). Opportunities to create convenience in this project have been achieved through primary processing and further processing.

Primary processing offers convenience

The products that were suggested in this category include boneless breast lamb, kebabs, free-flow lamb pieces (cubes, strips), oven roasts, restructured lamb, rotisserie lamb and table steaks.

The boneless breast lamb was suggested to take advantage of the popularity of boneless breast chicken. Boneless breast chicken is considered a highly convenient and consistent product.

Kebabs were suggested because a study (Ringkob et al., 1998) in the USA, aimed at developing and marketing all mutton products, concluded that Kebabs cubes were one of the products with the greatest market potential. Lamb kebab is already a popular product in British restaurants (Heany, 2001) and is seen by consumers as a convenient product. In the present study, a rotisserie was modified to be used for cooking kebabs. Pre-prepared frozen kebabs, skewered and ready for use, may be another way value can be added to lamb, particularly the legs and shoulders.

Free-flow lamb pieces are intended for the fast-growing home meal replacement or meals solution sectors and other similar sectors of the food industry. A diversity of production/distribution solutions is being offered to consumers, with the aim of partially or fully replacing homemade meals (Costa et al., 2001). In this sector, processors appreciate uniform, discrete meat pieces with flow characteristics that allow mechanisation of production. For instance, a home meal producer of curried lamb could incorporate an automatic metering device to deliver a certain quantity of meat chunks for either batch or continuous product manufacture. Chilled meat pieces tend to stick together, which limits their use where flow is required.

Oven-roasted lamb is one of the most popular menu items from lamb. Recent surveys in Britain indicate that 70% of those who buy lamb are 45 years or older and are among the higher income group. It is important to attract younger consumers to buy lamb. Because younger consumers tend to have less income and smaller family size, creating roasts for one or two individuals rather than using a whole leg or boneless rolled and netted shoulder may be a way to create convenience and attract the younger consumer and may even be more attractive to the older current buyers of lamb.

The products in the restructured lamb category include cold-set restructured steaks from either whole legs or shoulders, or smaller cuts such as loin or tenderloin. Restructuring enhances the use of variably shaped and sized cuts of meat. Another advantage of restructuring is portion control, which is particularly important in the food

service sector. With restructuring, whole boneless shoulder or legs, which are bulky, could be restructured and sliced into steaks which can be made to look like and have the eating quality of expensive cuts. Thin expensive cuts of lamb such as the tenderloin could be restructured to give a steak with a better plate cover and with good portion control.

Rotisserie whole chicken is popular in many outlets. Rotisserie and other forms of direct flame cooking or roasting impart a unique flavour that consumers like. Currently, little or no rotisserie cooking of whole-tissue lamb is done. Some cuts, like the insides, knuckle and rump, have suitable sizes, shape and texture for use in rotisserie cooking. In a previous study, rotisserie lamb as rated highly by in-house consumer panels (Farouk, 2001).

Currently, the consumption of steaks is increasing (Sloan, 2000). Considering that New Zealand lamb is generally tender, table steaks from shoulder and legs could be produced to add value to capture the trend in increased meat consumption.

Further processing offers convenience

Product opportunities suggested in this category include appetisers/nibbles, battered and crumbed lamb, BBQ-ready steaks, burgers (fresh and pre-cooked), corn-on-the-cob-neck, deep fried products, meatballs (raw and precooked), precooked roast, ready meals and toppings.

Finger foods are becoming very popular and snack bars are expected to be one of the fastest-growing eating-place segments (Sloan, 2001). Most restaurants list some appetiser/nibbles of some kind in their menus. Appropriate cuts of lamb can be used to fill this growing need for finger foods and snacks. Corn-on-the-cob-neck is an idea taken from corn-on-the-cob to describe a product that can be nibbled like a corn-on-the-cob.

Breaded products are popular finger foods (Sloan, 2001). Many breaded or breaded and crumbed poultry and marine products exist, but there are hardly any such products from red meats, particularly lamb and beef. One of the biggest problems with breading red meats is the shrinkage of the meat portion during cooking, resulting in the crumbs or breadings falling off. Selecting the right meat cuts and developing batters or processing techniques can solve this problem. Successful development of breaded lamb products has good potential to add value to lamb, since battered and breaded beef were found to be highly acceptable by consumers (Farouk, 2001).

There has been a tremendous growth in the use of barbecues and grills for outdoor cooking in the USA (Sloan, 1999). While beef is one of the grilling favourites in US, New Zealand successfully promoted lamb in that market as a suitable meat for barbecuing or grilling (Meat New Zealand, 1998). Developing barbecue/grill-ready lamb steaks that are ready for use and are free-flowing for added convenience will be a good way to add value, particularly to lamb legs and shoulders.

Burger - particularly beef burger - purchases have risen considerably recently (Fowler, 2001). Frozen lamb burgers and precooked frozen lamb burgers are a good way of adding value to trim from processing lamb carcasses. Gourmet burgers from leaner lamb may be another way of creating a convenient item for meals, barbecuing and capturing the increasing popularity of this item. Precooked burgers will enhance the convenience of burgers and may be a suitable product for some niche markets.

Deep frying imparts a unique flavour to foods that consumers have become accustomed to and expect in certain foods mainly for snacking. The consumer familiarity with the deep fried taste can be exploited to produce pre-cooked products from lamb that can be sold frozen or chilled to be reheated in a microwave for convenience. Certain lamb muscles are small enough to be used for deep frying.

Meatballs are popular with children (Fowler, 1999). Precooked frozen free-flow meatballs ready to be added to a sauce is a convenient method of using lamb trim and a good way to add value.

Precooked oven roast, chilled or frozen, using suitable cuts of lamb could be a good away of adding value due to the convenience it offers. Foodservice is a fast-growing sector in the USA, and opportunities exist in that sector for precooked, pre-seasoned, pre-sliced and portion-control products (Salvage, 2000).

Ready meals are the fastest-growing part of the meat products sector (Fowler, 1999). Using lamb as an ingredient in a ready meal will add value to lamb, particularly the shoulder and legs.

Toppings – convenience and variety

Product opportunities in this category were suggested to provide variety and novelty to consumers. In addition, the products also offer opportunity to use some of the most underutilised parts of a lamb carcass - example bones. The product suggested include, breakfast lamb/lamb bacon, lamb jerky, lamb soup/lamb soup stock, pet treats and ring sausages.

Bacon is traditionally made from pork bellies. Although many societies have religious and cultural taboos about eating pork, there are no such restrictions on sheepmeat (Young et al., 1994). With giant international fast food outlets, such as McDonald and Burger King, using bacon in their products, providing a substitute from lamb for countries or communities with taboos against pork, could be a way for using an underutilised lamb cut and a way of adding value. Bacon-like product made from beef was found to be highly acceptable by consumers who are traditional pork eaters (Farouk, 2000, 2001).

Jerky is one of the fastest-growing snack item in the USA. Therefore, developing snack products from hindquarter muscles to capture the trend will add value.

Soups are popular, particularly during winter time, and are a popular product among hospital patients and those in nursing homes and hospices. One way of adding value to lamb bones is to develop soups and soup stocks or flavourings.

Dried rib bones or sliced flab was found to be too fatty for human consumption and was suggested to be used as a pet treat. The dried bones were for cats and the dried flap strips were targeted to both dogs and cats. A huge amount of money is spent on pet treats locally and internationally and will be a way to add value.

Recent studies have shown that hotdogs and meatballs are very popular with children (Fowler, 1999). Ring sausages are skinless sausages that may find acceptance with children as a novelty item.

Could the strategies for adding value to other redmeats be applied to venison?

There is a dearth of literature on adding value to venison in spite of the fact that the industry has recognised the importance of adding value for some time (Wallis and Faulks, 1977; Sadler, 1989; Swan, 1995; Gutzke and Tobin, 1998). The available information on venison added value products falls under the following categories:

- Whole-tissue products
- Restructured products
- Coarse and fine emulsion type products
- Co-products

Added value whole tissue products

The added-value products in this category include venison ham, venison roast, air dried hams and jerky. Sadler (1989) suggested that the shoulder or neck muscles

could be used for venison ham or roast. The venison ham should be cured and sold to supermarkets or service delicatessens and the venison roast should be sold to hotels or restaurants. Air-dried hams and jerky were produced as part of a MIRINZ study in which venison smallgoods were developed for the German markets (Swan, 1995). The air-dried hams and jerky were made from venison leg cuts.

Added value restructured products

The added value products in this category include cooked hams, steaks and roasts. Gutzke and Tobin (1998) developed restructured steaks and roasts from venison shoulder muscles using different cold-binding systems. They (Gutzke and Tobin, 1998) studied a number of processing factors that affect the quality of restructured steaks and roasts such as type and concentration of binder, meat particle size, optimum temperature and pH and concluded that acceptable added-value products could be produced from venison shoulder muscles using restructuring technology.

Coarse and fine emulsion type added-value products

A number of products are included in this category. Swan (1994; 1995) produced scalded sausages (Brühwurst), salami, patties and aspic products from venison for the German consumers and found that the products were highly rated in that market. Detail recipes for the manufacture of these products are contained in an earlier report by Swan (1993).

Co-products of deer processing

Co-products of deer can be categorised into two groups/types: those harvested from the deer while alive (velvet antler and blood) and those removed from the deer after slaughter such as blood (for dried powdered blood), skins, tails, pizzles, sinews, hearts, livers, tongues and kidneys (Wallis and Faulks, 1977). Among the co-products of deer processing, the edible offals are the least value added. The remaining have found various uses as valuable products in oriental medicines.

Conclusion from added value work on venison

The following are the conclusions drawn from the available literature on adding value to venison:

- There is a dearth of published literature on adding value to cheaper cuts of venison or offal, indicating that not much value is being added to these categories of meat.
- Literature is available on adding value to beef and lamb which could be used as a guide to add value to venison.

Potential opportunities arising from added value section

- The use of CAP technology for venison should be re-evaluated as it may be able to offer new opportunities for chilled venison.
- The use of carbon monoxide in pack atmospheres should be investigated as it may offer significant commercial advantages for the retail display of venison.
- Adding value work similar to that carried out for beef and lamb should be undertaken for venison.
- Value should be added to venison offal.

4 "Blown Pack" Spoilage of Vacuum-Packed Chilled Meats: Cause(s) and Control

4.1 Introduction

At storage temperatures between -1.5 and 0°C, vacuum-packed fresh meat with a normal ultimate pH between 5.4 and 5.8 typically spoils after approximately 10 to 12 weeks. This spoilage is usually attributed to the growth of lactic acid bacteria and is manifested by relatively inoffensive sensory characteristics. Even at the end of shelf life of vacuum-packed chilled meats, the packaging typically remains tight around meat and no gas or only small gas bubbles are present inside the pack.

