Effects of Selection for Low Residual Feed Intake on Meat Quality of Major Muscles from Angus, Charolais and Kinsella Composite Cattle

by

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Abstract

Residual feed intake (RFI) is a measure of animal feeding efficiency. Selection of high feed efficient cattle using RFI does not affect production performance; low RFI animals consume less feed to reach a similar body weight gain as high RFI animals and thus are considered as feed efficient cattle. However, reducing production costs by selecting low RFI cattle will be beneficial only if beef quality of efficient animals is not adversely affected. Moreover, the response of different beef breeds or crossbred cattle to RFI selection may not be consistent. Therefore, the main objective of this research was to determine the effects of breed and RFI on carcass quality, objective meat quality and sensory palatability of five major beef muscles: m.longissimus lumborum (LL), m.longissimus thoracis (LT), m.triceps brachii (TB), m.semimembranosus (SM) and m.gluteus medius (GM). Twelve high and twelve low feed efficient Angus, Charolais and Kinsella Composite cattle were selected for this study. RFI selection had limited influence on performance traits, carcass quality, instrumental meat quality, sensory palatability assessed by a trained panel and a consumer panel, with some exceptions. RFI selection for feed efficient animals negatively affected consumer acceptability of ribeye steaks from Angus and Kinsella Composite and sensory quality of TB for all breeds. Moreover, the interaction between breed, RFI and postmortem aging showed that postmortem aging did not improve tenderness of ribeye steaks from low RFI animals of any breed, thus breeding for feed efficient animals using RFI selection may affect meat quality and consumer preference of steaks from some beef muscles. Breed significantly affected most traits assessed in this study; steaks from Angus had better meat sensory

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quality assessed by both trained and consumer panels, while Charolais had better yield quality with greater hot carcass weight, carcass ribeye area and lean meat yield. The interaction between breed and RFI selection showed that the effects of RFI on meat quality was not consistent among breeds. Therefore, RFI may be a beneficial tool to reduce feed costs by selecting feed efficient animals, but it had possible adverse influence on quality and consumer preference of meat in some breeds.

Preface

This thesis contains two studies to investigate the effect of selecting feed efficient animals using RFI beef quality. Beef cattle were reared at University of Alberta Kinsella cattle herd according to Kinsella animal care protocol: Aup 00000777. For the first study (Chapter 2), the data were provided by Agriculture and Agri-Food Canada's Lacombe Research and Development Centre and were analyzed by me. This study received research ethics approval from University of Alberta Research Ethics Board, Project Name "Genetics of the eating quality of high connective tissue beef", No. Pro00054386, Date: June 24, 2015. This chapter was written and formatted for publication in the *Journal of Animal Science* with Dr. Wendy Wismer, Dr. Manuel Juarez, Dr. Carolyn Fitzsimmons, Dr. Changxi Li, Dr. Graham Plastow, Dr. Jennifer Aalhus as co-authors.

For the second study (Chapter 3), the experimental design, data collection and data analyses are my original work, with the assistance of Dr. Wendy Wismer and Ha Nguyen. This study received research ethics approval from University of Alberta Research Ethics Board, Project Name "Consumer evaluation of beef steak", No. Pro00064155, Date: August 28, 2016. This ethic application also received amendment approval, Project Name "Consumer evaluation of beef steak", No. Pro00064155_AME2, Date: October 06, 2016. This thesis chapter was written and formatted for publication in the journal *Meat Science*, with Dr. Heather Bruce, Ha Nguyen, Dr. Manuel Juarez, Dr. Carolyn Fitzsimmons, Dr. Changxi Li and Dr. Wendy Wismer as co-authors.

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Abbreviations

ADG	Average daily gain
AFAT	Average backfat thickness
AMSA	American Meat Science Association
ANOVA	Analysis of variance
BF	Back fat thickness
BW	Body weight
CBBC	Canadian Beef Breeds Council
DMI	Dry matter intake
DOT	Days on test
FAO	Food and Agriculture Organization
FCR	Feed conversion ratio
FUFAT	Final ultrasound backfat
GPA	Generalized Procrustes analysis
HCW	Hot carcass weight
IMCT	Intramuscular connective tissue
LD	m.longissimus dorsi
LL	m.longissimus lumborum
LMA	Longissimus muscle area
LMY	Lean meat yield
LT	m.longissimus thoracis
LY	Lean yield
MWT	Metabolic body wight
PCA	Principle Components analysis
PREFMAP	Preference mapping
QDA	Quantitative Descriptive Analysis
REA	Carcass rib eye muscle area
RFI	Residual feed intake

RFIf	RFI value adjusted for backfat thickness
SEM	Standard error mean
SM	m.semimembranosus
ТВ	m.triceps brachii
USDA	United States Department of Agriculture
WBSF	Warner-Bratzler shear sorce
WHC	Water holding capacity

Chapter1: Introduction and literature review

Normally, when researchers investigate the effects of genetics, management or other factors on beef quality, the three major components of carcass quality, meat quality and meat palatability are measured. Why are these three components important for beef studies? According to a previous study (Warriss, 2010), agricultural systems, including the beef production system, usually have three phases of development. The first phase is to produce enough meat to meet human needs. When the first aim has been achieved, beef producers are motivated to increase the benefits of production by raising beef more efficiently. Thus, attention to the carcass yield of beef, back fat depth, fat to muscle ratio and other carcass quality characteristics are crucial to produce more edible meat, and then to increase the total value of the animals. Finally, when consumers have plenty of choices to purchase meat, meat quality and palatability become increasingly important. Ultimately, after achieving the goal of sufficient meat production, beef producers look to reduce production costs, while consumers want to pay less for the best quality meat. Therefore, to increase the profits of beef production, beef producers should either reduce production costs or increase beef quality to achieve an increased unit value of meat.

One of the effective methods of reducing production costs is to reduce costs of feed, because the major cost of beef production is feed (Herd et al., 2003; Arthur et al., 2004). Therefore, increased feed efficiency could result in higher profitability of beef production. Beef industries have attempted several ways to improve feed efficiency, such as selection for highly efficient animals within the same breed or selection for highly efficient breed types (Herd et al., 2003). To calculate and select animals with high feed efficiency, several methods have been used such as feed conversion ratio (FCR) and gross efficiency. Selection for animals with reduced or low residual feed intake (RFI), initially suggested by Koch et al. (1963), has become a popular method to select high feed efficient animals, because of its potential to increase beef production efficiency (Basarab et al., 2003).

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Although selection for highly efficient animals may reduce feed costs and increase profits of a beef enterprise, breed type mainly determines the profitability related traits such as mature cattle size and growth (Greiner, 2005). Thus, selection or crossing of breeds is another method to reduce costs of animal production and increase value of meat when sold (Chewning et al., 1990), because there exist genetic differences among breeds and even among animals within the same breed (Smith et al., 2011). Moreover, the response of selection for RFI may not be consistent in different pure-bred or cross-bred cattle (Baker et al., 2006). Comparison of growth performance and carcass and meat quality traits among different breeds can help producers select the most appropriate breeds for their specific environments and handling systems (Campo et al., 2000).

Consumer demand for consistent and high-quality beef products makes it important to study the influence of beef breeding programs (i.e. selection for breeds or selection for low RFI animals) on carcass and meat quality. Thus, several quality traits of meat, including pH, water-holding capacity, shear force and sensory characteristics, should be assessed as part of breeding programs, because it is important to ensure that the palatability of beef products is not negatively affected by selection for low RFI animals from a specific breed. Therefore, understanding the relationships among RFI, breed and carcass and meat quality of beef is the primary objective of this thesis.

1.1 Feed efficiency

Feed efficiency is the ability of an animal to utilize feed and turn feed (inputs) into meat (outputs) and has a major influence on the unit cost of production (Basarab et al., 2003). Feed efficiency is particularly important to beef production systems. Approximate 5% of dietary energy intake is utilized by beef cattle to build muscle, while the utilization rate of poultry and pork is higher at about 20% and 14%, respectively (Ritchie, 2000). Beef production is a relatively inefficient livestock production system to utilize energy and requires more feed to produce the same amount of meat compared to chicken and pork production (Kelly, 2015). Feed costs account for the majority of costs

in beef production and constitute 60% to 70% of the total costs of production (Karisa, 2013). Thus, increasing the feed efficiency of beef is important to reduce overall feed costs and then increase net profit of beef production.

Increased feed efficiency not only shows significant economic benefits, but also potential benefits in environmental sustainability (Arthur et al., 2004; Bezerra et al., 2013). According to a recent report by Food and Agriculture Organization (FAO) (Gerber et al., 2013), beef cattle accounted for 41% of the total emissions of global livestock due to the huge amount of methane produced in the rumen. As reviewed by Bezerra et al. (2013), increased feed efficiency has the potential to reduce environmental impact by reducing methane emissions.

Dry matter intake (DMI) and average daily gain (ADG) are two major traits evaluated by beef researchers and beef producers to monitor the growth performance of animals. Dry matter is the dry portion of the feed and indicates the total amount of nutrients available in the feed. To meet the energy and nutrient requirements to maintain normal body functions, a certain amount of dry matter will be needed for cows every day. DMI of an animal depends on body size and weight.

