
Accepted Manuscript

Genetic and environmental effects on meat quality


PII: S0309-1740(10)00177-4
Reference: MESC 5083

To appear in: Meat Science

Received date: 24 February 2010
Revised date: 28 April 2010
Accepted date: 30 April 2010


This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.
Genetic and environmental effects on meat quality

R. D. Warner\textsuperscript{1*}, P.L. Greenwood\textsuperscript{2}, D.W. Pethick\textsuperscript{3} and D.M. Ferguson\textsuperscript{4}

\textsuperscript{1} Department of Primary Industries, 600 Sneydes Rd, Werribee, 3030, Australia
\textsuperscript{2} Industry & Investment NSW, Beef Industry Centre of Excellence, University of New England, Armidale NSW 2351, Australia
\textsuperscript{3} Department of Veterinary and Biomedical Sciences, Murdoch University, WA, Australia
\textsuperscript{4} CSIRO Livestock Industries, FD McMaster Laboratory, Chiswick, Locked Bag 1 Armidale NSW 2350, Australia.

* Corresponding author. Telephone +61 3 97420477. Email address robyn.warner@dpi.vic.gov.au
ABSTRACT

In order for livestock industries to consistently produce high quality meat, there must be an understanding of the factors that cause quality to vary, as well as the contribution of genetics. A brief overview of meat tenderness is presented to understand how genotype and environment may interact to influence this trait. Essentially, meat tenderness is determined from the contribution of connective tissue, sarcomere length determined pre-rigor and rate of proteolysis during ageing, as well as contributions from intramuscular fat and post-mortem energy metabolism. The influence of mutations in myostatin, the callipyge gene, the Carwell or ribe eye muscle gene as well as the calpain system on meat tenderness is presented. Specific examples of interactions between the production or processing environment and genetics are presented for both sheep and cattle. The day-to-day variation in tenderness is evident across experiments and this variation needs to be controlled in order to consistently produce tender meat.

Keywords – genetics, environment, GxE, meat quality, stress, tenderness, consumer, heritability, cattle, sheep

CONTENTS

1 Introduction ............................................................................................................ 3
2 Meat quality defined .............................................................................................. 6
3 A brief overview of meat tenderness ................................................................. 7
   3.1 Effects of sarcomere length ................................................................. 8
1 Introduction

The supply of meat which is wholesome, safe, nutritious, and of high quality to the consumer will ensure continued consumption of meat. In affluent countries, consumers are increasingly demanding meat products which are of high quality 100% of the time. In order for livestock industries to consistently produce high quality meat, there must be an understanding of the factors that cause quality to vary, and implementation of management systems to minimise quality variation.

Beef and sheep meat quality are the primary subjects of this review, though reference is made to evidence from pigs, where the results are considered applicable. There have previously been reviews on the effect of genetics on meat quality of cattle (Burrow, Moore, Johnston, Barendse & Bindon, 2001; Hocquette, Lehnert, Barendse,
Cassar-Malek & Picard, 2007; Lehnert, Wang, Tan & Reverter, 2006; Mullen, Pannier & Hamill, 2009) and sheep (Bishop & Karamichou, 2009; Safari, Fogarty & Gilmour, 2005; Warner, Pethick, Greenwood, Ponnampalam, Banks & Hopkins, 2007b). Obviously there are also reviews on the multitude of effects of environment on meat quality. This review focuses on the relative contribution of genetics and environment, and any interactions, in determining meat quality. Recent research is discussed as well as specific genes known to influence meat quality in cattle and sheep.

The environmental effects on meat quality are best defined as those not attributable to genetics, and include on-farm, pre-slaughter, and post-slaughter processing factors. Genetics is defined as heredity and a unit of heredity that occupies a specific locus on a chromosome is defined as a gene; a sequence of bases on a DNA molecule (Stenesh, 1989). While there are many examples of single-gene-locus traits, current thinking in biology discredits the notion that a small number of genes, and that genes alone, can determine most complex traits. At the molecular level, DNA interacts with signals from other genes and from the environment. At the level of individuals, particular genes influence the development of a trait in the context of a particular environment. Thus, measurements of the degree to which a trait is influenced by genes versus environment will depend on the particular environment and genes examined.

It is recognised that environmental influences, eg. in utero nutrition, can have effects beyond a single generation (Gluckman, Hanson & Beedle, 2007), which is the study of epigenetics. In addition, mitochondrial DNA is transmitted exclusively through dam lines and a mitochondrial maternal effect has been described (Burrow, 2001).
The effect has been defined as an effect of dam on offspring performance, additional to the direct additive, maternal and permanent environmental contributions and may have both a genetic and environmental origin (Burrow, 2001). Epigenetics and maternal effects through mitochondrial DNA are potentially exciting new areas of research but are beyond the scope of this review.

Intra- and inter-muscle variation in a trait also occurs thus the effect of genes on a trait will depend on the muscle, and the location within a muscle, and should be defined as such. Generally, muscles respond to their environment as they grow and develop, eg. tension on a muscle, nerve supply, blood supply carrying nutrients and oxygen, frequency of nerve stimulation, hormonal messages, etc. Thus a muscle develops partly in response to its environment, as well as in response to its genes.

A phenotype can be simply described as the sum of the genetic and environmental variation in a measured outcome (trait) such as tenderness or intramuscular fat. Phenotype can also be described as a result of the interaction between genotype and environment, in addition to variation not readily attributable to either (Peaston and Whitelaw, 2006) Heritability is the proportion of phenotypic variation in a population that is attributable to genetic variation among individuals.

Variation in a trait of interest can also be influenced by genetic-environment (G x E) interactions where the expression of a genetic trait, or genotype, changes in response to the environment. In our review of the literature, we found seven studies where there was some evidence of a G x E interaction for meat quality traits and these are
presented in the relevant sections. This evidence indicates that G x E interactions may be a relatively important contributor to the phenotypic variance in meat quality traits.

2 Meat quality defined

Meat quality is defined by those traits the consumer perceives as desirable which includes both visual and sensory traits and credence traits of safety, health and more intangible traits such as ‘clean’ and ‘green’ or welfare status of the production system (Becker, 2000). Important visual traits include; colour and texture of the meat, fat colour, amount and distribution of fat as well as the absence of excess water (purge) in the tray (Glitsch, 2000). Once cooked, consumer satisfaction is largely determined by how tender the meat is as well as its flavour/odour and juiciness (Glitsch, 2000). Consumers of lamb in Australia usually place the highest weighting on flavour/odour, followed by tenderness and lastly juiciness (Pethick, Pleasants, Gee, Hopkins & Ross, 2006). This is in contrast to consumers of beef who generally rate tenderness as the most important palatability trait (Huffman, Miller, Hoover, Wu, Brittin & Ramsey, 1996; Robinson, Ferguson, Oddy, Perry & Thompson, 2001; Watson, Gee, Polkinghorne & Porter, 2008) although this can vary with country (Glitsch, 2000). Recent consumer data from Meat Standards Australia indicate that flavour has increased in importance to beef consumers, most likely as a result of reducing the variation in tenderness (Rod Polkinghorne, pers. comm.).

Over the last two decades in Australia, the genetic and environmental contributions to the variation in beef and sheepmeat quality traits, particularly tenderness, have been
characterised and these will be discussed further within this review. The results from large scale beef and lamb progeny evaluation programs in Australia show that despite our knowledge, there is still considerable variation in meat tenderness which remains unexplained. This review will focus primarily on meat tenderness simply because quantification of the genetic and non-genetic variation in beef tenderness, and to a limited extent sheep tenderness, has received more attention compared with the other quality traits such as colour, flavour and water-holding capacity.

3 A brief overview of meat tenderness

A brief overview of meat tenderness is presented in order to understand how genotype and environment may interact to influence this trait. Tenderness is an important meat quality trait and the biological, structural and physiological mechanisms underlying meat tenderness have been extensively investigated (Dransfield & Jones, 1980; Harper, 1999; Koohmaraie, 1988; Tornberg, 1996). Essentially, meat tenderness is determined by the amount and solubility of connective tissue, sarcomere shortening during rigor development, and postmortem proteolysis of myofibrillar and myofibrillar-associated proteins (Koohmaraie & Geesink, 2006). Intramuscular fat also indirectly influences meat tenderness (Harper, 1999; Hocquette, Gondret, Baéza, Médale, Jurie & Pethick, 2010; Nishimura, Hattori & Takahashi, 1999; Tornberg, 1996) as does the rate and extent of post-mortem energy metabolism (Thompson, Perry, Daly, Gardner, Johnston and Pethick, 2006. These are discussed below.