Until the late 1980's premature spoilage of vacuum-packed chilled meat due to pack blowing was usually attributed to the growth of Enterobacteriaceae and linked to temperature abuse of the product. However, since 1989, a number of spoilage incidents where temperature abuse has not occurred were reported in New Zealand and overseas (Dainty et al. 1989, Kalchayanand et al. 1989, Broda et al. 1996, Kalinowski and Tompkin 1999). In these incidents meat products of normal ultimate pH spoiled, as manifested by gross pack distension, within 4 to 6 weeks of storage at temperatures between -1.5 and 1°C. Clostridial aetiology of "blown pack" spoilage was initially inferred from the detection of butyl compounds, i.e. typical clostridial metabolites, in the gaseous atmospheres of spoiled vacuum packs. Subsequently, psychrophilic clostridia were isolated from blown packs, and a causal relationship between these microorganisms and "blown pack" spoilage was established in a definite manner by fulfilling Koch's postulates.

In the early 1990's, "blown pack" spoilage of vacuum-packed chilled meats was often thought to be a scientific curio and, consequently, the dearth of worldwide research on this spoilage condition and its causative agents was considered justified. However, during the past decade research continued to challenge the minor chilled meat spoilage role traditionally assigned to clostridia. In the past five years, "blown pack" spoilage of vacuum-packed chilled meats became recognised worldwide as a spoilage condition causing major economic losses in the beef, venison and lamb processing industries. Recently, it became evident that the development of effective measures to control "blown pack" spoilage is not possible without a thorough knowledge of its causative agent(s) and that the existing knowledge on this topic is very fragmented. The recognition of psychrophilic clostridia as a serious processing threat prompted significant changes to "blown pack" spoilage research worldwide; research projects that were in the past conducted by very few universities/research institutes, usually

resulting in publicly accessible knowledge, became replaced by intellectual property (IP) -protected applied studies that are conducted by individual meat companies and other commercial groups. Consequently, to reduce product loss due to “blown pack” spoilage, meat producers now have little choice but to independently direct, fund and conduct research programmes on this spoilage condition and its causative agents.

The aim of this review is to present the current status of worldwide research on “blown pack”-causing clostridia and its control in a meat plant environment, and to identify gaps in existing knowledge required to minimise product loss due to “blown pack” spoilage of vacuum-packed chilled meats.

4.2 Current status of knowledge on “blown pack” spoilage

4.2.1 Characteristics of “blown pack” spoilage of vacuum-packed chilled meats

Basic characteristics of “blown pack” spoilage

“Blown pack” spoilage of chilled anaerobically stored meats is known to occur with vacuum-packed fresh beef, lamb and venison primal cuts; chub-packed fresh ground beef; vacuum-packed roasted beef; and cooked pet food rolls packed in gas-impermeable plastic casings (Dainty, Edwards and Hibbard, 1989; Kalchayanand, Ray, Field et al., 1989; Broda, De Lacy, Bell et al., 1996 a and b; Kalinowski and Tompkin, 1999). This spoilage condition usually manifests around the 4th to 6th week of storage at temperatures between -1.5 and 1°C, or higher.

Sensory attributes of “blown pack” spoilage

“Blown pack” spoilage of vacuum-packed chilled meats is characterised by copious gas production causing gross pack distension (Fig. 6). Meat usually has the purple colour typical of anoxic tissue, but can occasionally be dark in colour with a green or reddish-green discolouration. Large amounts of drip accumulate in the pack. Although the texture of spoiled meat is usually firm, a partial proteolysis of meat tissue is sometimes reported. Meat odours shortly after pack opening are decidedly offensive ranging from “sulphurous”, “faecal” and “sewage-like” to “strong dairy” or “cheesy”.

Figure 6. Commercial sample of vacuum-packed venison showing gross pack distension.

Headspace volatiles

Analyses of volatile compounds from “blown” packs detect large amounts of carbon dioxide and hydrogen, but oxygen is usually present at levels of less than 0.5 ppb. 1-butanol, butyric and acetic acids, butyl acetate and butyl butyrate are typically detected in spoiled meat packs (Dainty, Edwards and Hibbard, 1989; Broda, De Lacy, Bell et al., 1996 a and b). Significant amounts of sulphur compounds, such as hydrogen sulphide, dimethyl sulphide or dimethyl disulphide, are also found in the headspace of “blown” packs.

Microbiological characteristics of “blown pack” spoilage

Typically, microscopic examination of Gram stained drip from “blown” packs shows a significant proportion of large sporeforming rods. However, routine microbiological analyses of meat from “blown” packs do not show a significant departure from the expected norm, with lactic acid bacteria dominating the floras. Enterobacteriaceae and other known gas-producing spoilage microorganisms are typically not detected or present in only low numbers.

4.2.2 Characteristics of causative agents of "blown pack" spoilage

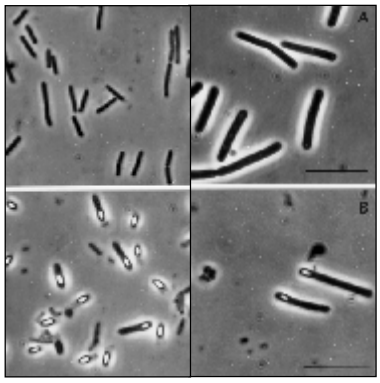
Taxonomy of "blown pack"-causing clostridia

Presence of sensory attributes of "blown pack" spoilage is attributed to the metabolic activity of microorganisms belonging to the genus *Clostridium*. This genus includes predominantly Gram-positive, rod-shaped, anaerobic and spore-forming bacteria. Two clostridial species, *Clostridium estertheticum* and *Clostridium gasigenes*, are recognised as causative agents of "blown pack" spoilage (Collins, Rodrigues, Dainty et al., 1992; Broda, Saul, Lawson et al., 2000). Another species, *Clostridium laramiense*, which was described in 1989 and confirmed as a causative agent of "blown pack" spoilage of vacuum-packed raw and cooked chilled beef (Kalchayanand, Ray, Field et al., 1989), was recently found to display a high degree of genomic similarity to *Cl. estertheticum*. It was proposed that *Cl. laramiense* be considered a different strain of *Cl. estertheticum*. Consequently, *Cl. laramiense* is now re-classified as *Cl. estertheticum* subspecies *laramiense*, while *Cl. estertheticum* carries species epithet *Cl. estertheticum* subspecies *estertheticum* (Spring, Merkhoffer, Weiss et al., 2003).

Recently, four new species of psychrophilic clostridia were obtained from an Antarctic microbial mat. *Cl. frigoris*, *Cl. lacusfryxellense*, *Cl. bowmanii* and *Cl. psychrophilum*, each share over 97% 16S rDNA sequence similarity with, and therefore, are phylogenetically related to, *Cl. estertheticum* (Spring, Merkhoffer, Weiss et al., 2003). It is yet unknown whether any of these newly described species is capable of causing pack blowing in chilled meats. However, psychrophilic strains of *Cl. estertheticum*-like clostridia designated K21 and K24, were isolated locally from gas-distended vacuum-packed chilled venison and were subsequently confirmed as abundant gas-producers. Whilst being somewhat similar to *Cl. estertheticum*, these strains carry many unique characteristics and appear to represent previously undescribed clostridial species. This review will focus both on the two described clostridial species recognised as causative agents of "blown pack" spoilage, *Cl. estertheticum* and *Cl. gasigenes*, and on local "blown pack"-causing *Cl. estertheticum*-like strains K21 and K24.

Morphological, physiological and biochemical characteristics

Cells of *Cl. estertheticum*, *Cl. gasigenes* and strains K21/24 are Gram-positive, motile rods producing subterminal or terminal spores during the late-stationary growth phase (Fig. 7). Colonies of these clostridia on sheep-blood agar are 0.7 to 3.0 mm in diameter, cream-white through dirty yellow to greyish and are β-haemolytic.



DB1A K21

Figure 7. Phase-contrast micrographs of vegetative (a) and sporulated (b) cells of "blown pack"-causing clostridia. Bar lengths equal 3.5μm for *Cl. estertheticum*-like strain K21, and 10μm for *Cl. gasigenes* strain DB1A.

Cl. estertheticum, *Cl. gasigenes* and strains K21/24 will not grow on aerobic solid, or in oxygenated liquid media (Collins, Rodrigues, Dainty et al., 1992; Kalchayanand, Ray and Field, 1993; Broda, 1999; Broda, Saul, Lawson et al., 2000; Spring, Merkhoffer, Weiss et al., 2003). The organisms are psychrophilic, with optimum growth temperature of 6-8°C for *Cl. estertheticum* subsp. *estertheticum*, 15°C for *Cl. estertheticum* subsp. *laramiense* and strains K21/K24, and 20-22°C for *Cl. gasigenes* (Fig. 8). The upper temperature limits for growth are 13°C, 21°C and 26°C, respectively. All these "blown pack"-causing clostridia are capable of growth at -1.5°C. The optimal pH for anaerobic growth is about 6.5 to 7.0 (Fig. 9).

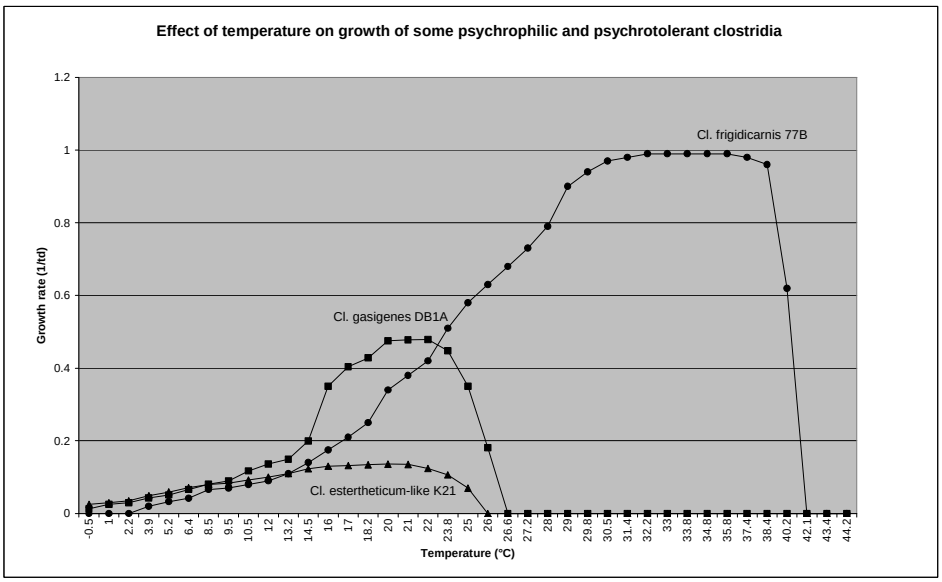


Figure 8. Comparison of growth rates of "blown pack"-causing psychrophiles *Cl. gasigenes* and *Cl. estertheticum*-like strain K21, and psychrotolerant *Cl. frigidicarnis* in PYGS broth at various temperatures.