ADG is the average gain in weight per day of the animal raised over a period of time and is largely depend on the DMI. Researchers calculate feed efficiency by combining feed input (DMI) and output (ADG). Archer et al. (1999) reviewed the traditional methods to measure efficiency, including FCR or gross efficiency, maintenance efficiency and partial efficiency of growth, which are all expressed as the ratio of feed: gain or gain: feed and are used for different purposes. FCR, one of the most widely used measures so far, is defined as feed intake per unit weight gained. Animals with high FCR are inefficient in converting feed into weight. According to the literature, FCR is correlated with growth performance of animals both phenotypically and genetically (Archer et al., 1999), indicating that selection for improved FCR improves feed efficiency. However, there are limitations of using FCR or other ratio traits in beef cattle selection for breeding. One disadvantage of selection for improved FCR is that it may not improve the efficiency of the entire production period even though it may improve the efficiency during the growth and finishing phases of beef production (Nascimento et al., 2016). An additional limitation is due to the nature of ratio traits. For example, low FCR animals consuming a low amount of feed may exhibit a small weight gain and thus take a long period to meet the minimum market weight for slaughter (Kelly, 2015), causing wastes of other operation costs, such as the pasture rental and veterinary medicine (Blawat, 2004). Animals consuming a high amount of feed may have a similar FCR to animals consuming less feed, as long as they have relative gain according to their feed intake during the set period. Thus, using only FCR or other ratio traits to measure feed efficiency for genetic improvement is not accurate (Moore et al., 2009).

1.1.1 Residual feed intake

Due to several drawbacks of using ratio traits in breeding programs, linear type measures have been studied to increase the accuracy of predicting feed efficiency. Currently, residual feed intake (RFI) is one of the popular methods to measure feed efficiency. RFI is defined as the difference between an animal's actual and expected feed intake for a specific test period (Koch et al., 1963; Basarab et al., 2003). The equation for RFI calculation is: RFI = DMI actual – DMI predicted based on ADG and metabolic body weight (MWT). RFI has proven to be a valuable method to select feed efficient animals (Kelly, 2015). Recently, the influence of RFI on production performance has been widely studied and showed a positive result of selecting for low RFI animals (Cruz et al., 2010; Zorzi et al., 2013). However, reducing the cost of feed will be beneficial only if the carcass and meat quality of beef are not affected significantly by breeding for low RFI animals. Thus, more studies on both traits should be performed to fully understand the influence of RFI.

1.1.2 Effect of RFI on carcass quality, meat quality and palatability

Almost all studies investigating the relationship between RFI and carcass quality agree that selecting low RFI animals will have little influence on carcass traits (i.e.

average back fat thickness (AFAT), USDA yield grade, marbling score, carcass rib eye muscle area (REA), quality grade, hot carcass weight (HCW), lean meat yield (LMY) (McDonagh et al., 2001; Basarab et al., 2003; Baker et al., 2006; Gomes et al., 2012; Zorzi et al., 2013). An insignificant phenotypic correlation between RFI and carcass traits further confirms the minimal effect of RFI selection on carcass quality (Cruz et al., 2010; Mao et al., 2013). However, Janelle (2015), using 156 Angus × Simmental steers, found that low RFI animals tended to have heavier HCW, larger REA, and higher LMY and concluded that higher lean yield of low RFI animals indicated less fat deposited during the growth period on the assumption that high RFI animals (or inefficient animals) would consume more DMI than required, and thus deposit more fat.

The effect of selection for low RFI animals on meat quality remains contradictory, as varying results have been reported to date. Baker et al. (2006) investigated the relationship between RFI and meat quality traits among 54 purebred Angus steers, but found no significant difference on meat quality (i.e. Warner-Bratzler shear force (WBSF) and sensory tenderness and flavor) between high and low RFI groups of animals. Moreover, the WBSF of all RFI groups fell within the industry standard range (< 4.1 kg) of Australia. Similar results on carcass quality and WBSF have been found by other researchers (McDonagh et al., 2001; Ahola et al., 2011; Gomes et al., 2012). On the other hand, some studies found possible negative consequences on meat quality of selecting efficient animals based on RFI. Zoizi et al. (2013) conducted a study with 59 young Nellore bulls to determine the possible relationship between RFI and meat quality and found that *longissimus* steaks of low RFI animals tended to have higher shear force values, indicating tougher meat. McDonagh et al. (2001) also concluded a possible negative effect of selecting low RFI animals on meat tenderness by observing a lower rate of myofibrillar fragmentation and higher calpastatin concentration in low RFI animals. The myofibrillar fragmentation index can be used to reflect the proteolytic activity (Zorzi et al., 2013), since a positive correlation of the ratio of μ -calpain to calpastatin (protease to inhibitor) with the rate of myofibril fragmentation was found (McDonagh et al., 2001). The high level of calpastatin in muscles inhibits the activity of

calpain (Koohmaraie and Geesink, 2006) increases the rate of myofibril fragmentation, reduces the rate of proteolytic activity, and subsequently reduces the tenderness of meat (Baker et al., 2006).

The influence of selecting low RFI on the sensory palatability of steaks is not clear as few studies have been conducted so far. In the study conducted by Baker et al. (2006), a nine-person trained sensory panel evaluated steaks from longissimus muscles and observed that the tenderness, flavor and off-flavor intensity of steaks were similar among animals from three RFI levels, while steaks from low RFI animals tended to be juicier. Ahola et al. (2011) also found no relationship between RFI and sensory attributes (i.e. tenderness, juiciness, flavor, and overall acceptability) assessed by a trained sensory evaluation panel. However, in the study conducted by Kelly (2015), a trained panel gave lower intensity ratings to three texture attributes (softness, tenderness, and rate of breakdown) in LM muscles from low RFI groups of animals versus high RFI animals. The authors suggested that although the texture attribute differences between the two RFI groups were statistically significant, these differences may not influence consumers' eating experiences. Since no study has investigated the influence of RFI on consumer preference for beef steaks, no conclusion can be made so far. From previous studies, it appears that selection for low RFI animals does not significantly influence carcass quality, meat quality and palatability, but possible negative influences on meat quality should be taken into consideration when selecting feed-efficient cattle.

1.2 Beef breeds and meat quality

As reviewed by Greiner (2005), the profitability of beef production can be impacted significantly by selection of breed in the breeding program because of the different growth rates, mature cattle sizes and meat palatability of different breeds. As shown in numerous studies conducted to measure and compare the meat quality of different beef breeds, clear differences of carcass traits (DMI, ADG, carcass weight etc.) and meat attributes (shear force value, proximate analysis, sensory attributes etc.) exist among different breeds (Pringle et al., 1997; Campo et al., 1999; Chambaz et al., 2003; Barton et al., 2006). Therefore, selection of the appropriate breed is crucial for the success of beef production. Breed selection should consider several factors: production system, market demands, feed stuff available, climate, breed complementarity and the availability of purebred seed stock (Greiner, 2005).

Most cattle breeds belong to two species: *Bos taurus* and *Bos indicus* (Buchanan and Lenstra, 2014). *Bos taurus* cattle are adapted to both temperate and subtropical climates, but are not heat tolerant (Girard, 2010), while *Bos indicus* cattle are tropically adapted. Thus, the main breed type in Canada is *Bos taurus* or *Bos taurus* crossbred cattle. According to Canadian Beef Breeds Council (CBBC) (Canadian Beef Breeds Council, 2017c), there are twenty-six breeds of purebred *Bos taurus* beef cattle in Canada. Studies focused on meat quality differences of these two species revealed consistent high-quality and tender beef from *Bos taurus* compared to *Bos indicus* (Marshall, 1994; Wheeler et al., 1994; Shackelford et al., 1995).

Angus and Charolais, both *Bos taurus* species, are two breeds widely used on cow-calf operations in Canada according to the beef cattle registrations report on the CBBC website (Canadian Beef Breeds Council, 2017a). Angus breeds originated in the British Isles (British breeds), while Charolais breeds originated in the European Continent (Continental breeds) (Girard, 2010). As concluded by Greiner (2005), British breeds, including Angus, Hereford and Shorthorn in Canada, are generally regarded as small in size but with high meat quality, while Continental European breeds, including Charolais, Gelbvieh, Limousin, Maine-Anjou and Simmental in Canada, are regarded as large in size but with low quality grade. The popularity of Angus cattle in Canada is due to its high carcass quality, such as superior marbling ability and tender meat, while Charolais cattle are famous for their great adaptability to the Canadian environment (Canadian Beef Breeds Council, 2017b), but with relative low marbling score and tenderness (Buchanan and Lenstra, 2014). Apart from these differences, Angus cattle tend to have a low to moderate lean-fat ratio and reach puberty at an early age, while Charolais cattle have a higher lean-fat value and reach puberty at a later age (Buchanan and Lenstra, 2014)

Significant meat quality and meat palatability differences exist among Bos taurus breeds, although these differences are smaller than the differences between Bos taurus and Bos indicus. Bures et al. (2006) found that longissimus lumborum (LL) muscles from Angus were rated with the highest overall liking for all sensory attributes (odor, flavor, texture, and juiciness) compared to samples from Charolais and Simmental bulls (Continental European breeds). However, different results were found in a study comparing meat quality of Angus, Simmental, Charolais and Limousin steers at the same marbling score (Chambaz et al., 2003). Longissimus dorsi (LD) muscles from Angus and Charolais had WBSF values, myofibrillar fragmentation index, total collagen and collagen solubility and sensory tenderness, indicating similar meat tenderness of these two breeds. The only significant difference was that Charolais LD muscle was rated as juicier than Angus. The dissimilarity of these study results compared to those previously discussed may be due to differences in slaughter endpoints; animals from this study (Chambaz et al., 2003) were slaughtered at the same intramuscular fat content (same marbling score) on different days, reflecting the importance of marbling in sensory attributes of meat. However, studies generally agree that the sensory rating of Charolais cattle is poorer than the Angus when both breeds reach similar maturity (Sinclair et al., 2001; Bures et al., 2006).