Interactions occur between these factors which can be difficult to separate (Harper, 1999) and each of these is influenced to a greater or lesser degree by genotype and the
pre- or post-slaughter environment. Furthermore, each of these factors are generally non-linear in their effect on meat tenderness, with continuing debate over the importance of each factor, depending on the muscle, the species, the age of the animal and the data available.

3.1 Effects of sarcomere length

Early work showed the pivotal effect of muscle sarcomere length on meat tenderness (Marsh & Leet, 1966) and the phenomenon of cold-shortening has been extensively discussed (Hwang, Devine & Hopkins, 2003). The effect of sarcomere length on meat toughness was traditionally thought to be mediated through the increased overlap of myofilaments (Dransfield & Rhodes, 1976) and that the toughness comes from the myofibrillar structure (see Lepetit, Grajales & Favier, 2000). Rowe (1974) observed that the angle that perimysium collagen fibres make with muscle fibres is dependent on post-rigor sarcomere length. More recently, Lepetit et al. (2000) showed that the amplitude of cold-shortening, and thus toughening, is dependent on the muscle collagen content and the toughening is strongly influenced by the cooking temperature.

Any one muscle will of course not have uniform sarcomere lengths throughout as each muscle fibre goes into rigor at different times. At intermediate rates of temperature decline, minimal shortening of sarcomeres occurs at rigor (Marsh, Ringkob, Russell, Swartz & Pagel, 1987b; Pike, Ringkob, Beekman, Koh & Gerthoffer, 1993). If the temperature decline is too rapid and glycolysis is slow, cold-shortening occurs resulting in a profound increase in toughness (Marsh et al., 1987b; Pike et al., 1993). If the temperature fall is too slow, and glycolysis is fast, rigor-
toughening can occur with associated tougher meat, due to a failure of the tenderising processing during ageing (Dransfield, 1994; Marsh, Ringkob, Russell, Swartz & Pagel, 1987a; Pike et al., 1993). Once rigor occurs, sarcomere length does not generally change with ageing (Koohmarai, Doumit & Wheeler, 1996). Thus the normal pre-rigor restraint on muscles through tendons and attachments, which can be increased through the post-slaughter application of tenderstretch (hanging by the aitch bone), can minimise the degree of sarcomere shortening and associated toughening.

3.2 Effects of proteolysis post-slaughter

The proteolysis contribution to meat tenderness is predominantly regulated by the protease levels in the muscle at slaughter, duration of post-rigor ageing and protease activity during ageing (Koohmarai et al., 2006). Sarcomere length at rigor also has been shown to contribute to proteolysis post-slaughter through its effect on limiting access of proteases to myofibrillar protein substrate in cold-shortened beef semitendinosus (Weaver, Bowker & Gerrard, 2008).

The mechanism of post-mortem tenderisation has been described in detail by Koohmarai (1994), Taylor, Geesink, Thompson, Koohmarai and Goll (1995) and Tornberg (1996). The rate and extent of post mortem proteolysis by the calpain system explains the majority of the observed improvement in tenderness with ageing of meat (Koohmarai, 1994; Taylor et al., 1995). The activity of these proteolytic enzymes is also regulated by the rate of pH and temperature decline during rigor development (Dransfield, 1994), ionic strength (Ouali, 1984), oxidation of calpain post-slaughter (Rowe, Maddock, Lonergan & Huff-Lonergan, 2004) and possibly other factors.
The effect of the calpain system on tenderisation post-slaughter relies on a balance between the rate of activation and activity, and the rate of inactivation or denaturation of the proteolytic enzymes (Dransfield, 1992; Dransfield, 1993; Dransfield, 1994; Dransfield, Etherington & Taylor, 1992; Dransfield, Wakefield & Parkman, 1992). The rate of activation and activity is reliant on the concentrations of calpain 1, calpain II and their inhibitor, calpastatin at slaughter and the conditions at slaughter (Koohmaraie, 1994). The inactivation of the calpain system through denaturation occurs more rapidly when the pH-temperature conditions associated with rigor-toughening occur (Dransfield, 1994).

3.3 Effects of connective tissue

The connective tissue contribution to meat tenderness is predominantly determined by the development of heat-stable cross-links as well as the total collagen content, and are predominantly established in the animal pre-slaughter (Harper, 1999) although there is some influence post-slaughter through sarcomere length (Lepetit et al., 2000).

Connective tissue is an integral component of muscle that transmits contractive forces from the myofibrils in the postnatal animal (McCormick, 1994). Connective tissue consists primarily of an extracellular matrix and is the predominant component of endomysium, perimysium and epimysium (Harper, 1999). Collagens are the major protein constituents of perimysial connective tissue (Dransfield, 1977), and as an animal matures, covalent cross-links between collagen fibrils become heat-stable and these links significantly increase meat toughness (McCormick, 1994).
Simplistically, the cross-links determine the extent of tension generated during heating and the residual adhesion between muscle fibers and hence, the greater the number of cross-links, the tougher the meat (Bailey, 1985; Light, Champion, Voyle & Bailey, 1985). It is therefore not the amount of collagen present, rather the degree of structural linkage in collagen that determines its contribution to meat tenderness (Bailey, 1985; Light et al., 1985). However as with all biology and meat science, there are always cases that do not fit our understanding. In Japanese black cattle, Nishimura et al. (1999) found that age-related increases in semitendinosus toughness were unrelated to collagen solubility or total collagen.

Connective tissue that is present within the muscle is macroscopically unaffected by slaughter *per se*, or by *rigor mortis* (Harper, 1999). It is generally considered that connective tissue does not change or degrade post-slaughter although small post-slaughter changes during tenderstretch hanging may contribute to increased tenderness (Harper, 1999). As discussed above, sarcomere shortening influences the perimysial collagen fibrils and Lepetit et al. (2000) has shown that at cooking temperatures above 60°C, the increased toughness of cold-shortened meat is derived from the myofibrillar as well as from the collagen components. The contribution of perimysial collagen in shortened muscles to the measured toughness, depends on the total collagen, the collagen thermal stability and on the cooking temperature.

### 3.4 Effects of intramuscular fat (IMF)
Marbling is defined as the intramuscular fat (IMF), or adipose tissue, deposited between perimysium surrounding muscle bundles, and is visible to the human eye as ‘flecks’ or spots of fat. Marbling is a visual score given to a piece of meat whereas IMF is the chemically measured fat content (includes membrane lipids) although the terms are often used interchangeably. Early work from the USA showed that higher levels of marbling were associated with improved palatability in cooked beef (McBee, Jr. & Wiles, 1967; Smith, Savell, Cross, Carpenter, Murphey & Aalhus, 1987) although generally marbling degree accounts for only 3-10% of the variation in sensory tenderness of beef (Nishimura et al., 1999). There has been extensive debate about the contribution that intramuscular fat makes to the sensory attributes of meat (Hocquette et al., 2010; Oddy et al., 2001).

Hocquette et al. (2010) stated that IMF directly affects juiciness and flavour, but tenderness was influenced indirectly. IMF appears to separate and dilute perimysial collagen fibres and disorganize the structure of intramuscular connective tissue that contributes to increased meat toughness (Hocquette et al., 2010). Nishimura et al. (1999) found that in heavily marbled (>8% IMF) Japanese black cattle, the perimysium is separated into thinner collagen fibrils.

In general, some IMF is important to palatability and at low IMF levels (<8%), this is most likely primarily through improved juiciness and flavour. At high IMF levels (>8%), disruption in the connective tissue in the perimysium appears to occur, which influences the toughness deriving from the structure of the muscle.
3.5 Effects of glycogen metabolism

Post-mortem energy metabolism in muscle and in particular, glycolysis and rigor onset, are important for determining sarcomere length (Thompson et al., 2006). The pre-rigor metabolism and availability of substrate, combined with muscle temperature, in any muscle in a carcass will determine the shortening occurring at rigor and thus the post-rigor sarcomere length (Koohmaraie & Geesink, 2006). As discussed above, cold-shortening and heat-toughening in muscles will influence the myofibrillar, the connective tissue and also the proteolytic contributions to meat tenderness. Thus it is evident that control of energy metabolism post-mortem is important for producing tender meat.