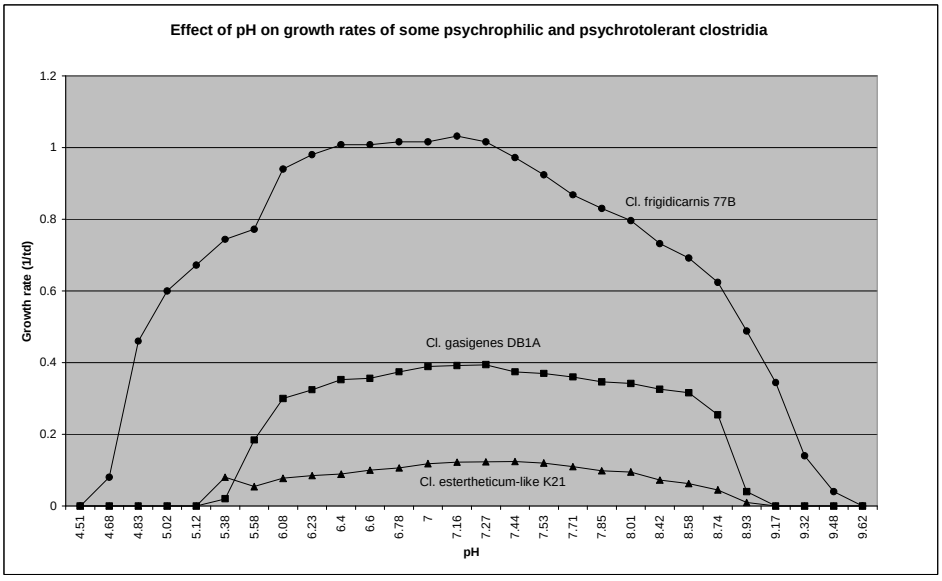


Figure 9. Comparison of growth rates of "blown pack"-causing psychrophiles *Cl. gasigenes* and *Cl. estertheticum*-like strain K21, and psychrotolerant *Cl. frigidicarnis* in PYGS broth at various pH values.

Cl. estertheticum subsp. *estertheticum*, *Cl. estertheticum* subsp. *laramiense*, *Cl. gasigenes* and strains K21/K24 are capable of utilising and fermenting a range of carbohydrates, including glucose and fructose. In contrast to *Cl. estertheticum*, *Cl. gasigenes* and strains K21/K24 hydrolyse gelatine. Of these clostridia, only strain K21 produces milk curding and meat proteolysis, as shown by milk reaction and meat digestion tests.

In Peptone, Yeast Extract, Glucose, Starch (PYGS) broth, the most abundant gaseous fermentation end products of "blown pack"-causing clostridia are hydrogen and carbon dioxide. In PYGS broth, the most abundant non-gaseous fermentation end products of "blown pack"-causing clostridia are butyrate, acetate, lactate and ethanol. In addition, *Cl. estertheticum* and *Cl. gasigenes*, but not strains K21/K24, produce significant amounts of 1-butanol.

When tested using botulinal neurotoxin mouse bioassay, supernatants of *Cl gasigenes* strain DB1A and strain K21 were non-toxic to mice. Recently, strains of New Zealand-discovered psychrotolerant clostridia were isolated from the blood culture of a patient in Germany (P. Phaller, personal communication) and these microorganisms are increasingly being perceived as emerging or re-emerging human pathogens. However, it remains improbable that psychrophilic clostridia which are unable to grow at body temperature could cause human medical conditions. Consequently, the presence of "blown pack"-causing clostridia in vacuum-packed meats does not endanger food safety and should be considered solely a food spoilage issue.

4.2.3 Factors affecting growth and survival of "blown pack"-causing clostridia

Factors affecting growth and survival of vegetative cells

The single most important factor affecting growth and survival of vegetative cells of "blown pack"-causing clostridia is temperature. Under laboratory conditions, vegetative cells of psychrophilic clostridia rapidly loose viability at temperatures above their maximum for growth. However, detailed data on rates of thermal inactivation of vegetative clostridial cells in laboratory media and/or meat matrices is not available.

Whilst growth of psychrophilic clostridia occurs at temperatures below 0°C, these microorganisms do not divide rapidly at low temperatures. At -0.5°C, generation times of 40 h were obtained for *Cl. estertheticum*-like strain K21 during incubation in PYGS broth, while generation times of 77 h were obtained for *Cl. gasigenes* under the same growth conditions (Broda, 1999; Broda, Saul, Lawson et al., 2000). At 4°C, these

microorganisms grew nearly twice and four times faster respectively, achieving doubling times of 20 and 23 h. Generation times rose further at elevated temperatures, though not as rapidly as at temperatures between -0.5 and 4°C. Generation times of "blown pack"-causing clostridia in vacuum-packed meat are unknown.

While it is believed that vegetative cells of psychrophilic clostridia pose significant resistance to freezing, little data is available to demonstrate this effect. Similarly, no information exists as to the temperature below which inactivation of clostridial cells may occur. Lack of success with the isolation of viable clostridia from "blown" packs that have been placed into frozen storage is usually attributed to the destruction of viable cells by high levels of metabolites that accumulated in the packs, rather than due to their thermal inactivation. It should be noted however, that DNA of "blown pack"-causing clostridia can be readily detected in packs stored at -18°C for 2 years or longer.

Little is known about how other factors such as water activity (a_w); salts or oxidation-reduction potential (E_h) affect the growth and survival of "blown pack"-causing clostridial cells. While carbon dioxide packaging inhibited "blown pack" spoilage to some extent in packs inoculated with low numbers of *Cl. estertheticum* spores and incubated at either -1.5°C or 2°C, the inhibitory effect appeared inconsistent (Broda, Penney, Miller et al., 2002). Results obtained to-date indicate that carbon dioxide packaging does not afford a means for preventing "blown pack" spoilage.

Factors affecting sporulation and survival of spores

"Blown pack"-causing clostridia do not sporulate readily in laboratory media. With *Cl. estertheticum*, spores are optimally produced under semi-starvation conditions, such as those created in a two-phase medium consisting of lower solid phase of cooked meat agar and upper liquid phase of deoxygenated water. In the two-phase medium, maximum levels of *Cl. estertheticum* spores are produced within 3 to 6 months of incubation at 7 to 10°C.

It is believed that psychrophilic clostridia produce spores that, while possessing some degree of heat resistance, are more heat-sensitive than spores of psychrotolerant or mesophilic clostridia. In a previous study, $D_{95°C}$ values (times required for 90% reduction in population of viable spores at 95°C) determined for psychrophilic clostridia ranged from 0.6 to 2.9 min, and $D_{95°C}$ values obtained for psychrotolerant clostridia ranged from 3.1 to 11.0 min (Broda, De Lacy and Bell, 1998). Little is known, however, about the effect of temperature on thermal inactivation of spores of "blown

pack"-causing clostridia. It is recognised that heat resistance of clostridial spores can be influenced by a number of genetic and environmental factors. Consequently, populations of spores of "blown pack"-causing clostridia may also vary in their heat resistance, depending on selection of species or strains, sporulation temperature and medium, and heating medium or matrix in which heat activation is occurring.

Spores of "blown pack"-causing clostridia survive freezing at -18°C for at least 2 years.

Factors affecting spore activation, germination and outgrowth

Because of the presumed sensitivity of spores of psychrophilic clostridia to heat, spore activating heat treatments may result in thermal injury to clostridial spores and should be avoided when attempting to culture these microorganisms from complex samples.

Spores of psychrophilic microorganisms are capable of activation in response to nutritional, rather than heat, stimuli. Comparison of *Cl. estertheticum* spore counts obtained with and without prior heat treatment, and with and without ethanol treatment, indicated that the majority of the initial spore inoculum is usually able to germinate without heat or ethanol spore activating treatments.

4.2.4 Isolation, detection and typing of "blown pack"-causing clostridia

Conventional culture-based detection

Nearly 15 years after first incidents of "blown pack" spoilage were reported, conventional isolation and enumeration of pack blowing clostridia remains difficult and technically demanding. Consequently, psychrophilic clostridia often go undetected in instances of overt meat spoilage unless stringent anaerobic techniques and low incubation temperatures are employed (Dainty, Edwards and Hibbard, 1989; Broda, De Lacy, Bell et al., 1996a and b). It is thought that difficulties with isolation and enumeration of "blown pack"-causing clostridia from the drip of "blown" packs may be due to toxicity of butyl compounds. When the carbon source becomes limiting after prolonged storage of meat, some clostridia may show an increased sensitivity to butyl compounds in concentrations that are normally non-toxic (Cortinas, Micalizzi and de Guzman, 1994). Alternatively, difficulties with isolation may be due to clostridial inhibition by, or direct competition with, lactic acid bacteria and their metabolites.

Generally, recovery of "blown pack"-causing clostridia should be conducted under strict anaerobic conditions using fully pre-reduced media. The best plating efficiency is usually obtained on non-selective solid media, such as Columbia Blood Agar containing 5% of sheep blood or PYGS agar. Selective media, even those devised for

enumeration of sulphite-reducing anaerobes in foods, are likely to fully or partially inhibit the growth of “blown pack”-causing clostridia (Broda, De Lacy and Bell, 1995, 1998b. For liquid enrichment, anaerobic PYGS broth is recommended. Anaerobic incubation is required at 7 to 10°C for 3 to 4 weeks. Isolates that are obligately anaerobic, catalase and oxidase negative, metronidazole sensitive Gram-positive large rods with terminal or subterminal spores that are unable to grow at about 25°C or above but grow at 4°C or below can be considered to be psychrophilic clostridia. Because a number of psychrophilic clostridia exist that are unable to cause pack blowing, the obtained isolates must be subsequently identified to the species level.

Non-selective media are not suitable for enumeration of “blown pack”-causing clostridia from vacuum-packed chill-stored meats, faeces, hides, soil, or other material where a competing microflora can be expected to be present in similar or higher numbers than psychrophilic clostridia. However, the probability of isolating these microorganisms from mixed microflora increases by the application of treatments that eliminate competing vegetative cells. Therefore, to ensure maximum spore recovery of these microorganisms, a treatment of sample with ethanol at a final concentration of about 50% is recommended. If heat treatment is used, the recovery of thermally injured spores of psychrophilic clostridia is enhanced by the inclusion of lysozyme in the medium. Up to 7 log increase in cfu of recovered spores may be achieved on a lysozyme- or egg yolk-containing non-selective solid media (Broda, De Lacy and Bell, 1998b).