1.3 Other factors that affect beef quality

1.3.1 Post-mortem aging

Post-mortem aging is the practice of storing meat at low temperature, and has long been known to improve beef palatability (Jones et al., 1991; Jiang et al., 2010), especially the tenderness of meat (Brewer and Novakofski, 2008). Endogenous enzyme activities, especially that of calpains (Lamare et al., 2002), on degradation of muscle cytoskeletal proteins (i.e. structural myofibrillar proteins), mainly contribute to the increase of tenderness (Spanier et al., 1990) and influence water-holding capacity of meat (Huff-Lonergan and Lonergan, 2005) during the post-mortem aging period. As reviewed by Girard (2010), calpains have been reported to degrade actin, myosin light chain, troponin T, titin, desmin, troponin I, vinculin, nebulin, synemin, and vimentin and thus result in improved meat tenderness and possible drip production (Huff-Lonergan and Lonergan, 2005). Monson et al. (2005) found that the differences of textural characteristics among breeds were reduced through long aging times (greater than seven days), proving the importance of aging during beef processing.

Due to hydrolytic activity during aging, significant alterations occur to flavorrelated components such as sugars, organic acids, peptides and free amino acids and metabolites of adenine nucleotide influence meat flavor (Spanier et al., 1997). However, the impact of aging on meat flavor is still controversial. No influence of aging times (1, 3, 7, 14, 21 and 35 days) on beef flavor intensity was found by Monson et al. (2005) using a trained panel. Similar results were also reported by Brewer et al. (2008) using a consumer panel. A significant influence of aging was observed in other studies (Jeremiah et al., 1991; Campo et al., 1999; Bruce et al., 2005). The study conducted by Jeremiah et al. (1991) showed that post-mortem vacuum packaged aging times (up to 28 days) increased the intensity of beefy flavor. Moreover, undesirable tastes such as bitter and sour are reported to increase during aging time (Spanier et al., 1997; Bruce et al., 2005) and negatively impact meat palatability.

The positive effect of aging on meat tenderness is well accepted, while the influence of aging on juiciness and flavor is not conclusive. Therefore, more studies should be conducted with a focus on the influence of aging on overall consumer preference.

1.3.2 Carcass muscle location

It is generally known that different muscles within the same beef carcass vary in size, shape, location, weight, pH, composition, ultrastructure and function (Ramsbottom

and Strandine, 1948; Searls et al., 2005), and thus vary in tenderness, flavor intensity, juiciness and cooking losses (Jeremiah et al., 2003; Calkins and Sullivan, 2007). Meat from rib and loin cuts have been proven to be tender and generally have higher value than meat from chuck and round, some of which are processed into ground products to improve their value (Belew et al., 2003; Lepper, 2013).

Factors such as muscle fiber characteristics, connective tissue and intramuscular fat contribute to variation in tenderness and other attributes (Torrescano et al., 2003; Dubost et al., 2013; Joo et al., 2013). Connective tissue, mainly collagen, located throughout the muscle contributes to "background" toughness (Torrescano et al., 2003; Calkins and Sullivan, 2007). Torrescano et al. (2003) assessed total and insoluble collagen of fourteen bovine muscles to evaluate the relationship between Warner-Bratzler shear force and collagen characteristics, and observed high positive correlations between both total and insoluble collagen content and Warner-Bratzler shear force. Samples from Triceps brachii (Chuck cuts) and Semimembranosus (Round cuts) had higher total and insoluble collagen and shear values compared with samples from Gluteus medius (Top sirloin cuts) and Longissimus lumborum (Loin cuts). Muscles used for maintaining posture (i.e., Longissimus) are more oxidative and contain more slow-twitch type I fibers than muscles used for rapid movements, containing more fast-twitch type IIa and type IIb fibers (Hill, 2012; Joo et al., 2013). Renand et al. (2001) found that muscles with larger fiber size (i.e., type IIb) were tougher than meat with smaller fiber size. Moreover, intramuscular fat content significantly affecting meat juiciness and flavor was positively correlated with type I content (Hwang et al., 2010; Joo et al., 2013).

Numerous studies have assessed WBSF, palatability attributes and consumer preference of different muscles of the beef carcass (Keith et al., 1985; Jeremiah et al., 2003; Torrescano et al., 2003; Belew et al., 2003; Calkins and Sullivan, 2007) and observed that several muscles were undervalued and underutilized. In muscle profiling research conducted by University of Nebraska and University of Florida, flat iron steaks from *Infraspinatus* muscles were found with exceptional tenderness (Calkins and Sullivan, 2007). Flat iron steaks were rated by a trained sensory panel with high initial and overall tenderness and flavor intensity and were similar to several steaks from rib and loin cuts (Jeremiah et al., 2003). These results indicate that there is potential to add value to steaks that are often underutilized in the market. Therefore, studies of beef meat quality should not only focus on high value steaks such as rib-eye and tenderloin, but also on lower value cuts such as round and chuck, to realize greater profit for meat industries.

1.3.3 Collagen content and soluble collagen

The amount, structure and composition of intramuscular connective tissue (IMCT) in muscles contributes to the toughness of meat (Nishimura, 2010; Dubost et al., 2013). McCormick et al. (1999) concluded that the variable amount of collagen, the major component of IMCT, and collagen crosslink maturity are two factors accounting for the development of the toughness of meat. Listrat et al. (2007) found that mature crosslinks (i.e., pyridinoline crosslinks) positively contributed to meat toughness. Collagen heat stability and solubility depend on the maturity of the crosslink (Nishimura, 2010) as agreed by McCormick (2009), who concluded that the increased conversion from immature crosslinks into mature crosslinks leads to decreased solubility of collagen in cooked meat. Thus, the concentration of soluble collagen in the muscle typically represents the percentage of immature crosslinking (Rompala and Jones, 1984).

Many studies have attempted to relate collagen amount to meat tenderness but failed to establish a clear conclusion. Some studies have found a significant influence of collagen content on meat tenderness (Light et al., 1985; Nishimura et al., 1995; Dubost et al., 2013). However, several studies have found a poor correlation between collagen quantity and overall tenderness and suggested that collagen quality influences meat tenderness to a greater degree than collagen quantity (Keith et al., 1985; Weston et al., 2002). An investigation conducted by Bosselmann et al. (1995) on the mature crosslink of bovines from different ages showed significant variation in meat tenderness associated with age while there was no significant difference in collage quantity, indicating that maturity of crosslink contributed to the toughness of beef meat. Some other researchers, however, found a low correlation between collagen maturity and tenderness of meat (Chambaz et al., 2003).

The contradictory findings may be due to the additive effect of collagen content and mature crosslink concentration on the toughness of meat (Weston et al., 2002). By comparing collagen concentration and collagen crosslinks in five different bovine muscles, McCormick et al. (1999) found that increasing concentration of both traits contributed to tougher meat. A lower impact on meat toughening development was found when the concentration of either of the factors was low.

1.4 Beef quality and measurement

As previously discussed, beef producers are willing to select the appropriate breed or adopt new production methods to improve beef quality to increase carcass value and meet consumers' needs. Before making changes to the beef production system, it is important to ensure the changes will not negatively affect meat quality. Therefore, it is essential to measure beef quality efficiently.

1.4.1 Carcass quality and measurement in Canada

Previous studies indicated that consumers are willing to pay more for guaranteed tender beef (Miller et al., 2001). Thus, to ensure that consumers can purchase beef with consistent good quality and that producers get paid for their efforts to improve beef quality, a beef grading system was developed and introduced in Canada in 1929 and has been improved to fulfill the need of beef industries (Government of Canada Publications, 1978).

The Canadian Beef Grading System measures several carcass quality traits, including AFAT, REA, marbling scores, HCW and LMY. The AFAT, REA and marbling score are all measured at the cut surface of the ribeye muscle or *longissimus* muscle between the 12th and 13th rib. Backfat helps prevent the carcass from chilling too quickly, resulting in reduced tenderness (Aalhus et al., 2001). Jeremiah et al. (1996) investigated

293 crossbred beef carcasses and found that the LT steaks with greater subcutaneous fat thickness (more than 1.40 cm) had better tenderness and overall acceptability by a trained panel and had a higher juiciness acceptability by a consumer panel than those with reduced fat thickness (less than 0.59 cm), indicating a positive relationship between fat thickness and palatability attributes. According to the Canadian Beef Grading System, youthful carcasses with bright red muscle, more than 2 mm backfat thickness, and firm, white fat color qualify for at least A grades, and are considered as high-quality products, while carcasses with less than 2 mm backfat are assigned to B grades (Canada Gazette, 2007).

Marbling score in beef has a small, but positive association with the better meat palatability and acceptance (Wheeler et al., 1994; Jeremiah, 1996; Morales et al., 2013). In Canada, marbling is measured on the basis of average amount, size and distribution of fat particle in the ribeye muscle, and is assessed as being either trace, slight, small or slightly abundant (Canada Gazette, 2007). Marbling in the ribeye muscle is an important factor of beef grade. The carcasses with at least slightly abundant marbling in the ribeye muscle will be assigned to Canada Prime, the highest quality grade in Canada. Canada AAA, AA, and A graded carcasses have less marbling than Canada Prime, but will have traces, slight and small of marbling in the ribeye muscle, respectively (Canada Gazette, 2007).