Muscle metabolism pre-rigor in any muscle of a carcass can vary significantly with (i) nutrition of the animal influencing pre-slaughter muscle glycogen levels (Knee, Cummins, Walker, Kearney & Warner, 2007), (ii) pre-slaughter stress influencing muscle metabolism at slaughter (Channon, Payne & Warner, 2000), (iii) muscle fibre type (Thompson et al., 2006), (iii) post-slaughter electrical stimulation of the carcass (Simmons, Daly, Mudford, Richards, Jarvis & Pleiter, 2006), (iv) genetics (Warner et al., 2006; Martin, Gardner, Thompson & Hopkins, 2006; Gardner, Kennedy, Milton & Pethick, 1998) as well as through unidentified causes. In theory, effective control of post-mortem pH and temperature decline can provide precise management of meat quality outcomes (Simmons et al., 2006) as well as allowing more precise measurement of heritability of traits of interest and variation between genotypes and breeds. In practice, this can be difficult to achieve under commercial conditions and
variable responses are usually due to animal history rather than processing conditions (Simmons et al., 2006).

Meat tenderness varies not only with the rate of glycolysis and rigor onset post-slaughter, but also with the extent of glycolysis, classically identified through the ultimate pH (pHu) achieved in a muscle. The relationship between meat pHu and tenderness is quadratic, with a peak in toughness at pHu 6.1 (Purchas & Aungsupakorn, 1993). The improved tenderness as ultimate pH increases above 6.1 appears to be largely attributable to improvements in water-holding capacity and consequent decreases in cooking losses, and to the greater activity of proteases at pH values close to neutrality (Yu & Lee, 1986). The reasons for beef becoming tougher with intermediate elevations in pH to around 6.1 appear to be due to reduced sarcomere length (Purchas et al., 1993).

3.6 Interactions between factors

As previously stated, the extent of myofibrillar shortening has a profound effect on initial tenderness. However, shortening can also influence aged meat tenderness as proteolysis will occur more slowly in meat with shortened sarcomeres for two reasons. Firstly, because rigor onset occurs at a later stage, ageing occurs more slowly due to the low initial temperatures in the muscle delaying activation of the calpains (Dransfield, 1994). The second reason is because of limited access of the proteases to the protein substrate within the sarcomeric structure, as shown by Weaver et al. (2008).
In muscles which have undergone some shortening due to high rigor temperatures, there is both rapid activation and inactivation of the proteases with a net reduction in tenderisation (Dransfield, 1994). Structure and function of muscle determine phenotype and therefore influence gene expression and metabolic profile. Interrelationships between structure and function make it difficult to establish cause and effect between any particular structural feature and a gross phenotype such as meat toughness (Harper, 1999).

4 Standardising pre- and post-slaughter conditions

Even under controlled pre- and post-slaughter management protocols there is still unexplained variation in traits like tenderness associated with the day, abattoir and year the animals were slaughtered, as shown in pigs (Purslow et al., 2008), cattle (Johnston, Reverter, Robinson & Ferguson, 2001) and sheep (Purchas, Sobrinho, Garrick & Lowe, 2002).

Robinson et al. (2001) showed that strict protocols should be used to control pre- and post-slaughter management in order to reduce the variation in tenderness measurements of the longissimus and semintendinosus muscles from 3,440 Bos indicus beef carcasses. Subsequent to the work by Robinson et al. (2001), Burrow et al. (2001) suggested that alternative methods of hanging of carcasses, such as tenderstretch, may reduce the amount of variation in shear force values observed. Wolcott, Johnston, Barwick, Iker, Thompson & Burrow (2009) compared tenderstretch and achilles hanging, within a beef carcasses, using 290 temperate and 290 tropical composite breeds to test the change in variation. The longissimus shear
force of tropical composite cattle was on average 7.4 Newtons tougher than the longissimus of temperate breeds with a similar reduction in shear force due to tenderstretch (8.2 N), for both breeds (Wolcott et al., 2009).

Despite efforts to standardise the effects of electrical stimulation and chilling rate on the temperature and pH fall post-slaughter, wide variation is often found between animals, abattoirs and also slaughter dates within abattoirs (Thompson, Hopkins, D' Souza, Walker, Baud & Pethick, 2005). This can be due to variation in electrode contact with the carcass, electrical resistance within the carcass and variation in wave form, current and frequency. But even with these standardised, the muscle metabolic response to electrical stimulation, as measured by muscle pH, can vary widely, depending on the metabolic state of the animal at slaughter. For example, Warner, Bond & Kerr (2000) reported that the muscle pH of lambs undergoing exercise pre-slaughter shows no response to post-slaughter electrical stimulation whereas the muscle pH of lambs un-exercised pre-slaughter drops by 0.5 pH units. Similarly in a mob of 70 cattle, the range in muscle pH drop in response to electrical stimulation was 0 (no change) to 0.8 pH units (drop from 6.6 to 5.8) (R. Warner, unpublished results). In addition, other electrical inputs such as at the immobiliser and hide puller, can contribute significant variation in muscle pH fall between beef carcasses.

Features of the animal’s pre-slaughter experience, including acute stress and physical activity, have been shown to influence markedly the toughness of beef (Warner, Ferguson, Cottrell & Knee, 2007a; Gruber, Tatum, Engle, Chapman, Belk & Smith, 2009) and lamb (Warner, Ferguson, McDonagh, Channon, Cottrell & Dunshea, 2005a) through mechanisms that are not well-understood. A farm animal which has
had a poor handling experience on the farm, may respond adversely to handling experiences between farm and slaughter, resulting in inferior meat quality (eg. in pigs; D'Souza, Warner, Dunshea & Leury, 1998). Warner, Dunshea, Ponnampalam & Cottrell (2005b) hypothesised that the release of nitric oxide during stress activates the calpain system and causes the increased toughness.

The origins for a day of slaughter, abattoir and animal effect on meat tenderness are not fully understood. Some factors that may contribute include variation; between animals in stress susceptibility and temperament, in handling and handlers at the abattoir, in stress during transport and loading/unloading, in inclement weather influencing the animals’ behaviour and variation in application of electrical stimulation as well as the carcass response to stimulation.

5 Specific gene or gene marker effects on beef and sheepmeat quality

Although many meat quality traits have generally been assumed to be under the control of multiple genes, there is considerable evidence that single genes account for a relatively large amount of variation for some traits (Burrow et al., 2001).

Gene markers for tenderness targeting expression of the calpain proteolytic system have been developed (Barendse, 2002; Barendse, Harrison, Bunch & Thomas, 2008; Page et al., 2002; White et al., 2005). The Carwell gene is another that has been characterised for its effects on tenderness and rib eye (loin) muscling in sheep (Nicoll
et al., 1998; Jopson et al., 2001). The *callipyge* gene in sheep (Cockett et al., 1993) and the *double muscling* gene in cattle (Arthur, 1995) have very large affects on carcass attributes but the specific causes of altered expression of carcass traits, chromosomal sites of mutations, and effects on meat tenderness differ substantially. This is discussed below.

5.1 Calpain system gene markers for tenderness in cattle

Gene markers have been used to identify cattle with better performance for commercial traits including meat tenderness (Barendse, 2002; Barendse et al., 2008; Page et al., 2002; White et al., 2005). Markers for tenderness have been identified and these markers are known to influence the expression of the calpain-calpastatin enzyme complex that regulates the rate of protein degradation in the live animal and post-mortem muscle (Koohmaraie, Kent, Shackelford, Veiseth & Wheeler, 2002).

The associations between these tenderness markers and meat quality traits were quantified within two concurrent experiments using 377 Brahman (*Bos indicus*) cattle at two different locations (Cafe, McIntyre, Robinson, Geesink, Barendse & Greenwood, 2010a; Cafe et al., 2010b). Cattle were selected for the study from industry and research herds at weaning based on their genotype for Calpastatin (*CAST, CAST:c.2832A>G*: Barendse, 2002) and Calpain 3 (*CAPN3, APN3:c.1538+225G>T*: Barendse et al., 2008) gene markers and the gene marker status for µ-calpain (*CAPN1-316, CAPN1:c.947C>G*: Page et al., 2002) was also identified. Each marker is for a single nucleotide polymorphisms (SNP) within genes...
controlling the calpain proteolytic system. The alleles differ in their effects on meat tenderness, with the favorable allele being associated with more tender meat.