Culture-independent detection

The difficulty with conventional detection of “blown pack”-causing clostridia led to development of methods for molecular detection of these microorganisms (Boerema, Broda and Bell, 1999c). The DNA-based method that is now widely adopted by meat industry and meat testing laboratories use primer sets and PCR amplification procedures that detect the presence of species-specific 16S rDNA gene fragments of *Cl. estertheticum* and *Cl. gasigenes* (Helps, Harbour and Corry, 1999; Broda, Boerema and Bell, 2003). Detection of amplicons is via horizontal gel electrophoresis or RealTime PCR melting curve analysis. With the developed 16S rDNA-based method, minimum levels of detection are 10^4 g^{-1} cfu for non-enriched meat samples, and 10^2 cfu g^{-1} for meat samples previously enriched in PYGS broth and incubated anaerobically at 10°C for 7 days. In contrast to standard PCR which offers only semi-quantitative detection of clostridia, recently developed RealTime PCR is capable of fully quantitative analysis (J. Corry and T. Nielsen, personal communications).

The two main shortcomings of the existing 16S rDNA-based method are low sensitivity of the assay for direct (without enrichment) detection of clostridia and its inability to detect the presence of spores. While in a “blown” pack clostridia are likely to be present in high numbers and in a vegetative cell form, on dressed carcasses and in a meat plant environment “blown-pack”-causing clostridia are likely to reside as low numbers of spores. At present, a rapid detection kit is being developed under a contract with DEEResearch to enable molecular detection of low numbers of clostridial spores in matrices other than meat or drip from “blown” packs.

With respect to 16S rDNA-based detection of “blown pack”-causing clostridia, some concern exists surrounding the recent isolation of new species of psychrophilic clostridia that share over 97% 16S rDNA sequence similarity with *Cl. estertheticum*. From comparison of 16S rDNA sequences of the newly discovered species, it appears that the existing *Cl. estertheticum*- and strains K21/K24-specific primers are likely to give PCR product when DNA of any of these new species is present in the PCR template. Should the ability of these microorganisms to cause “blown pack” spoilage of vacuum-packed chilled meats be confirmed, the existing 16S rDNA-based method will remain suitable for detection of causative agents of this spoilage condition. However, if any of these species is found incapable of causing pack blowing, an alternative procedure based on a DNA target that differentiates between pack blowing and non-blowing clostridia will need to be developed.

Identification

If culture-dependant detection yields isolates that are later confirmed as psychrophilic clostridia species-level identification is necessary to confirm that these isolates represent species of “blown pack”-causing clostridia. When conventional microbiological methods are used, morphological and biochemical characteristics of “blown pack”-causing clostridia do not allow for their differentiation from closely related species. Similarly, no dichotomous keys permit ready identification of these microorganisms. Consequently, species identification of cultured isolates is best achieved with PCR amplification of species-specific 16S rDNA fragment, restriction fragment length polymorphism (RFLP) analysis of complete 16S rDNA gene, or when the identity of PCR-amplified product needs to be proved without any doubt, with DNA sequencing (Broda, Musgrave and Bell, 2000).

Typing

Strain-specific differentiation of clostridia may be useful for tracing “blown pack”-causing clostridia from spoiled product back to their sources in a processing plant environment. Strain-level differentiation of “blown pack”-causing clostridia can be

achieved using RFLP analysis of total genomic DNA (Broda, Musgrave and Bell, 2000), or with some species, with 16S-23S rDNA internal transcribed spacer analysis (Broda, Musgrave and Bell, 1996; 2003).

Strategies for detection of "blown pack"-causing clostridia

Generally, the frequency of sampling, the choice of sampling sites and the choice of the method for detection of "blown pack"-causing clostridia depends on the overall aim of the testing.

(1) Hide/faecal samples testing

Aim: To establish primary source(s) of "blown pack"-causing clostridia in the processing plant. To identify farms that supply animals carrying high numbers of clostridial spores. To confirm whether a relationship exists in processing plants between pack blowing and animal presentation for slaughter.

Sampling material: Hide swabs, faecal samples, and/or kill floor swabs at operations that involve handling of pelt-on animals (e.g. a hide puller). Swabs are typically gauze pads of sufficient size for swabbing of approx. 25 cm^2 surface area. The sample is taken from rump or the dirty areas of the belly. A wet and dry swabbing technique is used, with the first swab being moistened with sterile saline or dilution fluid prior to swabbing, and the second dry swab being subsequently used on the area of the first swab. With faecal samples, material that is least exposed to air is collected in the stockyard pens or on the viscera table. Samples should be collected into sterile plastic bags, packed anaerobically and placed under refrigeration for transport to the laboratory.

Sampling frequency and time: The testing is preferably conducted during the high-risk season for "blown pack" spoilage. Repeat annually, as requested, or throughout the year where the high-risk season for "blown pack" spoilage is unknown.

Method for detection of "blown pack" clostridia: Samples are incubated anaerobically for 3 to 4 weeks in a Peptone Yeast Extract Glucose Starch (PYGS) broth. An aliquot of this enrichment is then used for PCR detection of the two clostridial species that are recognised as causative agents of "blown pack" spoilage, *Cl. estertheticum* and *Cl. gasigenes*. PYGS enrichment is used to minimise PCR inhibition by soil or faecal particles that may persist in the processing plant/farm environment and to allow the outgrowth of clostridial spores which otherwise are difficult to lyse in a standard DNA isolation protocol.

(2) Environmental testing

Aim: To establish secondary source(s) of "blown pack"-causing clostridia in the processing plant from which clostridial spores may spread onto clostridia-free carcasses/cuts. To confirm that the plant environment is clostridia-free after the high-risk season. To evaluate the efficacy of cleaning procedures, cleaning agents or interventions.

Sampling material: Swabs collected in the ventilation system throughout the plant (e.g. air socks, fans and surrounds), carcass coolers (e.g. drip trays and drains) and boning rooms (e.g. conveyer belts and vacuum packaging machine).

Sampling frequency and time: The testing is preferably conducted during or at the end of the high-risk season for "blown pack" spoilage and, as a quality assurance measure, can be repeated annually. For evaluation of the efficacy of cleaning procedures, testing is conducted as required.

Method for detection of "blown pack" clostridia: With this testing, gauze swabs are collected and the PYGS enrichment method is used, as described previously for hide/faecal samples testing.

(3) Carcass testing

Aim: To quantify the load of clostridial spores on dressed carcasses. To rapidly identify dressed carcasses carrying high loads of clostridial spores prior to boning and packaging. To establish the time to the onset of blowing for vacuum-packed cuts derived from carcasses carrying "blown-pack"-causing clostridia. To decide the fate (chill or freeze) of venison from carcasses carrying high numbers of clostridia before it is prepared for chilled storage.

Sampling material: Carcass swabbing is conducted using gauze swabs, as described previously for hide/faecal samples testing, except that the swab area should be increased to approx. 500 cm^2. With conventional dressing system, flanks and/or hinds should be swabbed on the entry to, or exit from, the carcass cooler.

Sampling frequency and time: Throughout the year. When more information about the seasonality of "blown pack" spoilage incidents in a processing plant is gained, tests can be limited to the high-risk season for pack blowing.

Method for detection of "blown pack" clostridia: Because "blown pack"-causing clostridia on dressed carcasses are likely to be present as spores, the PYGS enrichment method would appear best suited for this testing. However, anaerobic incubation for 3 to 4 weeks is not acceptable, if test results are to be used to determine the cut disposition on-line. Rapid detection methods, such as RealTime PCR or rapid PCR kits, to be used in conjunction with short spore germination-enhancing enrichment, are being developed at present and these methods may provide testing more suited for this purpose.

(4) Final product testing

Aim: To confirm the causative agent during initial investigations of "blown pack" spoilage. Subsequently, this testing is only required when the plant has in the past experienced blowing incidents that were caused by microorganisms other than "blown pack"-causing clostridia.

Sampling material: Swab or drip samples from the vacuum-packed meat.

Sampling frequency and time: As required.

Method for detection of "blown pack" clostridia: Direct PCR detection without prior enrichment.

4.2.5 Epidemiology

Reservoirs and incidence of "blown pack"-causing clostridia in natural environments

In the past, psychrophilic clostridia were isolated from permanently cold, oxygen deprived environments, such as deep marine sediments of Antarctica (Mountford, Rainey, Burghardt et al., 1997). Whilst the presence of *Cl. estertheticum* was recently detected in Antarctic soils (Brambilla, Hippe, Hagelstein et al., 2001), it appears that this microorganism is also ubiquitous throughout the farm environment of temperate climate countries. On farms, *Cl. estertheticum* and *Cl. gasigenes* are commonly detected in soil, mud, and creeks and through waters (Boerema, Broda and Bell, 2000 a and b). These microorganisms are also present on farm vegetation, such as grass or lucerne.

In farm soils, *Cl. estertheticum* and *Cl. gasigenes* are commonly encountered at levels of at least 10^3 to 10^4 cfu g^{-1}. Prevalence of these microorganisms in farms can vary greatly, with all samples tested being negative for presence of clostridia through to

100% samples being positive. Regardless of the prevalence of “blown pack”-causing clostridia in farm environments, meat manufactured using ovine stock supplied by some farms proved more prone to accelerated pack blowing than meat of alternative origins (Boerema, Broda and Bell, 2000 a and b). With ovine species, it appears that some farms are at higher risk of supplying carcass meat that will subsequently produce “blown” product than others.

Reservoirs and incidence in foods

In the past, “blown pack”-causing clostridia were isolated from vacuum-packed chilled beef, lamb and venison (Dainty, Edwards and Hibbard, 1989; Kalchayanand, Ray, Field et al., 1989; Broda, De Lacy, Bell et al., 1996 a and b; Broda, De Lacy, Cook et al., 1997), as well as from vacuum-packed roast beef (Kalchayanand, Ray, Field et al., 1989). However, our knowledge of clostridial reservoirs in foods other than meat is limited. It is unknown what proportion of raw meat may contain spores of “blown pack”-causing clostridia.