The carcass lean yield (LY) of all carcasses receiving Canada A grade or above is assessed as Canada 1 (more than 59% estimated yield), Canada 2 (54% to 58%) and Canada 3 (less than 53%) according to the Canadian Beef Grading Agency. LY of a carcass is measured with a Yield Ruler, developed by Agriculture and Agri-Food Canada's Lacombe Research Station in 1992 and modified in 2011 (Kelly, 2015) based on the equation Lean% = 63.65 + 1.05 (muscle score) – 0.76 (grade fat). Muscle scores are determined by the width and length of *longisimus thoracis* muscle (rib-eye muscle) as rib-eye muscle size reflects carcass size (Karisa, 2013), while grade fat is determined by backfat depth (Aalhus et al., 2014). Consistent with other assessments, the ruler is applied on the surface of *longissimus* muscle between the 12^{th} and 13^{th} rib to determine muscle score and fat class. Therefore, carcass traits evaluated in the beef grading system, including AFAT, REA, marbling score, HCW, LMY, should be measured in research studies to ensure that breed selection and processing practices will not adversely influence meat quality grade and yield grade.

1.4.2 Instrumental meat quality analysis

As discussed above, improving carcass quality helps beef producers raise cattle with consistent high yield and better quality. However, carcass quality is not the only important factor for beef producers; they also need to pay attention to improving meat quality and palatability. For consumers, meat eating experiences play an important role when purchasing beef products (Maltin et al., 2003). Meat quality and palatability consist of a range of attributes, including tenderness, juiciness, flavor, color and water holding capacity. Several primary methods to evaluate meat quality are reviewed here.

1.4.2.1 Warner-Bratzler shear force

Tenderness is the most important sensory attribute for beef consumers (Verbeke et al., 2010). Moreover, the variability of meat tenderness is the major reason for consumers not to purchase beef products (Maltin et al., 2003). Numerous studies have been performed to understand tenderness and its measurement. Warner-Bratzler Shear Force is a traditional instrumental measure to evaluate beef meat tenderness. The idea to use a steel blade to slide through a sample to measure the amount of force needed to shear the meat sample was first proposed by K.F. Warner and colleagues in the 1920's, and was then modified by L.J. Bratzler to increase the test accuracy by standardizing the blade thickness, shape and speed (Zamarripa, 2014). The initial purpose of using an objective instrument is to measure meat tenderness consistently. However, the fact that different institutes use different equipment, cooking methods and other factors leads to variation of tenderness determination results (Zamarripa, 2014). Therefore, following a widely-accepted protocol is important and would make it possible to compare shear force values from different studies. In Canada, the most commonly used protocol is the

American Meat Science Association (AMSA) research guideline (AMSA, 2016), in which the blade is standardized with 0.046 inch thickness, 60 ° angle, half-round beveled cutting edge. The protocol advises the use of consistent diameter cores (0.5 inch) of steaks chilled overnight for the measurement of shear force values.

1.4.2.2 Water holding capacity and pH of meat

The water holding capacity (WHC) is defined as the ability of meat to retain moisture when external force or treatments are applied (Bouton et al., 1972). WHC is a key characteristic of meat as it is associated with meat appearance and perceived juiciness (Warriss, 2010), meat nutrient loss and carcass yield (Huff-Lonergan and Lonergan, 2005). It is reported that unacceptable WHC of meat causes millions of dollars in losses for the meat industry every year (Huff-Lonergan and Lonergan, 2005). Thus, controlling water loss during the post-mortem period and other processing procedures is important for beef producers to reduce loss and increase meat palatability.

The mechanisms of drip or purge loss have been well reviewed by Huff-Lonergan et al. (2005). In general, the rate of pH decline and final pH are two factors affecting the water-binding capability of fresh meat. The rapid pH drop in the warm muscle results in denaturation of myofibrillar protein, bound with a large proportion of water in meat. The extreme low ultimate muscle pH can lead to high drip loss. Breed selection and product handling play major roles influencing the WHC of meat, while other factors, such as aging period, storage condition and time, also influence moisture loss of meat.

Drip loss or purge loss measurements are two commonly used methods to quantify an estimate moisture loss from raw meat (Huff-Lonergan and Sosnicki, 2010). As discussed above, drip and purge loss are mainly caused by the rate of pH decline and ultimate pH of meat, affected by a variety of factors. Cooking loss, however, is mainly affected by cooking methods and cutting sizes. As reviewed by Zamarripa (2014), heat during cooking leads to evaporation of water, protein denaturation, collagen fiber shrinkage, and sarcomere length shortening, and then results in water unbound with myofibrillar protein.

1.4.2.3 Proximate analysis

Proximate analysis assesses important chemical components of meat, including moisture, protein, lipid and ash. Analytical methods of proximate analysis are described by the Association of Official Analytical Chemists (AOAC, 1995) and thoroughly reviewed by Ono et al. (1984).

Nutrients of meat, including protein, fat and minerals, are critical to human health (Karakök et al., 2010). Meat is a great source of high quality proteins and minerals such as iron, copper, zinc and manganese (Bender, 1992), and various fats, providing some essential fatty acids like alpha-linoleic acid (ALA) and omega-3 fatty acid (Williams, 2007). Although saturated fatty acids in the diet, supplied by meat fat, have been investigated as a contributing factor of coronary heart disease (Bender, 1992), the trend of preparing meat by trimming external fat makes it possible to reduce fat consumption with relative low fat content (<7%) in trimmed lean red meats (Williams, 2007).

The chemical composition of meat influences meat palatability in addition to its nutritional value. Collagen, the major component of connective tissue, is one of the proteins in meat. Connective tissue contributes to the toughness of meat and thus influences meat palatability (Dubost et al., 2013). Intramuscular fat between the muscle fiber bundles, known as marbling, is positively related with greater beef flavor intensity and other sensory attributes (Morales et al., 2013).

1.4.3 Meat quality analysis by sensory evaluation

In addition to instrumental and chemical measurements, sensory evaluation is a widely-used method for meat quality assessment. Since the 1900s, numerous studies have been conducted to understand the relationship between meat attributes and the eating experience. Trained panels and consumer panels are two available options for meat scientists to use to conduct sensory evaluation of meat, depending on the objectives of the studies.

1.4.3.1 Trained sensory evaluation panels

Trained sensory panels have been widely applied to assess meat quality in research (Kerth and Miller, 2015) and the meat industry (Monsón et al., 2005) to examine the effect of factors, such as new production practices, breed and post-mortem aging, on the palatability of meat and meat products. For example, to determine if the palatability traits of eight muscles from beef chuck could be enhanced by adding water, salt, and phosphates, Molina et al. (2005) employed an 8-11 member trained sensory panel to evaluate five sensory attributes (juiciness, beef flavor intensity, overall tenderness, connective tissue, and off flavor). The results showed the palatability traits were generally enhanced by brine treatments.

A highly trained sensory panel, screened for superior sensory acuity and trained to improve their ability to evaluate samples consistently, can be regarded as an objective measurement to provide accurate sensory descriptions of products. The AMSA sensory guideline (2016) provides a list of descriptive analysis methods for the trained panel, including Quantitative Descriptive Analysis® (QDA), Spectrum®, Flavor and Texture profile methods, and detailed procedures to recruit, screen and train panelists. Wellorganized training will help panelists understand the sensory test procedure, improve their ability to identify the sensory attributes, and also improve their reliability of sensory judgments (AMSA, 2016). A limited number of attributes are applicable to meat in most meat quality studies, such as beef flavor intensity, juiciness, connective tissue amount, muscle fiber tenderness, and overall tenderness. These attributes all are defined and reference standards are available (AMSA, 2016). The sensory panel leader in the studies can select and provide the sensory panel with a list of descriptors that are the most relevant to the objectives. Compared with non-sensory methods, trained panels can provide perception of meats to reflect the eating experience.

1.4.3.2 Consumer sensory evaluation panels

Consumers provide their acceptance or preference of products. Consumers' opinions are valuable to beef producers, as consumers are the potential customers who

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will purchase their products. One of the early beef consumer tests was conducted by Francis et al. (1977), who evaluated consumer rating of flavor, juiciness, tenderness and overall acceptability on steaks with two marbling levels and found that all the four sensory attributes were rated with higher values for the modest marbling samples than the slight marbling samples. Consumer data used in this study helped to understand the variability in beef palatability caused by the amount of marbling of the steaks. Since then, the consumer panel has become a research tool to evaluate the effects of various factors on meat quality acceptance (Kerth and Miller, 2015).

As trained and consumer sensory panels provide different information about samples (product descriptions and product preferences or acceptance, respectively), the two panels can be used together to comprehensively evaluate products. To investigate the breed effect on meat quality throughout the aging period, Monson et al. (2005) conducted both trained and consumer panels. The trained panel evaluated 10 sensory attributes (beef odour intensity, liver odour intensity, tenderness, juiciness, residue, beef flavor intensity, liver flavor intensity, bitter flavor intensity and overall acceptance), while consumers rated their acceptability of tenderness and flavor and overall liking. Both consumers and trained panelists found no significant differences in flavor, but significant differences in tenderness among the breeds, indicating that both types of panels can assess meat quality effectively from different perspectives.