In these studies, the gene markers had significant effects on shear force in the longissimus of Brahman cattle, but few effects on other meat quality traits (Cafe et al., 2010b, Table 1). Cattle with the favorable genotype for the three markers had reduced shear force compared to those with the unfavorable genotypes. The combined effects of the favorable marker alleles resulted in a 15.8 N reduction in shear force following 7 d aging (Table 1). Although the size of the effect of individual markers varied with experimental site, muscle, method of carcass suspension and aging period, there were no interactions (Cafe et al., 2010a,b). The results of Cafe et al. (2010a,b) are important as they provide evidence of few adverse effects due to the calpain system gene markers on production, carcass and beef quality related characteristics.

5.2 Callipyge sheep

The Callipyge sheep phenotype results from a mutation on chromosome 18 when present in heterozygous offspring that inherit the mutation from their sire (Cockett et al., 1994, 1996; Jackson, Green & Miller, 1997a; Freking et al., 1998a). They have extreme muscling, particularly in the hindquarters, reduced fatness and improved feed efficiency, but have extremely tough meat (Freking, Keele, Nielson & Leymaster, 1998b; Freking, Smith & Leymaster, 2004; Jackson et al., 1997a; Jackson, Miller and Green, 1997b,c; Koohmarai, Shackelford, Wheeler, Lonergan & Doumit, 1995). In Callipyge sheep, increased myofibre size is due to a greater proportion and size of type 2X/2B (fast glycolytic) muscle fibres and a reduction in the percentages of type
2A (fast oxidative-glycolytic) and 1 (slow oxidative) myofibres present (Koohmaraie et al., 1995; Carpenter, Owen, Cockett & Snowder, 1996; Lorenzen et al., 2000). The muscle of Callipyge sheep has increased calpastatin (Geesink & Koohmaraie, 1999; Duckett, Snowder & Cockett, 2000), reduced breaks in the I-band region during post mortem aging (Taylor & Koohmaraie, 1998) and no change in collagen cross-linking (Field, McCormick, Brown, Hinds & Snowder, 1996) compared to normal sheep, hence the myofibrillar rather than the connective tissue component is primarily responsible for the markedly increased toughness. Efforts to improve the tenderness and consumer acceptability of meat from Callipyge lambs have included electrical stimulation, prolonged aging, freezing prior to aging, injection of calcium chloride, the Hydrodyne® process, and different cooking methods, with variable degrees of success (Carpenter & Solomon, 1995; Solomon, Long, Eastridge & Carpenter, 1995; Clare, Jackson, Miller, Elliot & Ramsey, 1997; Shackelford, Wheeler & Koohmaraie, 1997, 1998; Duckett, Klein, Dodson & Snowder, 1998).

5.3 Carwell or Rib Eye Muscling (REM) gene in sheep

It has emerged that certain Australian Poll Dorset sires produce offspring with increased longissimus cross-sectional area and mass at equivalent live or carcass weights (Barendse, 1995; Banks, 1997; Nicoll et al., 1998; Greenwood, Davis, Gaunt & Ferrier, 2006a). This is apparently due to segregation at a locus on chromosome 18 known as the rib-eye muscling (REM) or Carwell locus (Nicoll et al., 1998; Jopson et al., 2001). This locus maps in close proximity to the Callipyge locus (see Section on Callipyge), however no published data is available concerning the precise chromosomal position of the sequence variant responsible. Association analysis has
been performed in half-sib progeny derived from Poll Dorset and White Faced Suffolk sires which demonstrated heterozygosity for DNA markers linked to the Carwell mutation (Kijas et al., data unpublished). Significant effects were detected for increased loin weight, as well as for muscling and leanness (CT scan measurements), however the magnitude of the effects were generally small (Kijas, personal communication). The Carwell phenotype may have inferior eating quality due to increased toughness (Cummins, 1997; Davis, Ferrier & Gaunt, 1999; Jopson et al., 2001; Hopkins & Fogarty, 1998), although this effect appears to be small and variable compared to that in Callipyge lambs.

Clearly, the development and application of markers for this and other alleles associated with increased muscling within the Australian sheep population will need to ensure that the increase in sheep meat yield does not come at the expense of sheepmeat quality characteristics including tenderness.

5.4 Mutations resulting in non-functional myostatin in cattle and sheep

The most extreme effects of specific genes on muscle characteristics are those associated with double muscled cattle and the Callipyge sheep phenotypes. Although mutations for these genes result in increased musculature, their chromosomal sites, specific causes and effects differ substantially, as summarised in Table 2.

The high degree of muscling that is evident in some European breeds of cattle (eg. Belgian Blue and Piedmontese) is in some cases associated with mutations in the gene regulating myostatin (Bellinge, Liberles, Iaschi, O’Brien & Kay, 2004). Mutations in
the myostatin gene (MSTN, also known as growth differentiation factor 8, GDF8) have been documented to alter muscling and muscle structure in cattle (Lines, Pitchford, Kruk & Bottema, 2009). If the gene is inactivated through the mutations, the protein is non-functional, causing the ‘double muscled’ phenomenon (Joulia-Ekaza & Cabello, 2006). The most common mutation is caused by an 11 base pair deletion (del 11) at base 821, evident within the Belgian Blue breed (Switonski, 2002). Animals can be homozygous positive for the mutation, and exhibit extreme muscling, or heterozygote for the mutation, expressing muscling intermediate between the homozygous positive and the wild-type. In Belgian Blue, the mutation has been associated with significant reductions in the shear force and a decrease in total collagen content (Ngapo, Berge, Culioli, Dransfield, De Smet & Claeys, 2002).

Within the Limousin cattle population, a gene variant of myostatin, F94L, which increases muscle mass also results in meat which is more tender, through a reduction in the collagen/elastin content of muscle (Lines et al., 2009). The mechanism for this gene variant is not known.

A study investigating the benefits of incorporating single MSTN loss-of-function alleles into breeding programmes to improve muscularity was conducted in two Australian beef cattle herds of Angus origin (O’Rourke et al., 2009). Both the research and commercial herds had Low and High muscling lines which were homozygous for ‘normal’ (also called wild-type) myostatin and a High muscling line which was heterozygous for the 821 del11 myostatin allele that results in non-functional myostatin. There were no differences due to genotype for shear force or compression in *longissimus* samples from both herds (O’Rourke et al., 2009) (Table 3). A reduction in shear force for the *m. semitendinosus* samples was evident due to
genotype in the commercial herd, but not in the research herd. Tatum, Gronewald, Seideman & Lamm (1990) and Wheeler, Shackelford, Casas, Cundiff and Koohmaraie (2001) found increased tenderness in Piedmontese which were heterozygous for the 821 del11 myostatin allele and similar findings were found in Charolais heterozygotes (Levéziel et al., 2006; Casas et al., 1998). However, Gill, Bishop, McCorquodale, Williams and Wiener (2009) did not find significant effects of heterozygosity on tenderness.

A mutation (g+6723G>A) in the myostatin gene (GDF8) that causes translational inhibition has been shown to result in increased muscling in Texel and other sheep (Laville et al., 2004; Clop et al., 2006; Kijas, McColloch, Hocking Edwards, Oddy, Lee & van de Werf, 2007), and other mutations in the sheep myostatin gene have also been identified that influence muscling and fatness (Kijas et al., 2007). The g+6723G>A mutation increased the percentage of fast glycolytic myofibres (Laville et al., 2004), but did not affect shear force (Kijas et al., 2007). However, it did reduce intramuscular fat and reduce sensory scores for eating quality including tenderness (Kijas et al., 2007). In another study, objective meat quality traits including longissimus and semimembranosus shear force were not affected by a QTL encompassing GDF8, which influences carcass composition in Texel sheep (Johnson, McEwan, Dodds, Purchas & Blair, 2005).

In general, mutations in the myostatin gene in cattle often, but not always, result in improved tenderness which has been attributed to a decrease in the amount of collagen in muscle relative to myofibres (Boccard, 1982; Uytterhaegen et al. 1994) and in the amount of stable non-reducible cross links (Bailey, Enser, Dransfield,
Restall, and Avery, 1982). Mutations in the myostatin gene in sheep appear to have no influence on shear force, although there may be a reduction in consumer perception of tenderness, perhaps due to intramuscular fat-related effects.