Processing plant reservoirs and mode of entry onto fresh meat

Psychrophilic and psychrotolerant clostridia are extremely common in a processing plant environment. Early culture-based studies determined that major sources of these microorganisms in a processing plant are slaughter animals themselves (Broda, Boerema, De Lacy et al., 1996; Broda, Bell, Boerema et al., 2002). About 100% of beef, 97% lamb and 96% of deer slaughter stock carried culturable psychrophilic and psychrotolerant clostridia. With “blown pack”-causing clostridia, *Cl. gasigenes* was isolated from hide/faeces of approximately 13% of beef, 0% of lamb and 10% of deer, slaughter stock (Boerema, Broda and Bell, 1997 a and b; Broda, Boerema and Bell, 1997), while *Cl. estertheticum*-like clostridia were isolated from about 16%, 6% and 23% of beef, lamb and deer slaughter stock, respectively. Prevalence of clostridia varied greatly between sampling days, with *Cl. gasigenes* isolated from approximately 5 to 18%, and *Cl. estertheticum*-like isolates obtained from 10 to 40% of deer slaughter stock. With molecular methods, “blown pack”-causing clostridia are typically detected in nearly 100% of hide and faecal samples (Boerema, Boerema and Bell, 1999, 2003). The numbers of *Cl. estertheticum* and *Cl. gasigenes* in these samples are estimated to exceed 10^3 to 10^4 cfu g^{-1}.

In a processing plant environment, “blown pack”-causing clostridia are commonly detected in stockyards on railing, ramp and floor surfaces, and in stockyard drinking water. On the slaughter floor, these microorganisms are present at many operations that involve handling a pelt-on animal, for example, sticking, bleeding, conducting opening cuts, skinning or hide pulling (Boerema, Broda and Bell, 2000 a and b). At

these operations, “blown pack”-causing clostridia are often detected on rails, floors, walls, fans and equipment surfaces. Except when these microorganisms are present at dressed carcass surfaces, *Cl. estertheticum* and *Cl. gasigenes* are rarely detected at evisceration or trimming, and are normally absent in carcass chillers and boning room. However, in plants where the process hygiene is compromised, “blown-pack”-causing clostridia can colonise some carcass chilling or boning room operations and sporadic instances of the above are known to the author of this report. “Blown pack”-causing clostridia may be occasionally detected in air, especially when reversed (i.e. from “dirty” to “clean”) air movement is observed in a processing plant due to its physical design or inappropriate location of air exhausts, intakes or fans.

“Blown pack”-causing clostridia seem to originate from the farm environment. It appears that spores of psychrophilic clostridia that are unable to grow above 25°C can survive passage though the gastrointestinal tract of warm-blooded animals and the exposure to oxygen and elevated temperatures during transfer on the animal’s hide. It is thus probable that “blown pack”-causing clostridia enter processing plants on hides and in faeces of slaughter animals and from these primary processing plant reservoirs are transferred onto dressed carcass with first opening cuts. In the absence of these microorganisms in plant operations other than those involving handling pelt-on carcasses, it should be concluded that slaughter animals represent a significant reservoir for isolates that actually cause pack blowing.

Incidence of “blown pack” spoilage

“Blown pack” spoilage is a worldwide issue affecting meat manufactured in Canada, Germany, Ireland, Netherlands, New Zealand, United Kingdom and the United States. Beef appears to be the main vehicle for “blown pack” spoilage in Ireland, UK and USA, while in New Zealand venison is the main meat species affected by this condition. The number of “blown pack” spoilage incidents occurring worldwide remains unknown, as it is proprietary information held by affected companies. However, this spoilage condition is no longer considered sporadic. Industry information suggests that “blown pack” spoilage condition causes major product losses (e.g. a whole day’s production) in major meat manufacturing companies in Ireland, UK and USA. The economic losses borne to-date by New Zealand processors remain unknown.

Worldwide, a number of issues are commonly observed with the majority of “blown pack” spoilage incidents. Initial episodes of “blown pack” spoilage usually manifest themselves in a processing plant suddenly, leading to a belief that the source of carcass/cut contamination with pack blowing clostridia may reside in a single slaughter or boning operation and, thus, that the re-occurrence of the spoilage could be

prevented by enhanced hygiene measures applied at this processing site. Alternatively, the sudden occurrence of “blown pack” spoilage is tentatively linked to current changes to processing practices. Unfortunately, commercial experience indicates that one-off clostridial colonisation of a single processing operation or immediate changes to processing are rarely the causes of the onset of “blown pack” spoilage.

“Blown pack” spoilage incidents commonly re-occur at the same time of the year and/or during similar weather conditions. Thus, the occurrence of these incidents can be indirectly linked to cold, wet or, conversely, dry weather, and, directly, to the animal presentation for slaughter. Other common issues experienced during “blown pack” spoilage incidents include variation in the frequency of product blowing between plants that have the same stock supplier, variation in the intensity of blowing between vacuum packs sold by different retailers, and variation in the intensity of blowing occurring between vacuum packs of the same shipment.

4.2.6 Factors affecting time to the onset of “blown pack” spoilage

The influence of packaging techniques and storage temperature on the onset of ”blown pack” spoilage was the subject of a study commissioned by the New Zealand Game Industry Board and Cryovac Sealed Air Corporation (Broda, Penney, Miller et al., 2002). This multivariant study investigated individual process variables in the context of earlier research reports (Bell, Moorhead and Broda, 1999; 2001) indicating that heat shrinking of vacuum-packs accelerated the onset of clostridial pack blowing. The study found that post-packaging storage temperatures, levels of *Cl. estertheticum* spores present initially on venison, type of packaging film, heat shrinking temperatures and duration, post-shrinking immersion chilling of vacuum packs and cut size, each individually and collectively influence time to the onset and severity, of “blown pack” spoilage.

The single most important factor that accelerates the onset of “blown pack” spoilage is post-packaging storage temperature. Even a modest 1.5°C rise in storage temperature above the -1.5°C optimum reduces the time to gas production by 33%.

Over the long storage periods employed in commercial trade, even the presence of low numbers of *Cl. estertheticum* spores can produce pack blowing. A preliminary study indicated that as few as 6 spores per cm^2 of meat surface can cause the onset of “blown pack” spoilage (Broda, Boerema, Miller et al., 2002). In practical terms, the quantitative relationship between *Cl. estertheticum* spore load in faeces/on hides of

slaughter animals and later on dressed carcasses, and time to the onset of pack blowing needs to be assessed.

Of other factors, the use of Cryovac BB7L packs, 78°C rather than 83°C shrinking temperature, 1-2 s rather than 7-8 s shrinking time, post-shrinking immersion cooling and small cut size, each delay the onset of "blown pack" spoilage.

4.2.7 Control of "blown pack" spoilage and its causative agents

The rationale behind "blown pack" spoilage control

Typically, an attempt to control the incidence of meat spoilage is best initiated by identifying the sources of the causative agents in a processing plant environment. Such knowledge of processing plant reservoirs for carcass contamination with "blown pack"-causing clostridia opens the possibility of intervention to eliminate the spoilage. Once contamination sources are identified, either they can be eliminated or control measures can be developed to (1) stop the transfer of "blown pack"-causing clostridia from their sources onto dressed carcasses, (2) remove clostridia from carcasses, and (3) prevent clostridia that are present on carcasses from growing.

Identification of processing plant sources of "blown pack"-causing clostridia

Comprehensive surveys of a number of New Zealand processing plants showed that "blown pack"-causing clostridia enter the abattoir on hides and in faeces of slaughter animals (Boerema, Broda and Bell, 2000 a and b; confidential consultancy reports). Consequently, in the majority of plants affected by "blown pack" spoilage primary sources for carcass contamination with these microorganisms cannot be eliminated.

Due to the continuous entry of "blown pack"-causing clostridia to, and their ubiquitous presence in, a processing plant environment, some processing operations are at risk of becoming colonised by these microorganisms. As experienced by some processing plants, vacuum packaging machines, fans, and even refrigeration units may occasionally become secondary contamination sources from which "blown pack"-causing clostridia are spread onto carcasses/cuts. When initial incidents of "blown pack" spoilage occur, it is recommended that a processor conducts a thorough process assessment and perhaps molecular microbiological screening to determine whether its processing operations contribute to secondary carcass/cut contamination with "blown pack"-causing clostridia. Should "blown-pack"-causing clostridia be harboured at processing operations other than those involving handling pelt-on carcasses, a cleaning/sanitising regimen must be established to eliminate "blown-pack"-causing clostridia from these secondary contamination sources.

Prevention of transfer of clostridial spores from the source onto carcasses

With primary sources of “blown pack”-causing clostridia in a processing plant being the slaughter animals themselves, an expectation that the surface of a dressed carcass and/or raw cut be free from these microorganisms is unrealistic. In a commercial processing plant environment, hides cannot be removed aseptically and even with the most hygienic dressing techniques, some transfer of clostridial spores from hides onto dressed carcass surface will occur. However, the extent of this transfer may be significantly limited by applying stringent hygienic measures to the conduct of dressing, especially first-opening cuts. Contamination of dressed carcasses can be further reduced by minimising contact between carcass flesh and hides during hide removal or between carcass and intestinal contents during dressing and evisceration.

Direction of air circulation is paramount in controlling air-borne contamination of dressed carcasses and unpackaged cuts with spores of ”blown-pack”-causing clostridia. Air flow in the whole processing plant and, specifically, slaughter floor(s) and boning room(s) should be from “clean” to “dirty” at all time. Physical design of slaughter floor, location of air intakes and exhausts, and work and product flows may each contribute to the air borne spread of “blown pack”-causing clostridia. These production features need to be periodically assessed with respect to their contribution to carcass contamination.

Transfer of “blown-pack”-causing clostridia from carcasses/cuts carrying high numbers of clostridial spores onto clean carcasses/cuts frequently occurs via direct contact during carcass cooling and, during carcass boning and cut packaging. It is recommended that direct carcass contact in chillers and cut contact on boning tables/conveyers be avoided.

It is thought that control measures which result in reduced transfer of spores of “blown-pack”-causing clostridia from hides onto dressed carcasses are preventing carcass contamination in the first place, and, consequently, are likely to have the greatest impact on subsequent onset of “blown pack” spoilage. The importance of dressing hygiene in “blown pack” spoilage control cannot be overemphasised.

Removal of “blown-pack”-causing clostridia from carcasses

Little is known about the efficacy of carcass decontamination measures on removal of spores of “blown pack”-causing clostridia from dressed carcasses. In the past decade, only limited use of carcass decontamination techniques was approved in New Zealand

and, consequently, research on control of pack blowing clostridia has focused on alternative measures.