1.4.3.3 Consumer sensory evaluation to develop product profiles

Trained panels are customarily used for profiling product characteristics of foods, because researchers traditionally believe that trained panels have improved sensitivity due to dedicated training sessions and they doubt consumers' ability to discriminate and profile the products accurately (Worch et al., 2010). However, most studies using trained panels rather than consumer panels do not provide evidence of superiority of trained panels over consumer panels (Moskowitz, 1996). In fact, several studies comparing data from trained and consumer panels refute the notion that the responses of consumers to product characteristics are not reliable and valid (Moskowitz, 1996; Husson et al., 2001; Worch et al., 2010)

Worch et al. (2010) conducted a study to compare profiling data from expert and consumer panels on the same 12 perfumes. The results of the study supported the hypothesis that consumers were similar to trained panelists; their evaluations were reproducible and they were able to discriminate the differences among products when they were well instructed. However, this study also observed some disadvantages of using consumer panels for classic product profiling; trained panelists displayed more consistent evaluations than consumers because of their better knowledge of perfume products, and consumers were limited to their own list of attributes because they were not trained to use the extensive sensory lexicon. Therefore, these issues should be addressed when using consumers to profile products. Firstly, consumers selected for descriptive sensory tasks should be familiar with and be knowledgeable about the tested products to keep consistent evaluation of products. Also, the size of the consumer panel should be larger than the trained panel. Additionally, the terms used in the consumer panel should be straightforward and easily understandable as descriptors used in the traditional descriptive analysis might not be easily understood by consumers, leading to the misunderstanding of sensory attributes (Le and Worch, 2014). For example, three basic attributes, beef flavor intensity, overall tenderness and juiciness, can be used in the consumer panel to evaluate steaks.

The advantages of using consumers rather than experts or trained panelists for profiling products include reduced time and costs and the generation of hedonic information and other valuable information such as ideal intensity of product attributes (Le and Worch, 2014). The sensory characteristic product profile and consumer preference obtained from a consumer panel can facilitate research and product development (Worch et al., 2010)

1.4.3.4 Generalized Procrustes Analysis (GPA) and preference mapping to understand consumer perceptions and preferences of products

GPA, generalized by Gower (1975), is one of the multivariate exploratory data analysis methods that provides graphic presentation of differences among products (Ferreira et al., 2008). GPA generates a consensus matrix by transforming individual data matrices through translation, rotation and scaling (Ferreira et al., 2008) and then Principal Components Analysis (PCA) can be applied to the consensus matrix to visualize the consensus (Society of Sensory Professionals, 2017b). Thus, GPA can be used to minimize differences among participants and find consensus in the sensory evaluation (Paulos et al., 2015). Moreover, GPA can provide graphic interpretation of product differences (Ferreira et al., 2008), relationships between products and panelists, and relationships between sensory attributes and products (Society of Sensory Professionals, 2017b). PCA can also be conducted to generate a multi-dimensional view of data to visualize the relationships between products and attributes (Society of Sensory Professionals, 2017c). However, GPA is superior to PCA in the presentation of such relationships based on panelist agreements (Meullenet et al., 2008). A comparison study of GPA and PCA conducted by Hunter and Muir (1995a) showed different configurations from these two methods and GPA showed better ability to differentiate samples and provide greater detailed information about differences between samples compared with PCA.

Preference mapping investigates the relationship between profiling data, generated from trained or consumer panels, and consumer hedonic data to provide a deeper understanding of consumers' liking of products (Costell et al., 2000). External preference mapping is based on the sensory map of a product generated by multivariate analysis (e.g., PCA, GPA) and with consumer hedonic scores fitted into the sensory space (Society of Sensory Professionals, 2017a). In the preference map, products are represented by points while consumer preference levels by vectors (Costell et al., 2000). The number of judges around a product indicate the popularity of it (XLSTAT, 2017). A colored plot can be drawn to calculate the percentage of consumers with above mean preference in a given area of the preference map (XLSTAT, 2017). The higher proportion of consumers with high preference scores, the hotter color the regions. When the preference map and contour plot map are superimposed, the preference ranking for samples can be concluded.

1.5 Conclusion

This literature review addressed the topics of influence of genetics (breeds, RFI, muscles) and other producing practices (aging) on beef quality, including growth performance, carcass quality, meat quality and palatability. The biggest challenges faced by the beef industry are the reduction of production costs, increasing their competitive ability, and improvement of meat quality, meeting consumers' demands for consistent high-quality products. Selection for appropriate breeds and animals with high feed efficiency is increasingly important for beef producers to reduce feed costs and reduce the environmental impact of beef production. RFI, a measure of feed efficiency, has shown its potential and value for use in breeding programs to select animals with high feed efficiency. However, concerns of adverse effects of using RFI on overall quality of beef make it vital to understand the influence of selection for reduced RFI animals. The interactions among RFI, breed and traditional production practices (aging) should be investigated to fully understand the effect of RFI selection on beef cut quality of popular Canadian breeds (Angus and Charolais). Additionally, the effect of RFI on consumer acceptance of beef steaks need to be evaluated, as consumers' satisfaction is the key to the success of the beef industry.

1.6 Summary and thesis structure

Based on the literature review of Chapter 1, reducing high feed costs is crucial to the success of beef industry. RFI has the potential to select breeds and animals with high feed efficiency without negatively influencing most production performance traits and meat quality traits, such as marbling score, size of rib-eye area, carcass weight, proximate composition and WHC. However, a possible negative influence of selecting high feed efficient animals using RFI on backfat thickness, shear force value and other quality traits has also been reported. Therefore, additional research would be valuable to understand the relationship between RFI and carcass and meat quality. In this study, we hypothesize that selection for reduced RFI may not affect beef carcass and meat quality. The primary objective of this thesis was to determine the influence of RFI on carcass and meat quality of different beef breeds as described in both chapter 2 and chapter 3.

Moreover, it is also important to understand the interaction between RFI and traditional production practices (i.e. post-mortem aging) used to increase meat tenderness (chapter 2 and chapter 3). The hypothesis is that RFI selection for high feed efficient animals will not adversely influence the tenderization of meat during post-mortem aging. In most research studies, meat quality analysis has been conducted only on the meat from high-value beef cuts, such as the *longissimus* muscle. However, the influence of RFI on the other meat muscles should be studied as well to understand the comprehensive influence of RFI on beef quality (chapter 2). We hypothesize that selection for low RFI will not influence the meat quality of high value meat cuts, such as loin and top sirloin, as well as the quality of tough cuts, like round and chuck.

As reviewed in chapter 1, there is currently limited research that investigates the effect of RFI on meat quality using sensory evaluation, an important measure to judge meat quality. Therefore, trained and consumer panels were used in chapter 2 and chapter 3, respectively, to ensure RFI does not affect meat palatability and consumer preference. The hypothesis is that animal selection for low RFI does not influence meat sensory palatability assessed by trained panel and consumer panel, as well as the consumer preference of meat.

This thesis is organized in four chapters with the literature review in chapter 1, a concluding summary in chapter 4 and two studies in chapter 2 and chapter 3 as described below:

Chapter 2 describes a research study to investigate the effect of RFI on beef quality and palatability of m.*longissimus lumborum* (LL), m.*triceps trachii* (TB), m.*semimembranosus* (SM), and m.*gluteus medius* (GM) from Angus, Charolais and Kinsella Composite steers. In this study, traditional measures of meat quality were applied, including biochemical analysis of muscle, Warner-Bratzler Shear Force test, pH evaluation, water-holding capacity test and a trained sensory evaluation panel. This chapter was written and formatted for publication in the *Journal of Animal Science*.

Chapter 3 presents the second study of the thesis. Consumers were recruited to evaluate rib-eye steaks of steers from two RFI levels and three breeds (Angus, Charolais, and Kinsella Composite steers). Generalized Procrustes Analysis (GPA) and preference mapping of the data provided additional information about eating experiences from the consumer perceptive. Additionally, a survey of beef purchasing and consumption habits was conducted to illustrate the importance of product attributes for consumers. This chapter was written and formatted for publication in the journal *Meat Science*.
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Chapter 2: Effects of selection for low residual feed intake on meat quality of major muscles from Angus, Charolais, and Kinsella Composite cattle

2.1 Introduction

Increasing the feed efficiency of an animal is important to increase the profitability of beef production (Arthur et al., 2004), and also is beneficial to the environment as it conserves resources (Capper, 2011) and reduces methane emission (Fitzsimons et al., 2013). Residual feed intake (RFI) is a measure of animal feeding efficiency and is defined as the difference between an animal's actual and expected feed intake for a specific test period. Compared to the traditional measures of feed efficiency, specifically feed conversion ratio and gain to feed ratio, studies show that RFI is independent of body growth (Bezerra et al., 2013), meaning that low RFI animals consume less feed at the same body weight gain as high RFI animals and are therefore considered efficient. However, reducing the cost of feed per animal will be beneficial only if beef quality is not affected negatively by selecting for efficient cattle (Nascimento et al., 2016). The influences of RFI on meat quality are still inconclusive and this may be due to inconsistencies in results due to breeds. The response of different beef breeds to selection for efficiency may not be the same (Baker et al., 2006). Additionally, few studies have been conducted to investigate the effect of RFI on beef quality using sensory evaluation, an important measure of meat quality. Most studies conducted quality measurement only on high-value beef muscles like m.longissimus (Zorzi et al., 2013). However, muscles from different carcass positions with different functions should be studied to reveal the comprehensive influence of RFI on beef quality. Studies of tougher cuts of beef (e.g. cross rib, inside round) may achieve greater value. Therefore, the aim of this study was to investigate the effect of RFI on beef quality and palatability of

m.*longissimus lumborum* (LL), m.*triceps brachii* (TB), m.*semimembranosus* (SM), and m.*gluteus medius* (GM) from Angus, Charolais, and Kinsella Composite steers.