6 Partitioning of genetics and environmental effects on beef quality

6.1 Heritability estimates and genetic correlations for beef quality traits

In their extensive reviews of genetic parameters for beef quality traits, Marshall (1999) and Burrow et al. (2001) reported that objective tenderness measures (shear force) and intramuscular fat percentage were moderately heritable \( h^2 = 0.2 - 0.3 \) whilst objective (reflectance; \( L_*, a* \) and \( b* \) values) and subjective assessments of meat colour and water holding traits (drip loss, cooking loss percentage) were less heritable \( h^2 = 0.1 - 0.25 \). For consumer panel scores of tenderness, juiciness and flavour, the average heritability was also low ranging from 0.05 – 0.2 with panel tenderness score being the highest at 0.22 (Marshall, 1999; Burrow et al., 2001). From this we can conclude that of the total phenotypic variance in beef quality traits, genetics accounts for 5 – 30%, depending on the trait. Secondly, given the low to moderate heritability for the traits, the opportunity for genetic improvement is somewhat limited but would appear best for traits such as tenderness and intramuscular fat percentage. With respect to tenderness, this opportunity is greater however for Bos indicus or tropically adapted breeds given the higher heritability (longissimus shear force \( h^2 = 0.30 \); consumer panel tenderness score \( h^2 = 0.31 \)) and phenotypic variance compared to Bos taurus
breeds (shear force $h^2 = 0.09$; consumer panel tenderness score $h^2 = 0.1$) (Johnston, Reverter, Ferguson, Thompson & Burrow, 2003).

There is a genetic correlation between IMF and tenderness in beef carcasses. For temperate breeds of cattle, the genetic correlation between IMF and consumer panel tenderness or shear force is 0.61 and -0.38 respectively (Reverter et al., 2003a). As the genetic correlation between consumer tenderness and consumer flavour or juiciness is high (0.93 and 1.0 respectively) (Johnston et al., 2003a), this suggests the correlation between IMF and consumer panel tenderness is most likely driven by palatability traits other than tenderness. IMF can be measured on the live animal thus selection for IMF \textit{in vivo} will also result in some genetic improvement in shear force. Preliminary data from the meat of lambs in the sheep Co-operative Research Centre (CRC) in Australia suggest that the genetic correlation between shear force and IMF is also high (Sue Mortimer, \textit{pers. comm.}).

Whilst the phenotypic relationship between temperament and tenderness is low (Burrow, Seifert & Corbet, 1988; Colditz, Ferguson, Greenwood, Doogan, Petherick & Kilgour, 2007; Kadel, Johnston, Burrow, Graser & Ferguson, 2006; King et al., 2006), the same cannot be said for the genetic correlation ($rg = 0.3–0.4$) between these two traits in cattle (Kadel et al., 2006; Reverter, Johnston, Perry, Goddard & Burrow, 2003b). The mechanism is not clear but certainly this data together with data from Warner et al. (2007a), Gruber et al. (2009) and Voisinet, Grandin, O’Connor, Tatum and Deesing (1997) suggests that stress has a significant and negative effect on meat tenderness, when measured by shear force or consume panel. The most likely causative mechanism is through effects on the proteolytic system, as proposed by
Warner et al. (2005b), but it is also possible that another unknown mechanism is involved.

6.2 Production environment

Cattle are grown and finished in a range of different production environments. Under extensive pasture-based environments, diet composition and feed availability can vary considerably resulting in variable rates and patterns of growth and changes in energy metabolism and body composition which in turn, can directly and indirectly affect beef quality. The impact of nutrition on muscle properties and product quality has been extensively reviewed by Oddy et al. (2001) and Hocquette et al. (2007). The focus of the following section is to examine the interaction between animal genetics and the production environment.

In a large beef cattle genetic evaluation study, progeny (n = 7,781) from the same sires from either temperate or tropically adapted breeds were grown to three slaughter weights (220, 280 and 340 kg carcass weight) in both pasture and feedlot finishing systems. The design and experimental protocols have been described in detail by Upton, Burrow, Dundon, Robinson & Farrell (2001) and Perry, Shorthose, Ferguson & Thompson (2001). Pasture finished cattle had significantly darker (lower L* values) and tougher (higher shear force values and lower consumer tenderness scores) meat and had less intramuscular fat than those finished in the feedlot (Reverter et al., 2003a; Johnston et al., 2003). The effect of slaughter weight and hence slaughter age on tenderness and meat colour measurements was less consistent and surprisingly smaller in magnitude, compared to the effect of finishing system. Cattle finished at
heavier slaughter weights had higher intramuscular fat percentage and visual marbling scores (Reverter et al., 2003a).

The genetic correlations between traits at different slaughter weights or between pasture and feedlot finished cattle were generally high (>0.7) and close to unity for most meat quality measures including longissimus shear force and intramuscular fat %. These results do not lend support for G x E interactions as the ranking between sires was not influenced by the weight at slaughter or whether progeny were finished on pasture or in the feedlot.

Using a subset of this data, Johnston et al. (2001) published an earlier study examining the sources of variation in longissimus shear force. After the additive variance (genetic component), slaughter group was the second largest source of explained variance in shear force accounting for 17.8% of the total phenotypic variance (Table 4). Slaughter group was defined as all animals run together from intake (year, season and sex), finishing system, slaughter weight and slaughter date. Importantly, the partitioning of the slaughter group variance revealed that slaughter weight and finishing system only accounted for the 1.4% of the total phenotypic variance. This reinforces the point that when the post-slaughter conditions are controlled, the contributions of these production factors to the variation in tenderness are relatively low. Finally, slaughter date which also encapsulates differences between abattoirs accounted for the majority (15.3%) of the slaughter group effect. This highlights that there are other unidentified animal and/or environmental factors that contribute to the variation in tenderness. Efforts to better understand, characterise and control these may yield further improvements in this highly important consumer trait.
Effect of growth path on beef tenderness - The extent to which nutrition and growth path affect beef tenderness and other production traits has been investigated in various large-scale, long-term studies within the Beef CRC in Australia. In general, the closer the nutritional perturbation to slaughter, the greater the potential to impact on beef quality and in particular beef tenderness (Table 5). From this, it was concluded that finishing systems can have significant effects on tenderness, whereas growth and nutrition earlier in life have minimal or no impact. It is also notable that genotype × nutritional (environmental) interactions were not evident within the Beef CRC studies (Table 5).

Rapid growth of cattle at pasture over the entire post-weaning period to the same market weight or to feedlot entry may or may not improve beef tenderness (Perry & Thompson, 2005; McKiernan & Wilkins, 2007; Table 5), and any improvements are not consistent across breed types, geographic locations, and muscles (Perry et al., 2005). In this regard, while beef eating quality characteristics decline with age (Perry et al., 2005), serious adverse affects of backgrounding growth (also called ‘grow-out’ period, and corresponds to the post-weaning, pre-finishing period) on beef tenderness are only likely to be evident if retarded growth results in animals being substantially older when they reach the same market weight, by about 9 months, for example Purchas, Burnham & Morris (2002).

More rapid growth of cattle during finishing or use of feedlot compared to pasture finishing (Table 5) generally improves meat tenderness (Johnston et al., 2003; Perry et al., 2005), although not always depending upon factors such as prior nutrition and the
duration of concentrate feeding (Troy, Murray, O’Sullivan, Mooney, Moloney & Kerry, 2002).

6.3 Post-slaughter environment

A key point to highlight here is that unlike some beef quality traits such as intramuscular fat percentage, the tenderness phenotype is not fixed at slaughter and can be significantly affected by the post-slaughter processing conditions that prevail during the first 24 h after slaughter and duration of ageing. Failure to control the post-slaughter processing conditions may allow cold shortening or heat-toughening to occur, particularly under rapid chilling regimens for the former. This of course can be avoided through the application of effective electrical stimulation which accelerates the rate of post-mortem glycolysis. As discussed above, cold shortening results in profound changes to the phenotypic variance in tenderness. However it can also influence the estimate of heritability for shear force as demonstrated by Johnston et al. (2001). They showed that the exclusion of only three slaughter groups (n = 173) where electrical stimulation was not applied, from the full dataset (n = 2661), resulted in a 33% reduction in the phenotypic variance and an increase in the longissimus shear force heritability from 0.19 to 0.31.

Another post-slaughter strategy to minimise cold shortening is tenderstretch. Wolcott et al. (2009) conducted a study on 2180 Brahman and tropical composite feedlot steers where one side was tenderstretched and the other hung normally via the Achilles tendon. They showed that tenderstretch reduced the average shear force by 8.2 Newtons, for both breeds. The genetic and phenotypic variance for both breeds
was substantially reduced by tenderstretch, but importantly, the heritability was essentially unchanged ($h^2 = 0.32-0.33$ for Achilles hung, $h^2 = 0.30$ for tenderstretch (standard errors were the same for Achilles hung and tenderstretch) (Wolcott et al., 2009). Moreover, the genetic correlation between shear force measures in the tenderstretched and Achilles hung sides was quite high ($r_g = 0.77 \pm 0.11$).