In processing plants overseas, steam vacuum and hot steam decontamination are routinely used at pre- and post-evisceration and sequential interventions, including hot water wash, cold water wash and acid wash are applied to the carcasses prior to their entry to the carcass chillers. Preliminary research conducted at MIRINZ showed that the onset of "blown pack" spoilage has not been significantly delayed for meat carrying spores of "blown-pack" causing clostridia that underwent steam vacuum treatment (Broda, Boerema, Miller et al., 2002). Hot steam is unlikely to destroy heat-resistant clostridial spores. Consequently, both steam vacuuming and hot steam treatments may not be commercially effective for controlling "blown pack" spoilage. Similarly, there is little evidence on the efficacy of sequential interventions for the removal or destruction of spores of psychrophilic clostridia from dressed carcasses. Both warm pre-evisceration and subsequent hot/cold washes help to spread clostridial contamination onto carcass sites that normally carry a low load of clostridial spores. As yet, no scientific data is available on the efficacy of acids or acid-based preparations for removal or destruction of spores of "blown pack"-causing clostridia.

It is recommended that the efficacy of decontamination techniques is carefully evaluated before introducing new, and/or re-adjusting existing, interventions to enable removal of spores of "blown pack"-causing clostridia from carcasses. It is recognised that the majority of carcass decontamination technologies target, and are most effective against, Gram negative microorganisms, but their efficacy against Gram positive, and especially spore-forming, microorganisms is limited. Consequently, the destruction of Gram negative bacteria may enable better growth of "blown-pack"-causing clostridia on meat due to the elimination of their natural competitors. On the other hand, abrupt changes in the chemical composition of anti-bacterial washes to enable their increased sporicidal action may result in enhanced survival of Gram negative bacteria, including many food-borne pathogens.

Prevention of germination, outgrowth and multiplication of "blown-pack"-causing clostridia

Due to an inevitable presence of spores of "blown pack"-causing clostridia on carcass meat, the most practical way of preventing "blown pack" spoilage appears to be by inhibiting their multiplication in vacuum-packed product.

Generally, thermal inactivation, food preservatives that lower pH or water activity (a_w), and curing agents can control growth of "blown-pack"-causing clostridia in meats by

inhibiting outgrowth of clostridial spores. With fresh meat however, spoilage control cannot rely on thermal inactivation or additives. Of alternative measures, no single control step proved effective for preventing the onset of “blown pack” spoilage (Broda, Penney, Miller et al., 2002). However, a combination of approaches was found effective for delaying the onset of pack blowing for venison carrying low or high numbers of *Cl. estertheticum* clostridial spores beyond nominal shelf life expiry date of normal vacuum-packed chilled meat. For small venison cuts packed in BB7L packaging film, this combination consisted of heat shrinking at 78°C for 1 to 2 s and post-shrinking immersion in water at 4 to 5°C for 4s, followed by post-packaging storage at -1.5°C.

The single most important factor that inhibits multiplication of “blown pack”-causing clostridia is post-packaging storage temperature. Previous research showed that, following the application of optimised heat shrinking regimen, holding meat that carries *Cl. estertheticum* spores at -1.5°C effectively prevents clostridial gas production in vacuum-packs for up to 105 days. Therefore, the top priority for preventing the onset of “blown pack” spoilage is maintaining the cold chain during post-packaging storage, transport and retailing of meat.

Alternative control measures

Recent developments in rapid detection of “blown pack”-causing clostridia have made it possible to screen carcasses for presence of clostridial spores prior to chiller entry. On-line identification of carcasses carrying high numbers of clostridial spores would allow meat producers to exclude high-load carcasses from the chilled market and, therefore, to gain partial control over “blown pack” spoilage. In addition, export markets would not be endangered by supplying product that subsequently spoils.

On-line screening and an exclusion from chilled market may be useful during the high risk season for “blown pack” spoilage. Seasonal changes in animal presentation for slaughter due to wet, or conversely, dry weather, and/or during the annual moult often result in an increased transfer of “blown pack”-causing clostridia from hides onto dressed carcasses and subsequently, in accelerated onset of pack blowing. Carcass testing for the presence of pack blowing clostridia during the high risk season may help processors to come to an informed decision about the fate (chill or freeze) of carcasses carrying spores of “blown-pack”-causing clostridia.

4.3 Recent developments in research on "blown pack" spoilage and its causative agents

The majority of research papers and industry reports on "blown pack" spoilage and its control were published between 2000 and 2003. The findings from these publications are included in earlier sections of this review. Except for these works, which are mainly of AgResearch (formerly MIRINZ) staff authorship, there is a dearth of published information on "blown pack"-causing clostridia and their control in a processing plant environment.

At AgResearch (formerly MIRINZ) Food Safety, research on "blown pack" spoilage was initiated in 1993 and, since then, our group has assumed world leadership in research on this spoilage condition. Recently, however, the Foundation for Research, Science and Technology decided to withdraw financial support for the programme on "blow-pack" causing clostridia. While DEEResearch and Meat and Wool Innovation continue to support clostridial programmes, the loss of the New Zealand government sponsorship effectively means that only limited resources can be made available for research on "blown pack" spoilage. Consequently, progress in research on control of this spoilage condition is much slower than in the past.

In sharp contrast with the current research effort undertaken locally, marked expansion in research on "blown pack" spoilage control can be observed overseas. The author of this review is aware of significant investments in clostridial programmes currently being made by major meat producers in USA and UK. Most of these research programmes are conducted by individual companies that operate their own molecular testing facilities, some including such specialised equipment as RealTime PCR at a cost of approximately $100,000 per each machine. Results of these programmes are covered by IP protection and are not publicly accessible.

4.4 Industry practice relevant to "blown pack" spoilage

4.4.1 Process standards and practices

At present, there are no industry agreed standards that optimise venison processing with respect to "blown pack" spoilage control. Information below summarises industry practices observed during hygiene audits of beef, lamb and venison processing plants in New Zealand and overseas.

Dressing techniques

Dressing systems are largely governed by the species being processed. Previous research has found that the location of the initial microbial load on the dressed carcass is strongly correlated with the area where the first carcass opening cuts are made. From this site, microorganisms are subsequently spread to other carcass sites during dressing, trimming and boning. With the conventional dressing system, the majority of microbial contamination is found on the flank and hindquarter areas of the carcass. With the inverted dressing system, the majority of microbial contamination is found on forequarter areas of the carcass. Depending on the dressing system used, different cuts pose the highest risk of clostridial pack blowing although inverted dressing systems offer the greatest protection of high value cuts against “blown pack” spoilage.

Considerable variation exists between meat processing facilities when evaluating hygiene of carcass opening cuts, skinning and evisceration. For the first carcass opening cuts, a “single-knife” or a “two-knives” procedure may be used. Carcass opening cuts may be carried out hygienically with knives being washed and sterilised between cuts, but knives may also be sterilised in-between animals, rather than between single cuts. In addition, opening cuts may be initiated on the “dirty” (bung) area of the carcass and continued onto “clean” (crotch and belly) areas, or may be initiated on “clean” areas. Similarly, carcass opening cuts at evisceration may be conducted with or without knife washing and sterilisation between carcasses.

Skinning may employ a manual or automated upwards or downwards hide puller. Hide pulling using an automated downward puller minimises transfer of clostridia from hides onto skinned carcasses. Hide pulling may be carried out without or with roll back, and hair or other particles of hide origin may or may not be visible on the dressed carcass. It is suggested that for maximum control of clostridia the hygiene of skinning is monitored closely for roll back and hide-flesh contact during the high-risk season for “blown pack” spoilage.

Organisation of boning

The boning operation may be organised around conveyer belts, with separate conveyers carrying cuts obtained from hind- and forequarters. Such an arrangement offers significant protection against cross-contamination between cuts that pose the lowest risk and the highest risk of clostridial pack blowing. Alternatively, within a single boning lane, the majority of unpackaged cuts are at risk of becoming cross-contaminated with “blown pack”-causing clostridia via contact with cutting board and conveyer belt surfaces, and when allowed to accumulate before packaging, via direct

contact with cuts carrying high numbers of clostridial spores. To avoid cross-contamination of the most valuable cuts (e.g. middles), the boning process should be conducted with a minimal handling of cuts, using boning tables, cutting boards and boning operators dedicated to the preparation of these cuts, rather than adding them to the normal meat flow carried on the fabrication room conveyer.

Heat shrinking

Heat shrinking conditions used in processing plants frequently differ from those recommended for cuts carrying spores of “blown-pack”-causing clostridia, but also from those recommended by packaging film manufacturers. Heat shrinking conditions may employ temperatures and times as low as 64°C for 3 s, and as high as 90°C for 10 s, with shrinking by immersion of packs in a dip tank or by spraying in a tunnel. Following heat shrinking, packs may or may not undergo cooling by immersion in, or spraying with, cold water. Post-shrinking immersion cooling in a bath of 4 to 5°C water for about 4s is likely to delay the onset of blowing in packs carrying *Cl. estertheticum* spores and can be recommended as an additional measure to control “blown pack” spoilage.

Product storage

Regardless of the meat species, MAF/NZ FSA requirement for carcass cooling is such that the deep tissue temperature is reduced below 7°C within a specified time. This requirement has its origins in early studies which determined 7°C as a temperature at which an inhibition of the growth of major food-borne pathogens occurs. Following carcass cooling, boning and packaging, vacuum-packed cuts are stored at refrigeration temperatures. While the majority of processors store packaged product at 0°C or below, there are instances when storage temperatures between 0 and 4°C are employed. There is a great variation between processing plants with regard to cooling rates that are used to bring packaged product to its final storage temperature. Similarly, temperatures experienced during product load-out, transportation and retailing differ significantly between various processing plants.

From a regulatory view point, it appears that processors may exercise significant freedom in selecting storage temperatures of the packaged product. It needs to be stressed that, with psychrophilic microorganisms such as “blown pack”-causing clostridia, temperatures around 7°C are optimal for growth, and those between -1.5°C and 4°C are most certainly within their range for growth. Even a modest rise in storage temperature above the -1.5°C optimum for product storage accelerates the onset of “blown pack” spoilage. The importance of maintaining temperatures as close as

possible to -1.5°C throughout storage, transportation and retailing of the packaged product cannot be overemphasized.

Other sources of process variation

At present, many processing plants operate pathogen reduction programmes that may include interventions, extended or modified cleaning regimens or other enhanced hygiene measures. These programmes frequently involve use of antimicrobial compounds in the form of bactericidal carcass washes, antimicrobial contact sprays (e.g. applied directly on the boning conveyers or tables) or cleaning agents. Antimicrobials most commonly target microorganisms that are natural competitors of clostridia on carcass meat and, therefore, their use may permit accelerated growth of "blown pack"-causing clostridia in vacuum-packed product.

In the past, processors were often reluctant to admit that they had a problem, because they were concerned that "blown pack" spoilage incidents may reflect poor hygiene in a plant. Paradoxically, it appears that the emergence of clostridial spoilage may, on many occasions, be a direct consequence of enhanced hygiene measures.