2.2 Materials and methods

The experiment was carried out following the guidelines of Canadian Council on Animal Care (1993). Approval for the use of human subjects in trained sensory panel was received from a Research Ethic Board at the University of Alberta following review of the study protocol for its adherence to ethical guidelines.

2.2.1 Experiment design and animals

A 3 x 2 factorial design was used to characterize the effects of breed types and RFI levels on meat quality and palatability of beef muscles from the strip loin, inside round, cross rib and top sirloin cuts. Calves from three different cattle breeds, including purebred Angus, purebred Charolais and crossbred Kinsella Composite, were born in April or May and reared at University of Alberta Kinsella cattle herd in 2014 and were uniquely identified by ear tags. Kinsella Composite crossbred steers were producing by crossing Angus, Charolais, or University of Alberta hybrid bulls with a hybrid dam line, described in detail by Jiang et al. (2012).

2.2.2 Animals management and RFI determination

Calves were weaned at approximately 190 days of age, and the average age for RFI testing was about 422 days for Angus and Charolais and about 344 days for Kinsella Composite. Steers were placed in feedlot pens by breed and individual feed intake and feeding frequency were monitored daily during the finishing period using GrowSafe feeding systems (GrowSafe Systems Inc., Airdrie, Canada) for RFI evaluation. The use of GrowSafe feeding system was described in detail by Basarab et al. (2003). The test periods were around 75 days for Kinsella Composite, approximately 72 days for Charolais, and about 66 days for Angus. Performance and ultrasound traits measurements were described by Mao et al. (2013). Body weight (BW) of animals were weighed every 2 weeks, and ultrasound measurements of backfat thickness and longissimus thoracis area were performed every 28 days during the test period. Ultrasound traits were estimated using an Aloka 500V diagnostic real-time ultrasound with a 17 cm 3.5 MHz linear array transducer (Overseas Monitor Corporation Ltd., Richmond, BC, Canada).

Metabolic BW (MWT) was calculated as midpoint BW^{0.75}, where midpoint BW was the sum of initial BW and ADG multiplied by half of the days on test (DOT). Final ultrasound backfat (FUFAT) of individual cattle at the end of test were predicated from the regression equation of ultrasound fat depth measurements on time. RFI values of individual animals were calculated as the difference between an animal's actual dry matter intake (DMI) and predicted DMI. Therefore, RFI = DMI actual – DMI predicted based on ADG and MWT and RFIf (adjusted for backfat thickness) = DMI actual – DMI predicted based on ADG, MWT and ultrasound backfat measured at the end of the test. The models used to predict expected DMI and DMI adjusted for ultrasound backfat thickness were described by Mao et al. (2013) and are presented here:

 $Y_i = \beta_0 + \beta_1 ADG_i + \beta_2 MWT_i + e_i$

 $Y_i = \beta_0 + \beta_1 ADG_i + \beta_2 MWT_i + \beta_3 FUFAT_i + e_i$

Where β_0 is the intercept; $\beta_1 \beta_2$ and β_3 are the coefficients on AGD, MWT and FUFAT, respectively; e_i is the residual.

When the RFI values for each animal were available, animals were classified as negative (or low) RFI group and positive (or high) RFI. Then twenty-four steers from each breed were selected for the subsequent experiment; twelve were negative RFI while the others were positive RFI. Following the RFI evaluation, steers were fed a finishing ration until they reached a minimum 2 mm back fat at the 12th - 13th rib site, the minimum requirement for a carcass to be eligible for the Canada A quality grades.

Steers were slaughtered by breed group from July to September in 2015, with the mean kill age of 493, 518, and 454 days for Angus, Charolais, and Kinsella Composite,

respectively. All cattle in the study were slaughtered in the federally-registered Agriculture and Agri-Food Canada research abattoir in Lacombe, Alberta. On each slaughter day, 6 steers from each RFI group were processed within a breed for a total of 12 steers per kill day. Cattle were randomly selected and were rested at the abattoir for approximately 2 hours with *ad libitum* access to water. After recording the live weight, steers were stunned with a captive bolt pistol and humanely slaughtered, exsanguinated and dressed. Animals and their carcasses were inspected by Canadian Food Inspection Agency inspectors to determine qualification to enter the human food system.

2.2.3 Carcass and objective meat quality traits determination

Average backfat thickness (AFAT), carcass rib eye area (REA), lean meat yield (LMY), and marbling scores were collected for all carcasses (n = 72) according to the Canadian beef grading system (Canada Gazette, 2007). The carcass backfat thickness, REA and marbling score were measured at the cut surface of the ribeye muscle between the 12th and 13th rib. Carcass marbling score can be classified as trace marbling of 100 to 199 (Canada A quality grade); slight marbling of 200 to 299 (Canada AA quality grade); slight marbling of 300 to 399 (Canada AAA quality grade); slightly abundant or more marbling of 400 to 499 (Canada Prime).

At 72 h post-mortem, the target muscle samples were removed from the right sides of the carcasses and fabricated into steaks for the analysis of meat quality traits as described by Girard et al. (2012) and Holdstock et al. (2014). Three readings of intramuscular temperature and pH were recorded on each muscle using a Fisher Scientific Accumet AP72 pH meter (Fisher Scientific, Mississauga, ON) equipped with an Orion Ingold electrode (Udorf, Switzerland). Mean pH and temperature values were calculated and used for statistical analysis. One steak from each muscle was weighed to determine drip loss, expressed in milligrams of water lost per gram of muscle. For the chemical composition of each steak, one hundred g of ground sample were weighed and placed in a gravity convection-drying oven at 102°C in stainless steel beakers (Model 1370 M, VWR Scientific, Mississauga, ON). After 24 h, the samples were weighed to determine moisture loss. The dried samples were pulverized (Grindomix Model GM200, Retsch Inc., Newton, PA) and analyzed for fat content according to Method 960.39 of Official Methods of Analysis (AOAC, 1995) by extraction with petroleum ether using a fat extractor (Foss Soxtec System Model 2050; Foss Analytical AB, Hoganas, Sweden). Crude protein content was determined using a Nitrogen/Protein Determinator CNS2000 (Leco Corp., St. Joseph, MI) on the fat-free samples based on the Method 992.15 of Official Methods of Analysis (AOAC, 1995).

For the measurement of sarcomere length, two g of each muscle trimmed of connective tissue and large deposits of fat were hand-minced, immersed in 20 mL of 0.02 M EGTA/0.25 M sucrose solution in a 50 mL centrifuge tube, and homogenized for 10 s at 6000 rmp (Polytron Homogenizer PT3100 and a 2 cm generator; Brinkmann Instruments Inc., Mississauga ON). A drop of sample was placed on a microscope slide and covered with a cover slip to prevent dehydration. A phase contrast microscope (Axioscope, Zeiss, West Germany) equipped with a Sony DXC 930 Color Video Camera (Sony Corporation, Japan) and Image Pro-Plus software V4.0 (Mediacybernetics, Silver Spring, MD) was used to capture 12 images of sarcomeres per steak sample. Mean sarcomere length value of each steak was calculated from the best ten images and expressed in micrometers for statistical analysis.

For the analysis of shear force and cooking loss, non-aged steaks (day 3) and aged steaks (day 13) were cut into 2.5-cm-thick slices. The steaks were weighed and grilled at approximately 210°C (Garland Grill ED30B, Condon Barr Food Equipment Ltd., Edmonton, AB). The internal temperature of each steak was monitored and recorded at 30s intervals using a spear point temperature probe (Type T copper-constantan,10 cm in length, AllTemp Sensors Inc., Edmonton, AB) placed in the geometric center of the steak. When the internal temperature reached 35.5 °C, the steaks were turned and cooked to a final temperature of 71°C (monitored with a Hewlett Packard HP34970A Data Logger, Hewlett Packard Co., Boise ID), and removed from the grill and left to cool in sealed polyethylene bags in an ice bath, before refrigeration at 2°C for 24 h. The following day, the final weight of each steak was recorded to calculate cooking loss,

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expressed in milligrams of water lost per gram of raw steak. Six 1.9-cm diameter cores were removed parallel to the muscle fiber from cooked steaks. Peak shear force of the cores was measured with a Texture Analyser (Model TA.XT plus, Texture Technologies Corp, New York) equipped with a Warner-Bratzler shear head. The crosshead speed was set at 200 mm min-1. Peak shear force values were recorded in kilograms (Texture Exponent 32 Software, Texture Technologies Corp., Hamilton, MA) and determined by averaging values of 6 cores from each steak.

2.2.4 Trained sensory panel evaluation of meat quality

One steak from each of 4 muscles was obtained and individually vacuumpackaged, and aged for 13 days at $4 \pm 1^{\circ}$ C and then stored at $-20 \pm 1^{\circ}$ C until evaluation, happening at September and October in 2015. The day before evaluation, steaks were removed from the freezer and thawed overnight in a cooler at $2 \pm 1^{\circ}$ C until 15 min prior to grilling. Initial weight of each steak was recorded. A thermocouple was inserted into the geometric center of each steak (AMSA, 2016). Steaks were broiled on a grill to 71°C internal temperature as previously described for cooking loss measurement.