Collectively, these results indicate that the genetic or sire variance in shear force may not be underpinned by differences in the mechanisms regulating post-mortem myofibrillar contraction. Wolcott et al. (2009) also reported that the genetic correlations between the difference in shear force ($\Delta SF$) due to hanging method ($\Delta SF = \text{Achilles hung SF} - \text{tenderstretch SF}$) and Achilles hung SF and tenderstretch SF were $0.92 \pm 0.03$ and $0.49 \pm 0.19$, respectively. This difference in the magnitude of the $r_g$ is quite salient as it is suggestive of a small G x E interaction.

As discussed above, the rate and extent of post-mortem proteolysis also governs the expression of tenderness. In two separate studies, the estimated heritability for beef longissimus shear force varied with the duration of ageing (Figure 1) (O'Connor, Tatum, Wulf, Green & Smith, 1997; Wulf, Tatum, Green, Morgan, Golden & Smith, 1996). Contrasting results between the studies were reported with respect to the interaction between breed or sire within breed and ageing duration. Wulf et al. (1996) found minimal re-ranking of sires for tenderness throughout the 1 – 35 days of ageing. In contrast, O’Connor et al. (1997) found a significant breed x ageing interaction where a more rapid rate of ageing (ie. tenderisation) was evident in Bos taurus compared with Bos indicus composites. The reduced proteolytic potential and therefore tougher meat in Bos indicus relative to Bos taurus cattle is reasonably well documented (e.g Shackelford Koohmaraie, Miller, Crouse & Reagan, 1991; Wheeler,
Savell, Cross, Lunt & Smith, 1990) but the breed difference in tenderness can be partially reduced through electrical stimulation (Ferguson, Jiang, Hearnshaw, Rymill & Thompson, 2000).

In a study of 273 Limousin x Jersey steers and heifers where the post-mortem conditions were very tightly controlled and rigor development and subsequent ageing occurred at a constant temperature of 15°C, Daly (2000) reported a 17 kg (New Zealand tenderometer measurements) difference in longissimus shear force between animals at rigor and a 10 fold difference between animals in the rate of post-mortem tenderisation. Notwithstanding the contributions to meat tenderness from other structural variants (eg. myofibrillar shortening, connective tissue), the mechanisms that underpin the variation in post-mortem proteolytic rate are probably the primary contributors to the genetic variation in tenderness.

### 7 Partitioning of genetics and environmental effects on sheepmeat quality

This subject has received much less research attention in sheep relative to cattle although a large scale progeny evaluation project has recently commenced in Australia.

#### 7.1 Genetic effects
Most published information on genetic variation in sheep meat quality is from inter-breed comparisons (Bishop et al., 2009) and there is a paucity of within breed genetic estimates for sheep meat quality traits. For example, in the review by Safari et al. (2005) of genetic parameter estimates for animal and quality traits in sheep, they only found published estimates for the heritability of meat pH and colour (0.18 and 0.09, respectively). However, new evidence is emerging as the heritabilities for tenderness traits have been investigated in two recent studies. Karamichou, Richardson, Nute, Wood and Bishop (2007) reported the heritability estimates of 0.15 for trained sensory panel score and 0.39 for shear force in Scottish Blackface sheep (n=350). Preliminary results of Mortimer et al. (2009) indicate a moderate heritability for shear force for 2,200 lambs with linked sires in seven flocks across Australia. More recent results (Warner, unpublished results) indicate that the heritability of shear force after 5 days ageing is 0.39, across 7 flocks, 90 sires and 2,500 progeny. This large study is ongoing and it aims to generate phenotype data on at least 10,000 lamb progeny across 7 flocks from four regions in Australia. This study will provide definitive answers for the effect of genetic x environment interactions on meat quality traits across the range of production systems for sheep in Australia.

Numerous studies have shown between breed, including sire differences, in lamb meat tenderness (Young, Reid & Scales, 1993; Fisher et al., 2000; Purchas et al., 2002; Hopkins et al., 1998; Martínez-Cerezo, Sanudo, Panea & Olleta, 2005; Martínez-Moreno, Sanudo, Medel & Olleta, 2005). Comparing six lamb crossbreds including Texels, Border Leicester, Poll Dorset and Merino crossed with Merinos, and second crosses, Hopkins et al. (1998) found no differences in shear force between crossbreds for either the longissimus or semimembranosus. In the same study, where the progeny
of the three Poll Dorset sires were evaluated, a significant sire difference of 11.7 Newtons was reported (Hopkins et al., 1998). The sire producing ‘tough’ meat in its progeny appear to be a line that is genetically tough and is called a ‘Carwell’ sire (see section above).

Selection for increased muscling in sheep can produce economic benefits through increased carcass yield but are there any negative effects on meat quality? Consumers have scored meat from lambs sired from high muscling rams as tougher, by ten units (scale of 0-100) (derived from Hopkins, Stanley, Toohey, Gardner, Pethick & Van De Ven, 2007b), relative to meat from lambs sired from low muscling rams, although no difference was found in shear force (Hopkins et al., 2007b). In other studies, the semimembranosus from lambs sired by high muscling rams had higher shear force values then that from lambs sired by low muscling rams (66.5 vs 61.4 N, respectively) (Hopkins, Stanley, Martin, Toohey & Gilmour, 2007a). The meat from progeny of sires which are extreme in either muscling or fatness have been found to produce meat which is unacceptable to the consumer via changes in tenderness (proteolysis, connective tissue) or juiciness/flavour (IMF) and such sires should be avoided (Warner et al., 2007b).

The role of intramuscular fat in the tenderness of meat from lamb carcasses has received less research attention then beef. Variation in sire breeding values for muscling and fatness affected consumer scores for tenderness of the longissimus lumborum with scores declining as muscling increased and as fat decreased, which appears to be driven by IMF (Thomson, 2009). Hopkins et al. (2007b) reported that the meat from lambs sired by rams with a high breeding value for fatness had higher
sensory tenderness scores than that from rams with low breeding values for fatness.

Hopkins, Hegarty, Walker and Pethick (2006) used consumer panels to show that a minimum of 5% IMF is required in order to achieve a failure rate of less than 10% for tenderness.

The heritability of ultimate pH in lambs ranges from 0.18 to 0.27 (Safari et al. 2005; Bishop et al., 2009; Fogarty, Safari, Taylor and Murray, 2003). There is evidence for breed effects on the way muscle glycogen metabolism is regulated in lambs. For example, there is clear evidence that Merino lambs are more susceptible to the high pH or dark cutting syndrome (Warner et al. 2006). This appears to be due to greater rates of glycogen loss between leaving the farm gate and slaughter (Gardner et al., 1998) suggesting Merino are more sensitive to stress than lambs sired by meat breeds. Further work has shown that selection for increased muscularity can influence the metabolism of glycogen in skeletal muscle. Thus lambs derived from sires with high genetic merit for muscularity have higher muscle glycogen concentration at any given level of nutrient intake (Martin, Gardner, Thompson and Hopkins, 2006). This strongly suggests that lambs with higher muscularity will be less susceptible to the high pH or dark cutting syndrome, which can impact on meat tenderness (Purchas et al., 1993).

7.2 Production Environment

There is a paucity of research into sheep meat breeds that attempts to understand the interactions of specific genetic selection indices on lamb performance, carcass composition, and meat quality (Pethick, Warner & Banks, 2006). In Australia, sheep
can be slaughtered for meat at ages ranging from 4 months to 3-6 years and are grown and finished under a range of environments and feeding systems, from green pasture in spring to hay or grain feeding in autumn and winter. The major breeds that contribute to the sheep meat industry are the Merino, Border Leicester and Poll Dorset. Research to understand the influence of carcass breeding values, age, growth path and nutritional supply on carcass and eating quality has been undertaken (Pethick, Warner & Banks, 2007). This research, as well as research from around the world, is reported below.

The age of the animal appears to be the largest environmental effect in sheep meat tenderness (Warner et al., 2007b). Meat from lambs is given a higher tenderness score by consumers, then meat from older sheep, for both the loin and leg muscles (Pethick, Hopkins, D'Souza, Thompson & Walker, 2005a). Surprisingly, the difference in consumer score between an 8-month old lamb and a 6 year old sheep is only 10 consumer points, for both the longissimus thoracis and the biceps femoris (Pethick et al., 2005a). This is also reflected in an increase in shear force with age of the sheep (Hopkins et al., 2007a) and is attributed to a reduction in collagen solubility (Young et al., 1993).