Variation in the hygiene surrounding pre-slaughter holding of stock, hygiene of carcass trimming, the type of carcass cooling, hot and cold carcass cross-overs during carcass transfer and holding in chillers and the boning room, product flow at boning and the organisation of the packaged product storage, may also directly or indirectly influence the spread of clostridial spores throughout an abattoir and contribute to the early onset of "blown pack" spoilage.

4.4.2 Industry identified issues relevant to "blown pack" spoilage

No surveys that might identify such issues were conducted recently.

4.5 Potential opportunities for further research

4.5.1 Interventions for "blown pack" spoilage control

The use of interventions has now been approved in New Zealand for venison companies that export to USA. The possibility exists to assess the efficacy of existing (e.g. activated lactoferrin wash, hot wash, steam vacuuming, hot steam) or new (e.g. peroxyacetic acid wash) carcass decontamination treatments on removal of spores of "blown pack"-causing clostridia from dressed carcasses.

4.5.2 Agents that block spore germination

Under mildly acidic conditions, rapid germination of clostridial spores may be induced by some chemical agents e.g. by low concentrations of sodium nitrite, glutaraldehyde or others. Such germinated spores are susceptible to even mild heat treatments. The possibility exists to develop a combination of physical and chemical treatments which, without changing sensory attributes of fresh vacuum-packed meats, would induce and inactivate clostridial spores present on vacuum-packed cuts.

4.5.3 De-hairing systems for use on the carcass opening cuts area

It is thought that control measures which result in a reduced transfer of clostridial spores from hides onto dressed carcass are preventing carcass contamination in the first place, and, consequently, are likely to have the greatest impact on subsequent onset of “blown pack” spoilage. Chemical or physical de-hairing systems may present an attractive measure for improved control of “blown pack” spoilage, as well as improved overall hygiene of manufactured meat.

4.5.4 Biopreservatives for “blown pack” spoilage control

Vacuum-packed chill-stored meat is, strictly speaking not fresh but a preserved product, with naturally-occurring populations of lactic acid bacteria (LAB) exerting bactericidal (e.g. by producing lactic acid and/or bacteriocins) or bacteriostatic (e.g. by competing for the same niche) effect against other microorganisms. It is thought that antimicrobials used in decontamination washes or sanitising sprays may have a detrimental effect on the natural preservative action of lactic acid bacteria. The possibility exists to develop LAB starter cultures producing bacteriocins against “blown pack”-causing clostridia. These cultures could be used as a biopreservative replacement of natural LAB populations to re-instate microbial balance on surfaces of manufactured meats.

4.5.5 Initial cooling rates of packaged product and an outgrowth of clostridial spores

There is a great variation between processing plants with regard to cooling rates that are used to bring packaged product to its final storage temperatures. It is thought that these initial cooling rates may, by resolving the initial lag phase influence the outgrowth of spores of “blown pack”-causing clostridia. It is proposed that the effect of initial cooling rates of vacuum-packed venison on the onset and severity of “blown pack” spoilage be determined.

4.5.6 Efficacy of cleaning agents and regimens

A variety of cleaning agents with sporicidal action are available through the commercial chemical supply companies and these agents usually range from food-grade peroxyacetic acid-based to hospital-grade glutaraldehyde-based chemicals. Processing plants that experience “blown pack” spoilage incidents often introduce changes to their existing cleaning routines, for example by conducting regular fogging with peroxyacetic acid-based cleaning agents and sanitizers (e.g. Oxonia Active or Bactipal). However, to the best of the author’s knowledge, little is known about the effect of the commercially available cleaning/sanitising agents on spores of clostridia causing “blown pack” spoilage. Until the sporicidal efficacy of these agents is evaluated, any recommendations on the use of chemicals for controlling “blown pack”-causing clostridia in the DSP environment would be conjectural.

4.5.7 Rapid detection of “blown pack”-causing clostridia

At present, a PCR kit is being developed for rapid detection of spores of “blown pack”-causing clostridia on dressed carcasses. The kit will allow early screening of dressed carcasses to determine carcasses carrying clostridial spores in numbers at or above the threshold for blowing. Following testing, positive carcasses could be excluded from the chilled market. It is proposed that the kit be further developed to the market-ready stage. In addition, to enable high throughput screening of carcasses for “blown pack”-causing clostridia, RealTime capabilities should be established.

Recently, four new species of psychrophilic clostridia were obtained from an Antarctic microbial mat. From a comparison of 16S rDNA sequences of the newly discovered species, it appears that the existing *Cl. estertheticum*- and strains K21/K24-specific primers are likely to give PCR product when the DNA of any of these new species is present in the PCR template. It is imperative that the newly described species are tested to determine their ability to cause “blown pack” spoilage of vacuum-packed chilled venison. Should the ability of these microorganisms to cause “blown pack” spoilage be confirmed, the existing 16S rDNA-based method will remain suitable for detection of causative agents of this spoilage condition. However, if any of these species is found incapable of causing pack blowing, alternative procedure based on a DNA target that differentiates between pack blowing and non-blowing clostridia will need to be developed.

4.6 Conclusions and recommendations

“Blown pack”-causing clostridia are very prevalent in New Zealand slaughter stock and processing plants. Consequently, their control in a processing plant environment is exceptionally difficult. To-date, no single control measure has proven effective for

controlling “blown pack” spoilage. On the basis of current knowledge on “blown pack” spoilage control, it appears that by using a combination of control steps, incremental gains in shelf life of the product can be obtained, so that the nominal shelf life of vacuum-packed chilled venison can be achieved for product carrying clostridial spores. Most practical measures for reducing or preventing the onset of “blown pack” spoilage must assure that (1) the lowest possible numbers of clostridial spores are transferred onto the carcass during dressing, and (2) germination and outgrowth of remaining spores are kept to a minimum by maintaining the cold chain throughout product storage, transport and retailing; and by using the optimal regimen for heat shrinking of vacuum-packed product.

On the basis of current knowledge on “blown pack” spoilage control, the following can be recommended:

- Identify reservoirs for “blown pack”-causing clostridia carcass contamination in DSP’s affected by “blown pack” spoilage condition.
- For enhanced control of “blown pack” spoilage, undertake individual processing audits of venison plants affected by this spoilage condition.
- Develop deer processing industry standards that optimise processing with respect to control of “blown pack” spoilage.
- Continue research on measures to control “blown pack” spoilage.
- Consider ways of assuring research continuity and retaining research staff dedicated to projects on “blown pack”-causing clostridia, e.g. by funding a PhD studentship in this research area.

5 Summary of Process Improvements and Opportunities Identified in this Document

5.1 Processing

5.1.1 Industry initiatives

Benchmark existing quality

Experience from beef and lamb benchmarking has demonstrated that there have been significant shifts in quality outcomes over the last 10 years. Results from venison tenderness testing suggest that this may also be the case for frozen and chilled venison.

To address this, the recommendation is to benchmark venison quality from several North and South Island based venison plants under controlled conditions. This process should utilise the following protocol:

1. Initial plant audits to measure pre-rigor pH fall, loin and deep leg muscle cooling.
2. Standardised sample collection.
3. Ultimate pH measured.
4. Shear force measured in frozen product. Chilled product shear force measured after different periods of ageing that would be representative of delivery times to market.
5. Drip and purge measured in chilled product.
6. Simulated retail display and colour stability using colour meter and consumer evaluation.

Objective Measurement protocols

To develop a robust data base of other quality attributes in addition to tenderness. This could be conducted in NZ on product direct from the processing plants and also in the market.

1. Ultimate pH.
2. Purge or drip during vacuum packed aging and after simulated retail display.
3. Colour stability.

Complete revision of tenderness testing protocols

- Sample selection numbers need to be revised to be representative of current throughputs.
- Increased frequency of testing.
- All product types should be tested e.g. frozen product, short-term chilled (< 1 week, longer-term chilled (3 weeks, 6 weeks etc).
- Tenderness testing should be carried out using revised protocols and include other cuts in addition to striploin.

Effect of Halal stunning on pre-rigor pH fall and temperature decline

Clearly, Halal stunning is an acceptable stunning method for venison. The use of Halal slaughter would open up new market opportunities for venison. However, the use of electrical head-only (Halal) stunning will impact upon subsequent pH fall and thus quality outcomes. Therefore, Halal stunning and slaughter can be introduced, but the corollary to this is that other process modifications may be required to ensure there is no deterioration in quality.

Plants to measure ultimate pH

- Measure ultimate pH of all carcasses and remove those that have a pH of ≥ 5.8 from the chilled consignments.

5.1.2 Research opportunities

- Define processing specifications for frozen, chilled frozen, chilled (1 week to market) and chilled (3 – weeks to market product)
- Evaluate use of advanced low voltage stimulation procedures in venison
- Evaluate use of hot boning deep leg muscles. Combine with immersion chilling and shape manipulation technologies
- Develop texture profiles for chilled venison after different periods of storage.

5.2 Value adding

- The use of CAP technology for venison should be re-evaluated as it may be able to offer new opportunities for chilled venison
- The use carbon monoxide in pack atmospheres should be investigated as it may offer significant commercial advantages for the retail display of venison

- Adding value work similar to the that carried out for beef and lamb
- Value should be added to venison offal.

5.3 "Blown-pack" causing clostridia

- Identify reservoirs for carcass contamination with "blown pack"-causing clostridia in venison processing plants.
- For enhanced control of "blown pack" spoilage, undertake individual audits of processing plants affected by this spoilage condition.
- Develop industry standards that optimise processing with respect to control of "blown pack" spoilage.
- Continue research on measures to control "blown pack" spoilage.
- Consider ways of assuring research continuity and retaining research staff dedicated to clostridial projects, e.g. by funding a PhD studentship in this research area.

5.4 Industry consultation on viability of new opportunities as identified in this document

Research priorities have been assessed using

1. the scoring of the 11 topics that was put forward at a recent venison processors technical meeting as identified below and

2. asking many of the contributors (as identified on the covering page of this document) to comment on some of the research opportunities that have been identified in this document.

Table 4 Scoring of recently submitted EOI's	
Title	**Rating**
Survey on pre-slaughter treatment of deer	H
Timing the occurrence of bruises in deer carcasses	M/H
Mobile slaughter for improved welfare and quality	L
Pre-slaughter handling of deer	M/L
Technologies to extend chilled storage life	M
Tenderising or ageing prior to freezing	H
Benchmarking venison quality and tailoring processing for different markets	H
Packaging systems to assist colour retention	M/L
New product options for the forequarter and lower quality venison cuts	M
Added value product options for venison	M
Proactive food safety - Johnes Disease	H
Control of blown pack spoilage – venison	H

Rating based upon H – high, M – medium, L – low.