After cooking, fat and connective tissue were removed from each cooked steak, and the cooked steaks were cut into 1.3 cm^3 cubes and served to an 11-member trained sensory panel screened and trained based on AMSA sensory guidelines (AMSA, 2016). Panelists evaluated each sample for initial tenderness (rated on the first bite through the cut center surface with the incisors); initial juiciness (rated after 3–5 chews with the molars); beef flavor intensity and desirability, off-flavor intensity and amount of connective tissue (rated between 10 and 20 chews); sustainable juiciness, overall tenderness, and overall palatability (rated prior to expectoration). Panel scores were based on a 9-point descriptive scale (9 = extremely tender, extremely juicy, extremely intense and desirable beef flavor, no intense off-flavor, no perceptible connective tissue, extremely palatable; 1 = extremely tough, extremely dry, extremely bland and undesirable beef flavor, extremely intense off-flavor, extremely abundant connective tissue, extremely unpalatable) (Holdstock et al., 2014). Samples from 6 treatments groups

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were labeled with random 3-digit codes to avoid bias and evaluated at each panel session. Six samples of the same muscle from the same breed were evaluated by trained panel in each session and all samples were evaluated in 48 sessions. All testing was performed in well-ventilated, partitioned booths under 124 lx red lighting. Distilled water and unsalted soda crackers were provided to cleanse the palate between samples.

2.2.5 Data analysis

Effect of breed and RFI levels on most meat quality parameters and sensory characteristics were evaluated by two-way analysis of variance (ANOVA) using statistical analysis software R (Version 3.3.3). Slaughter group on each slaughter day was included as a random source of variation, but it was removed when it did not account for significant variation. The cooking loss and shear force of different muscles were compared at two different aging times, thus three-way analysis of variance (ANOVA) was applied. Least square means of each treatment were calculated and compared using the Tukey's Honestly Significant Difference test at the significance level of 0.05. When interactions of main effects were significant, only the least square means of the interaction were presented. Correlation analysis was performed to calculate correlation coefficients and determine linear relationships between independent variables using R (Version 3.3.3) with the package Hmisc (Version 4.0-2). Bonferroni correction was used to compensate for the likelihood of Type I error. Twenty-nine comparisons were made; thus, the significant value of correlations was 0.0017 calculated by 0.05/29 (Mahmood et al., 2016).

2.3 Results

Performance traits and carcass traits of Angus, Charolais and Kinsella Composite cattle with positive and negative RFI values were recorded and presented in Table 2-1. No significant difference in mean ADG was observed between low and high RFI animals. However, steers with low RFI had a significant lower DMI than high RFI steers, as expected (P < 0.05). There were no significant differences in any carcass traits between

different RFI groups. The results of this study indicated that Angus consumed significantly more feed (P < 0.0001) while they had similar ADG to the other two breeds. There was a trend for RFIf of different breeds to be different (P = 0.0890). Significant differences were observed among breeds for all the carcass traits assessed. Charolais had the highest mean HCW, while Kinsella Composite had the lowest mean HCW. Angus and Kinsella Composite carcasses had thicker back fat (P < 0.0001) but lesser REA (P < 0.0001) and lower LMY than Charolais (P < 0.0001). However, Angus steers had a higher marbling score than the other two breeds (P = 0.0029).

Pearson correlation coefficients of performance triaits (Table 2-2) indicated that RFIf values adjusted for ultrasound backfat were higly positively correlated with RFI values (r = 0.9952, P < 0.0001). Both RFI values were unrelated to ADG (P > 0.05) but positively related with DMI (P < 0.0001). ADG of animals was not correlated (P > 0.05) with HCW, AFAT, REA, LMY and marbling but positively related to DMI (P = 0.0006). HCW was positively associated with LMY (P = 0.0001), while both triaits were hilgtly correlated with AFAT (P < 0.001) and REA (P < 0.0001). Marbling scores were not correlated (P > 0.05) with any of the performance triats assessed.

The significance of the two main effects, specifically breed and RFI, on meat quality traits and sensory attributes of LL, GM, SM and TB steaks is shown in Tables 3 and 4, respectively. Overall, breed significantly affected most objective meat quality and sensory quality attributes studied. However, no significant differences (P > 0.05) were obtained between two RFI groups for any of the meat quality traits and sensory characteristics in LL, GM and SM muscles. The only objective meat quality measurement affected by RFI was pH value of TB steaks, where mean value of low RFI animals was significantly greater (P < 0.05) than high RFI animals. Selection for efficient animals negatively affected the flavor desirability (P < 0.05) and intensity (P < 0.05) and sustainable juiciness (P < 0.05) in the TB muscle.

Mean pH of muscle LL and mean temperature of muscle GM and SM were not affected (P > 0.05) by breed type effect. Mean muscle pH of both TB and GM were

significantly greater (P < 0.0001) in Kinsella Composite than the other two breed types, while pH of SM from Charolais and Kinsella Composite were similar. Charolais breed cattle were observed to have the lowest mean LL and TB muscle temperature (P < 0.05) compared with Angus and Kinsella Composite cattle. No significant interactions (P > 0.05) between RFI and breed were observed for the four muscles on both traits.

The results of proximate analysis of four muscles are presented in Table 2-3. LL, GM and SM steaks from Angus had the highest fat content, while steaks from Charolais and Kinsella Composite had the lowest fat percentage (P < 0.05). For TB muscles, the proximate analysis showed no significant difference (P > 0.05) among breed types in the percentage of moisture, fat, and protein. However, there was a tendency (P = 0.1117) for the fat percentage of TB muscles from Angus and Kinsella Composite to be higher than those from Charolais. No difference of mean protein percentage among breed types was detected in SM steaks. LL and GM steaks from Charolais had the highest moisture and protein percentage, while steaks from Angus steaks had the lowest percentage (P < 0.05).

Data for drip loss and cooking loss of four muscles are shown in Table 2-3. Drip loss was greater (P < 0.05) for steaks from the SM and TB muscles of Angus than Charolais and Kinsella Composite. However, no influences of breed effect on drip loss were detected for LL and GM muscles. For steaks from all four muscles, no significant influences (P > 0.05) associated with breed, RFI, aging effects or their interactions on cooking loss were observed.

The results of sarcomere length and shear force affected by breed, RFI and aging are shown in Table 2-3. Sarcomere length of LL, SM, and TB steaks were not affected by breed. GM steaks from Kinsella Composite were observed with shorter sarcomere length (P < 0.05) than steaks from Angus or Charolais. For all four muscles, shear force values were lower as the aging time increased (P < 0.05), where the values of LL showed a greater decrease during the aging period. Steaks from LL and GM muscles of Angus had lower shear force values (P < 0.05) than Kinsella Composite, while steaks from SM had similar shear force values among the three breeds (P > 0.05). For the TB, there was a two-way interaction between aging and breed (Fig. 2-1), in which shear force values of the three breed types were lower on day 13 (P < 0.05); steaks from Kinsella Composite showed the greatest decrease in shear force value during aging.

The results of trained panel evaluation of steaks from the four muscles are presented in Table 2-4. No significant differences (p > 0.05) were observed between steaks from the two RFI groups for most of the sensory descriptors evaluated by the trained panel; however, sensory attributes were influenced by a breed effect. In LL steaks, the breed effect was significant only for flavor intensity (P < 0.001) and sustainable juiciness (p < 0.01). For both attributes, Charolais were rated with the lowest score and significantly different from Angus, which had the highest score among the breeds. For both TB and SM muscles, the breed had a similar influence on all attributes with the exception of flavor intensity. TB steaks from different breeds significantly differed from each other in flavor intensity (p < 0.001), while SM steaks did not (p > 0.1). The initial tenderness, flavor desirability, connective tissue amount, sustainable juiciness, and overall palatability of both TB and SM steaks from Charolais had the lowest scores, while Kinsella Composite had the highest scores among studied breeds. For GM muscle, the texture attributes (including initial and overall tenderness, and connective tissue amount) of different breeds were perceived with similar value (P > 0.05), while the beef flavor intensity and desirability (P < 0.001), juiciness (P < 0.05) and overall palatability (P < 0.05) were not. Trained panelists evaluated GM steaks from Angus and Kinsella Composite as juicier, and more desirable than steaks from Charolais carcasses. Moreover, Kinsella Composite GM steaks were rated as having more intense beef flavor and less off-flavor intensity than Angus and Charolais GM steaks. There was no difference in initial juiciness and off-flavor intensity in either of the two muscles. There was no difference in initial juiciness and off-flavor intensity in either of the two muscles.

Mean overall tenderness of TB and SM steaks were both involved in significant two-way interactions (P < 0.05) with breed types and RFI (Fig. 2-2). For both cooked TB and SM steaks, the overall tenderness was perceived as similar between RFI levels for the same breed type and also similar among three breeds for low RFI animals; however,

steaks from Kinsella Composite had the highest value of overall tenderness among three breeds when from high RFI level animals.