Nutrition in the six weeks pre-slaughter (ME from 6.2 to 11.0 MJ/kg) and the plane of the nutrition from birth to weaning, appear to have no effect on consumer scores for tenderness (Pethick et al., 2005b; Hopkins et al., 2005b, respectively). Faster growth in lambs has been shown to result in thinner connective tissue, but there were no effects on objective or consumer measures of meat tenderness (Allingham, Barris,
Reverter, Hilsenstein, Van De Ven & Hopkins, 2009) and there was no interaction with genotype or sire.

There is little data on the effect of genetic-environmental interactions on lamb meat quality or tenderness. In a study by Hopkins et al. (2005) utilising 140 crossbred lambs, there was no interaction between growth rate from birth to slaughter and sire Estimated Breeding Value (EBV) for muscling for either eating quality or shear force. Subsequently, Hopkins et al. (2007b) found in 627 cross-bred lambs that for lambs which were weaned early and fed a ‘restricted’ diet, the lambs sired by fast growth sires had tougher longissimus, although this was not the case in early-weaned full-fed or late-weaned lambs. (Hopkins et al., 2007b) found subtle interactions between selection for growth or muscling or fatness traits and growth path and suggested that the impact of a period of restriction and refeeding on meat and eating quality will be mediated by the sire genetics.

Arsenos et al. (2002) showed an interaction between breed and slaughter weight for tenderness of the leg muscles of lamb carcasses. For the Boutsko breed, lambs slaughtered at 48% of their mature weight were significantly tougher than lambs slaughtered at 55% mature weight, whereas for the Serres and Karagouniko breeds, the tenderness of the meat was similar between slaughter groups.

Purchas et al. (2002) showed a genotype x year interaction with Romney sires producing significantly lower shear force values in the semimembranosus than Finn x Poll Dorset sires (8.40 vs 10.21 kg, respectively) in one year but not in the preceding two years.
7.3 Post-slaughter environment

Effective electrical simulation used on lamb and sheep carcases results in meat which is generally more acceptable to the consumer in terms of tenderness (Shaw, Baud, Richards, Pethick, Walker & Thompson, 2005) and as discussed above tenderstretch can be used to overcome the toughening effects of both cold-shortening and heat-toughening (Thompson et al., 2005).

Martinez-Moreno et al. (2005) showed an interaction between genotype, live weight and ageing time post-slaughter for consumer tenderness scores (Figure 2). At the start of the ageing period, there were no differences in trained taste panel scores for tenderness between meat and dairy breeds or between lambs slaughtered at 11 or 31 kg liveweight. With ageing for 4 or 8 days, marked tenderisation occurred in the meat breed slaughtered at 31 kg but not for meat lambs slaughtered at 11 kg. For the dairy breed, tenderisation occurred at a similar rate for both live weights.

Thompson, Gardiner, Thomson and Allingham (2007) showed that although the progeny of sires with high breeding values for muscling or growth had tougher meat, there was no interaction with hanging method (ie. Achilles v. tenderstretch) or electrical stimulation.
8 Summary and conclusions

Tenderness is an important quality trait to the consumer. It is vital to control the pre- and post-slaughter environment when attempting to determine the contributions of genetic and production factors to meat tenderness. Genetic improvement in meat tenderness is possible, via traditional quantitative and more recently by genomic approaches (e.g., calpain markers). The greatest gains in tenderness in beef cattle appear to be in breeds like *Bos Indicus* where the phenotypic variation is larger. Variation in proteolysis appears to be the dominant contributor to genetic variation in tenderness, certainly in cattle. Increasing muscle mass is achievable by targeted selection for specific mutations in the myostatin gene, but if the increase comes at the expense of tenderness, then we question if real gains have been made.

The progressive improvement in meat tenderness through selection of cattle and sheep known to have ‘tender’ genetics, will be agonising slow unless strict attention is paid to processing procedures. Interactions between genotype and environment contribute to the phenotypic variation in meat tenderness. There are certainly other factors that contribute to the variation in tenderness, including variation between slaughter dates and abattoirs. This highlights that there are unidentified animal and/or environmental factors that contribute to the variation in tenderness. Efforts to better understand, characterise and control these may yield further improvements in this highly important consumer trait.

Acknowledgements
Comments on the manuscript from James Kijas (CSIRO) are gratefully acknowledged, as well as biometrical advice from Kym Butler (DPI, Werribee). The assistance of Owen Young (New Zealand) and Roger Purchas (New Zealand) in finding references is also acknowledged and the helpful comments from the anonymous reviewers.
References


Barendse, W. J. (2002). *DNA markers for meat tenderness*. International patent publication W0 02/064820.


Cafe, L.M., McIntyre, B. M., Robinson, D. L., Geesink, G. H., Barendse, W.,
processing studies on calpain-system gene markers for tenderness in cattle: 2.
Objective meat quality. *Journal of Animal Science (submitted-revised
manuscript sent back Mar 2010).*

and composition of muscles from normal and Callipyge lambs. *Journal of
Animal Science*, 74, 388-393.

tenderise callipyge lamb. In *Proceedings of the International Congress of Meat
Science and Technology*, 41, 620-621.

Casas, E., Keele, J. W., Shackelford, S. D., Koohmaraie, M., Sonstegard, T. S., Smith,
hypertrophy locus with carcass traits in beef cattle. *Journal of Animal Science*,
76, 468-473.

slaughter handling and stunning method all influence pork quality. *Meat
Science*, 56, 291-299.

Improving tenderness of normal and callipyge lambs with calcium chloride.

Clop, A., Marcq, F., Takeda, H., Pirrottin, D., Toroir, X., Bibe, B., Bouix, J., Caiment,
F., Elsen, J-M., Eychenne, F., Larzul, C., Laville, E., Meisch, F., Milenkovic,


ovine *Callipyge* locus: I. Relative chromosomal position and gene action.

*Journal of Animal Science*, 76, 2062-2071.


Table 1
Shear force (N) for Achilles (AT) and tenderstretch (TS) suspended *longissimus* and Achilles suspended *semitendinosus* aged for 1 or 7 d for Brahman cattle with a combination of favourable and unfavourable genotypes for tenderness across 3 markers in New South Wales (NSW) and Western Australian (WA) experiments (Cafe et al. 2010b).

<table>
<thead>
<tr>
<th></th>
<th>AT <em>longissimus</em></th>
<th>TS <em>longissimus</em></th>
<th>AT <em>semitendinosus</em></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1 d</td>
<td>7 d</td>
<td>1 d</td>
</tr>
<tr>
<td>NSW herd</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0_0_0a</td>
<td>82.8</td>
<td>80.4</td>
<td>48.9</td>
</tr>
<tr>
<td>2_2_1</td>
<td>77.5</td>
<td>64.6</td>
<td>44.7</td>
</tr>
<tr>
<td>Difference</td>
<td>5.3</td>
<td>15.8</td>
<td>4.3</td>
</tr>
<tr>
<td>SED</td>
<td>4.53</td>
<td>4.04</td>
<td>1.29</td>
</tr>
<tr>
<td>P-value</td>
<td>0.243</td>
<td>&lt;0.001</td>
<td>0.001</td>
</tr>
<tr>
<td>WA herd</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0_0_0</td>
<td>50.4</td>
<td>55.2</td>
<td>53.6</td>
</tr>
<tr>
<td>2_2_1</td>
<td>49.2</td>
<td>42.2</td>
<td>46.5</td>
</tr>
<tr>
<td>Difference</td>
<td>1.2</td>
<td>13.0</td>
<td>7.1</td>
</tr>
<tr>
<td>SED</td>
<td>4.21</td>
<td>3.47</td>
<td>4.34</td>
</tr>
<tr>
<td>P-value</td>
<td>0.784</td>
<td>&lt;0.001</td>
<td>0.108</td>
</tr>
</tbody>
</table>

a: Marker combination of favorable alleles for *CAST_CAPN3_CAPN1-4751*. Note: 2_2_1 is a favourable combination and should result in more tender meat.
Table 2.
Comparison of characteristics of double-muscled cattle and *Callipyge* sheep with normal cattle and sheep, respectively (derived from Greenwood and Dunshea, 2009)