Generally it was agreed that priorities in the area of processing were to benchmark existing venison quality. While it is clear that the tenderness of chilled product that takes several weeks to get to market is highly acceptable, most contributors commented that there was significant concern regarding the tenderness of frozen and short-term chilled product. Notwithstanding this, some felt that there were problems with high purge levels and poor texture and flavour in long-term chilled product. Thus, processing options that may overcome some of these issues were regarded favourably.

While the majority of processors and other industry members are extremely interested in the opportunities of value-adding to venison, investment in these areas was not considered high priority at this stage. However, many commented that they would be interested in pursuing these opportunities on an individual company basis.

Similarly, research to identify techniques to reduce "blown-pack"-causing cloistridia spoilage was considered of high value and interest to the industry.

5.5 Research providers

A number of organisations are carrying out research on deer, both in New Zealand and internationally (Table 5). However, details from some companies are limited prior to the publication of results. The largest amount of research available apart from AgResearch is from Rural Industries Research and Development Corporation (RIRDC) in Australia, who also publish a comprehensive international bibliography of deer research publications.

Table 5. List of companies currently carrying out deer research		
Company	***Website***	***Research***
New Zealand		
AgResearch	www.agresearch.co.nz	Production, animal welfare, meat science, food safety, microbiology, velveting, added value.
MAF sustainable farming fund	www.maf.govt.nz	Production
Lincoln University	www.lincoln.ac.nz	Reproduction and physiology
Massey University	www.massey.ac.nz	Production, animal welfare, meat science
Otago University	www.otago.ac.nz	Production
International		
RIRDC	www.rirdc.gov.au	Production, velveting, transport, processing, meat science, added value
Deer farmer	www.deerfarmer.com	Production
Gatton deer research	www.uq.edu.au	Production, meat science
MaCaulay Institute	www.mluri.sari.ac.uk	Production
ADAS	www.adas.co.uk	Production, product quality, added value
Bristol University,	www.bristol.ac.uk	Microbiology

5.6 Current research programmes in the area of venison processing, value adding and "blown-pack"-causing clostridia contamination

5.6.1 Processing & value adding

In the area of processing for quality and value-adding, the only current work that could be identified either in New Zealand or internationally, is an objective in the current AgResearch FRST funded programme entitled Transforming deer industry through niche foods strategy - Contract number C10X0202. This objective commenced in July 2002 and will run until 2004, although there is an expectation that funding for this work will continue for the full 4-year period. The background and details of this work are identified below.

Venison has a shorter retail display life when compared to beef and lamb. While this is not an issue for catering outlets and restaurants, it is for retail outlets where product is displayed either in over-wrapped or modified atmosphere packs.

Trout & Gutzke (1995) compared colour stability of venison, beef, lamb and pork; In 1 day old overwrapped product displayed at 5°C, there was a three-fold difference in discolouration rate between pork, which was the most colour stable, and venison which was the least. This translates into just 1.6 days of acceptable colour (Stevenson-Barry, 1998).

Unfortunately, the reasons why venison should have such poor colour stability when compared to other species is not clear, although the current Agresearch research programme is exploring this issue and developing interventions that can be applied to enhance the visual appeal of chilled venison. In other areas of this report, possible strategies to overcome this commercially important issue have been put forward.

The time taken for meat to turn brown is the balance of reactions that oxidise myoglobin to the brown state (oxidation reaction) and reactions that reduce the brown, oxidised myoglobin back to its original state (reduction reactions). Both sides of the process need to be understood and evaluated.

We have developed a new procedure for measuring the ability of venison to reduce oxidised myoglobin back to its normal state. We have been able to measure the effects of processing (chilling rate, electrical stimulation) on the activity of the reduction reaction in venison, and how fast this activity decays during chilled storage. Upon completion of this work, we hope to be able to identify which chemical step in the reduction reaction is the weak link, and investigate methods of maintaining its activity.

The second part of the equation is the oxidation reaction. We are developing a method of measuring how vigorous the oxidation reactions are in venison, and how effectively the antioxidants that are normally present in meat are at suppressing these reactions. When this work is complete, we expect to measure how these reactions change during chilled storage, and to develop processing and storage procedures that suppress the oxidation reactions and encourage the antioxidant potential in venison.

As an initial stage to this work, it was clearly demonstrated that an increased rate of metmyglobin in venison was not due to lower levels of vitamin E levels when compared to grain fed beef (Stevenson-Barry, 1999a). However, the display life of venison could be significantly enhanced by the introduction of vitamin C either by drenching the animal prior to slaughter (Stevenson-Barry, *pers comm*) or by applying directly to the meat (Stevenson-Barry et al, 1999b).

It is likely that other naturally occurring anti-oxidants exist and thus could be added to venison in the manner reported above. These possibilities continue to be explored within the scope of this FRST funded work.

5.6.2 "Blown pack"-causing Clostridia

The DEEResearch-funded project aims to develop a rapid detection kit that will allow early screening of dressed carcasses for the presence of "blown pack"-causing clostridia. Following this screening, venison producers will be able to select the appropriate meat processing procedure prior to packaging/distribution of the product. Venison carrying clostridial spores could then be frozen and/or distributed locally. The project is progressing well and will be reported by mid November 2003.

Previous results have indicated that the onset of "blown pack" spoilage could be suppressed by using bacteriocins (Kalchayanand, Ray and Field, 1993). A current Meat New Zealand-funded project aims to determine the efficacy of *Brochothrix campestris* bacteriocins against "blown pack"-causing clostridia. Results to-date indicate that while these bacteriocins inhibit the outgrowth of *Cl. estertheticum* spores *in vitro*, their use does not appear to significantly extend the time to onset of "blown pack" spoilage of vacuum-packed meat carrying clostridial spores.

6 Popular summary article

After consultation with venison processors, DEEResearch requested a review of the research around venison processing as one mechanism to assist in identifying process improvements and new opportunities for the venison industry. To establish the scope of the review, input was sought from members of the processors technical committee, and people associated with the industry who have responsibilities for processing quality, auditing and final product quality and safety in the market. This consultative process resulted in three areas of interest that it was agreed would be covered in this review: processing for quality, value-adding opportunities and blown pack spoilage.

Current industry processing standards have been in place for several years and the majority of processors are working to these agreed standards. However, a number of quality issues are of concern to processors and it is recognised that new opportunities that have been exploited in the beef and lamb industries could be successfully applied in venison processing.

While the tenderness of chilled venison is generally highly acceptable, high purge losses in vacuum packs and poor retail colour stability are acknowledged as issues that can impact upon market acceptance. Similarly, many processors are identifying problems with the tenderness of their frozen product and are finding challenges meeting the tenderness specification as laid down in the industry standards when freezing is carried out one or two days following slaughter.

The original work to establish a process that would reliably generate tender chilled and frozen venison was carried out in 1991. However, recent benchmarking quality work in lamb has shown that, despite improved processing consistency over the last 15 years, product tenderness is falling well short of the specification. The reasons for this non-compliance are probably different sheep genetics in combination with modified production systems, leading to carcasses with different responses to processing conditions. There is some likelihood that venison may be suffering from a similar phenomenon and we would recommend that a quality benchmarking exercise be undertaken.

In the past five years, “blown pack” spoilage of vacuum-packed chilled meats has advanced from being considered a scientific curio to being recognised worldwide as a spoilage condition causing major economic losses in the beef, venison and lamb processing industries. This spoilage condition is caused by spore-forming anaerobic

microorganisms that are capable of growth and abundant gas production at refrigeration temperatures. This review describes current status of knowledge on characteristics of “blown pack” spoilage and its causative agents, environmental reservoirs of “blown pack”-causing organisms, their incidence in foods the DSP environment, and methods for their detection. Present recommendations for control of these microorganisms in a processing plant environment are discussed, with the emphasis on new opportunities and research directions for the future.

Given the changes in animal physiology, production systems, their associated microbiology, together with shifts in market expectations of product quality, it is undoubtedly appropriate to re-examine processing specifications for the venison industry. Recent work with beef and lamb has shown that different combinations of stunning, stimulation and chilling can be used to dramatically manipulate product quality, and so tailor processing conditions to different market requirements. Similar opportunities exist in venison to define a range of optimised processing specifications for frozen, short-term and long-term chilled product to ensure that, once in the market, the quality attributes match or exceed customer expectations.

Similarly, quantum leaps have recently been made in the area of value-adding to manufacturing grade beef and lamb by understanding and exploiting attributes of individual muscles. While limited information exists in this area for venison, fundamental knowledge that has been developed to utilise this technology for beef and lamb, can now be readily applied to venison.

The review discusses the principles underlying the application of process tailoring and value adding and outlines the potential to apply this underpinning knowledge to venison. It is important that such work remains dynamic and that, as new processing technologies evolve into commercial reality, the existing specifications can be refined to incorporate these new technologies. Some new processing opportunities that are rapidly approaching commercial reality are also discussed. Strategies by which some of these can be incorporated into venison processing have also been explored.

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Appendix 1. Tenderness Standard

Objective

To ensure that every carcass and every cut of meat is processed and handled in such a manner that the meat achieves the desired level of tenderness and food safety requirements.

Standard

Every carcass will be electrically stimulated and chilled to achieve the following tenderness standard.

The mean tenderness of the M. longissimus thoraces et labarum measured at the butt end, following 21 days ageing at -1^{o}C (± 0.5^{o}C) shall be less than or equal to 5 kg Force Score units, determined using standard procedures and a MIRINZ Tenderometer. There shall be no scores above 10 kg while 90% of all samples will have values of 8 kg or less.

Process Variations

1. Low Voltage Electrical Stimulation
    a. Time from stunning to low voltage stimulation: Not more than 5 minutes.
    b. Points in the slaughter dressing process where stimulation may occur:
        i. After stunning, but before sticking and shackling
        ii. After sticking, but before shackling and hoisting
        iii. After hoisting to the rail.
    c. For hall slaughter there can be no stimulation until the throat is cut.
    d. Where electric stun includes heart stop, electrical stimulation can be applied prior to sticking.
    e. Electrode placement on carcass – shall be placed such as to achieve effective stimulation. Generally one electrode (heavy duty battery clamp) on upper or lower lip of the jaw, the other the anus, or other appropriate site.

The operation of the stimulator shall be verified by either:

- the use of a stimulation monitor, or

- Undertaking a monthly electrical parameter check by registered electrician with results meeting parameters outlined in this standard. The parameters shall be recorded “as found” and “as left” readings.

A certificate of calibration is required from the organisation used.

In addition daily plant checks to ensure correct use shall include:

- measurement of the stimulation time
- inspection of leads for damage

f. Where a monitor is used it shall

  – Be approved by MIRINZ as being capable of measuring the stimulation parameters outlined in this standard

  – Be calibrated at least annually.