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Double-muscled cattle</th>
<th><em>Callipyge</em> sheep</th>
</tr>
</thead>
<tbody>
<tr>
<td>Specific cause</td>
<td>Myostatin (GDF8) mutation (non-functional myostatin)</td>
<td>Uncertain (DLK-1 involved)</td>
</tr>
<tr>
<td>Location of single nucleotide polymorphisms(s)</td>
<td><em>Bovine</em> chromosome 2 (Ovine chromosome 2)¹</td>
<td><em>Ovine</em> Chromosome 18</td>
</tr>
<tr>
<td>Genotype resulting in mutant phenotype</td>
<td>Homozygous for mutant allele (heterozygote has intermediate phenotype)</td>
<td>Heterozygote with mutant allele inherited from sire (polar overdominance)</td>
</tr>
<tr>
<td>Phenotypic expression</td>
<td>Prenatal and postnatal</td>
<td>Primarily postnatal</td>
</tr>
<tr>
<td>Location of muscle hypertrophy</td>
<td>More generalised</td>
<td>Hindquarter and loin</td>
</tr>
<tr>
<td>Myofibres of affected muscles</td>
<td>Hyperplasia</td>
<td>No hyperplasia</td>
</tr>
<tr>
<td></td>
<td>More type 2X</td>
<td>More type 2X</td>
</tr>
<tr>
<td></td>
<td>Less type 2A</td>
<td>Far less type 2A</td>
</tr>
<tr>
<td></td>
<td>May have type 2 hypertrophy depending on mutation and genetic background of cattle</td>
<td>Type 2 hypertrophy</td>
</tr>
<tr>
<td></td>
<td>More glycolytic</td>
<td>More glycolytic</td>
</tr>
<tr>
<td>Predominant mechanism in enhanced muscle growth</td>
<td>Increased protein synthesis</td>
<td>Reduced protein degradation (more calpastatin)</td>
</tr>
<tr>
<td>Meat quality</td>
<td>Similar or more tender, pale</td>
<td>Much tougher, pale</td>
</tr>
</tbody>
</table>

¹ Mutation in sheep (especially Texel breed) which affects translation into myostatin protein.
Table 3
Liveweight, carcass, yield and beef quality characteristics of steers in two beef cattle herds (A and B) containing animals varying in muscling selection line (Herd A only) and either homozygous normal for the myostatin gene (wt/wt) or heterozygous for the 821 del11 loss of function myostatin mutation (mh/wt) (derived from O’Rourke et al. 2009).

<table>
<thead>
<tr>
<th>Muscling Selection Line²</th>
<th>Herd A¹</th>
<th>Herd B¹</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Low</td>
<td>High</td>
</tr>
<tr>
<td>Myostatin gene³</td>
<td>wt/wt</td>
<td>wt/wt</td>
</tr>
<tr>
<td>n</td>
<td>19</td>
<td>14</td>
</tr>
</tbody>
</table>

Carcass and yield characteristics

<table>
<thead>
<tr>
<th></th>
<th>Herd A¹</th>
<th>Herd B¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carcass weight (kg)</td>
<td>366</td>
<td>360</td>
</tr>
<tr>
<td>Rib fat depth (mm)</td>
<td>18.3</td>
<td>15.7</td>
</tr>
<tr>
<td>EMA (cm²)</td>
<td>70.4c</td>
<td>77.0d</td>
</tr>
<tr>
<td>Retail yield (% CCW)</td>
<td>62.2c</td>
<td>63.5c</td>
</tr>
<tr>
<td>Fat trim (% CCW)</td>
<td>18.5d</td>
<td>17.0d</td>
</tr>
<tr>
<td>Bone (% CCW)</td>
<td>18.7d</td>
<td>18.9d</td>
</tr>
</tbody>
</table>

Longissimus quality

<table>
<thead>
<tr>
<th></th>
<th>Herd A¹</th>
<th>Herd B¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shear force (kg)</td>
<td>4.6</td>
<td>5.1</td>
</tr>
<tr>
<td>Compression (kg)</td>
<td>1.6</td>
<td>1.8</td>
</tr>
<tr>
<td>IMF (%)</td>
<td>4.0</td>
<td>4.1</td>
</tr>
</tbody>
</table>

Semitendinosus quality

<table>
<thead>
<tr>
<th></th>
<th>Herd A¹</th>
<th>Herd B¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shear force (kg)</td>
<td>5.2</td>
<td>4.7</td>
</tr>
<tr>
<td>Compression (kg)</td>
<td>2.5</td>
<td>2.3</td>
</tr>
</tbody>
</table>

¹Herd A established from Angus x Hereford cows, Herd B established from Angus and Charolais cows

²Muscling selection lines in Herd A steers; Low = low muscle selection line, High = high muscling selection line. Selection was based on muscle score as assessed at weaning.

³Myostatin gene; homozygous ‘normal’ (wild-type=wt) myostatin = wt/wt, heterozygous for the 821 del11 loss of function myostatin mutation = mh/wt

Within herds, mean values followed by different letters are significantly different at $P = 0.05$
Table 4
Contributions to the total and slaughter group variance in *longissimus* shear force (from Johnston *et al.* 2001).

<table>
<thead>
<tr>
<th>Overall variance components</th>
<th>Variance (kg²)</th>
<th>Slaughter group variance components</th>
<th>Variance (kg²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Additive genetic</td>
<td>0.216</td>
<td>Slaughter weight and finishing system</td>
<td>0.010</td>
</tr>
<tr>
<td>Slaughter group</td>
<td>0.122</td>
<td>Electrical stimulation system*</td>
<td>0.008</td>
</tr>
<tr>
<td>Herd and sex</td>
<td>0.036</td>
<td>Slaughter date</td>
<td>0.109</td>
</tr>
<tr>
<td>Residual</td>
<td>0.330</td>
<td>Total</td>
<td>0.127</td>
</tr>
</tbody>
</table>

*Low voltage versus high voltage electrical stimulation*
Table 5
Effects of nutrition and growth during different stages of development for beef peak force (N) at heavier market weights of cattle, from Australian Beef Cooperative Research Centre studies demonstrating the relatively large effects due to nutrition during finishing compared to earlier in life.

<table>
<thead>
<tr>
<th>Stage of growth and muscle</th>
<th>Breed type</th>
<th>Comparison</th>
<th>P-value</th>
<th>References^a</th>
</tr>
</thead>
<tbody>
<tr>
<td>Birth weight (Prenatal growth)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Longissimus</td>
<td>Temperate</td>
<td>Low</td>
<td>39.2</td>
<td>40.5</td>
</tr>
<tr>
<td>Semitendinosus</td>
<td>Temperate</td>
<td>High</td>
<td>46.2</td>
<td>46.4</td>
</tr>
<tr>
<td>Pre-weaning growth (Weaning weight)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Longissimus</td>
<td>Temperate</td>
<td>Slow</td>
<td>40.5</td>
<td>39.2</td>
</tr>
<tr>
<td>Semitendinosus</td>
<td>Temperate</td>
<td>Rapid</td>
<td>46.3</td>
<td>46.3</td>
</tr>
<tr>
<td>120 days post-weaning growth</td>
<td>Weight loss</td>
<td>Rapid growth</td>
<td>76.5</td>
<td>73.5</td>
</tr>
<tr>
<td>Longissimus</td>
<td>Tropically-adapted</td>
<td>50.0</td>
<td>48.1</td>
<td>&gt;0.10</td>
</tr>
<tr>
<td>Semitendinosus</td>
<td>Tropically-adapted</td>
<td>40.0</td>
<td>40.8</td>
<td>&gt;0.10</td>
</tr>
<tr>
<td>Backgrounding growth</td>
<td>Longissimus</td>
<td>Temperate</td>
<td>Slow</td>
<td>40.0</td>
</tr>
<tr>
<td>Finishing system</td>
<td>Longissimus</td>
<td>Temperate</td>
<td>Pasture</td>
<td>42.7</td>
</tr>
<tr>
<td>Semitendinosus</td>
<td>Temperate</td>
<td>Feedlot</td>
<td>48.6</td>
<td>44.1</td>
</tr>
<tr>
<td>Longissimus</td>
<td>Tropically-adapted</td>
<td>49.2</td>
<td>45.5</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Semitendinosus</td>
<td>Tropically-adapted</td>
<td>49.3</td>
<td>45.0</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Figure 1
Variation in within-breed heritability with days aged for *longissimus* shear force in beef for two studies. Each point is an estimate, with the standard error of the estimate indicated by a vertical line. The data is derived from O’Connor *et al.* (1997) and Wulf *et al.* (1996)