Pearson correlation coefficients of tenderness related quality traits (sarcomere length, shear force value, initial and overall tenderness and connective tissue amount) and overall palatability are shown in Table 2-5. Results indicated that sarcomere lengths of all four muscles were not correlated with any sensory tenderness traits or overall palatability (P > 0.1), while GM sarcomere were positively associated with 3-day and 13-day shear force value (P < 0.0174). Shear force value of both aged and unaged LL steaks were found negatively correlated with overall tenderness of samples (P < 0.001) and shear force values of LL steaks aged for 3 days were negatively related to initial tenderness (P = 0.0008). Connective tissue amount scores of steaks from all four muscles were positive correlated with both initial and overall tenderness (P < 0.0001) perceived by the trained panel. Both initial and overall tenderness of beef steaks from four muscles were positively related to overall palatability (P < 0.0001).

Correlation results of flavor related traits (marbling, fat, flavor desirability, flavor intensity, and off flavor intensity), juiciness related traits (pH, temperature, moisture, protein, drip loss, cooking loss, initial and sustainable juiciness) and overall palatability are presented in Table 2-6. Marbling scores were positively correlated with fat content (P < 0.0001), but negatively correlated with moisture content (P < 0.0001) for all muscles. Marbling scores were not correlated with any sensory flavor attributes of TB, SM and GM steaks (P > 0.1), while there was a trend that marbling scores were positively correlated with flavor desirability (P = 0.0093) and overall palatability of LL steaks (P = 0.0086). Muscle pH, temperature and protein content were not correlated with any sensory juiciness and flavor attributes (P > 0.1), except that temperatures of TB samples were positively correlated with flavor intensity (P < 0.0001) and pH of SM were negatively related to drip loss (P = 0.0002). Moisture and fat contents of LL and GM were correlated with sustainable juiciness (P < 0.002) and LL moisture was also correlated with flavor desirability (P = 0.0009) and intensity (P = 0.0003). Fat content of LL tended to be positively correlated with flavor intensity (P = 0.0023) and desirability

(P = 0.0041). Flavor desirability and intensity, and sustainable juiciness of all four muscles were highly positively correlated with overall palatability of steaks (P < 0.0001). Moreover, off-flavor intensity scores of steaks were also found to positively correlate with overall palatability of LL, TB and SM (P < 0.0001).

2.4 Discussion

The results of the current study agree with previous reports (Baker et al., 2006; Nkrumah et al., 2007; Ahola et al., 2011; Zorzi et al., 2013) on the relationship between RFI and ADG, and DMI. Low RFI animals consumed less dry matter to achieve similar carcass weight gain performance compared to high RFI animals, indicating that RFI is independent of growth and body size (Koch et al., 1963) in Angus, Charolais, and Kinsella Composite steers, and selection against RFI shows its potential to improve the overall profit of the beef industry, as feed costs are the largest input of beef operation system (Council and Network, Western Canadian Feed Innovation, 2011). Back fat thickness, rib eye area, hot carcass weight and marbling score are good predictors of retail product yield (Crouse and Dikeman, 1976; Tait Jr, 2002). In the current study, no influence of RFI on these factors was found, suggesting that it is possible to select animals from studied breeds (Angus, Charolais and Kinsella Composite) that consume less feed without compromising carcass product yield. However, the inconsistent results reported in the literature on the relationship between RFI and carcass composition may suggest the variable influence of RFI on different purebred or crossbred cattle (Baker et al., 2006). Studies conducted on purebred Angus steers by Baker et al. (2006), Nellore bulls by Zorzi et al. (2013), and Angus bulls by Ahola et al. (2011) agree with the current study that there was no influence of selecting low RFI animals on the back fat thickness and rib eye area. Our results on the lean meat yield are consistent with the study of Basarab et al. (2003) that LMY of different groups of RFI were similar. In contrast to our results, Ahola et al. (2011) found that efficient animals had reduced marbling; Richardson et al. (2001) reported decreased back fat thickness and intramuscular fat content in low RFI Angus cattle; and Basarab et al. (2003) found that low RFI steers tended to have less intramuscular fat than high RFI steers. The variable findings

of these studies indicate the possible negative influence of selecting low RFI animals in some populations; however, the cause of the inconsistency is not clear. Thus, additional studies on understanding the effects of RFI selection on carcass traits should be conducted.

Previous studies indicate that the accumulation hydrogen ions during the production of lactic acid (Zorzi et al., 2013) during the postmortem period results in lowered pH and increased denaturation of muscle protein (Bruce and Ball, 1990), resulting in decreased water holding capacity and increased drip and cooking loss of muscles (Bruce et al., 2004), which could affect the juiciness of beef steaks (Chambaz et al., 2003). High muscle temperature could accelerate this process, as high muscle temperature may promote glycolytic enzyme activity (Bruce and Ball, 1990). Since no significant difference between RFI groups was observed on carcass temperature and pH for all four muscles, no significant influence of selection for low RFI cattle on cooking and drip loss, initial and sustainable juiciness was expected. These results are consistent with the studies by others researchers (Baker et al., 2006; Ahola et al., 2011; Nascimento et al., 2016). Ahola et al. (2011) found no significant difference of water percentage, protein percentage, cooking loss and juiciness across different RFI Angus steers, and Baker et al. (2006) reported a slight tendency (not statistically significant) that protein and moisture of m. longissimus dorsi (LD) of Angus steers were affected by RFI selection. However, Baker et al. (2006) found a higher cooking loss, and higher juiciness of strip loin steaks from low RFI steers, which differs from the current study. These controversial results suggest there might be an influence of selection for RFI on meat quality and that more research is needed to assess this influence. In the current study, although there was no influence of RFI on any objective quality trait of steaks from four muscles, a negative influence of RFI on eating quality of steaks from TB muscles was found. The reason why flavor and juiciness of TB muscle were affected by RFI is still unknown, although a reduced muscle pH has been associated with increased flavor desirability (Yancey et al., 2005). The ultimate pH of the high RFI beef was indeed lower than that of low RFI beef (0.02 pH units), so a biological impact on flavor seems unlikely, but the lower pH in high RFI beef may be indicative of additional metabolites

related to anaerobic glycolysis such as adenosine monophosphate and ribose, which may contribute to enhanced meat flavor.

Different degrees of influence of breed effect on water-holding capacity were observed among the muscles in the current study. Most studies have reported clear breed type differences in cooking and drip loss or juiciness (Shackelford et al., 1995; Chambaz et al., 2003; Bures et al., 2006). In the present study, however, cooking loss of all the four muscles, drip loss of muscle LL and GM, and initial juiciness of LL, SM, and TB steaks were similar across breed groups. This contrasted with study conducted by Chambaz et al. (2003), who found clear differences in cooking and drip loss and juiciness of LD muscle from Angus and Charolais steers. Despite the slight difference in cooking loss of four muscles, the overall juiciness of steaks from these muscles were quite different among breed type. Higher juiciness of Angus than Charolais was reported by Bures et al. (2006) but was in contrast with the study of Chambaz et al. (2003), who indicated that steaks from Charolais were juicier than those from Angus. A possible explanation for the different results may be the smaller slice size of Angus steaks, which had a higher cooking loss, used by Chambaz et al. (2003). These results indicate that the breed differences in drip loss were greater than those in cooking loss and that different muscle from the same breed presented different water-holding capacity. Moreover, that postmortem aging of muscles did not affect cooking loss regardless of breed was also reported by Holdstock et al. (2014), but was in contrast with the previous studies carried out by Bruce et al. (2004) and Nascimento et al. (2016), both of whom found that cooking loss increased during aging. A possible reason for this variation in results is the shorter aging period in the current study.

The moisture, protein, and fat percentage of muscles, especially the *longissimus* muscle, are important factors that contribute to meat characteristics and quality (Zorzi et al., 2013). Water is the major component of meat and is related to carcass yield and palatability of cooked beef (Faustman et al., 1998). Intramuscular fat of *longissimus* muscle is important for meat quality, as it reflects the degree of marbling (Welch et al., 2012), a key component of the grading system and also crucial to desired meat

palatability attributes such as flavor, juiciness, and aroma (Zorzi et al., 2013). Selection for RFI did not affect the chemical composition of the four muscles from different breeds, which is consistent with previous studies (Reis et al., 2010; Welch et al., 2012; Zorzi et al., 2013). However, most previous studies found that selection for efficient cattle reduced extracted intramuscular fat content (Basarab et al., 2003; Nascimento et al., 2016), thus affecting meat quality negatively.

The results of chemical composition of the four muscles revealed that the intramuscular fat percentage of muscles from Angus was higher than that from Charolais, which is in agreement with other authors (Wheeler et al., 2005; Bartoň et al., 2006; Bures et al., 2006). However, mean protein and moisture content of LL and GM from Angus were the lowest among the breeds, indicating that increased fat content is related to reduced protein percentage (Bures et al., 2006) and thus lowered moisture content (Gregory et al., 1994; Baker et al., 2006). Bures et al. (2006) reported that increased juiciness of beef steaks sometimes is related with increased fat content. Data of the current study also showed a positive correlation between sustainable juiciness and fat content of steaks from LL muscle.

Tenderness is one of the most important factors affecting consumers' decision to purchase a beef product. Considerable studies have shown that consumers are willing to pay more for "guaranteed tender" steak (Dransfield, 1998; Lusk et al., 2001), indicating the relationship between the commercial value of a beef cut and its tenderness. The two most commonly used methods to quantify meat tenderness are Warner-Bratzler shear force analysis and trained panel sensory analysis. The sensory score of tenderness and WBSF analysis revealed that the toughness of the four muscle steaks was similar between RFI groups, indicating no influence of selection for RFI on tenderness of beef steaks. Similar to our results, McDonagh et al. (2001), Baker et al. (2006) and Ahola et al. (2011) also reported no difference in tenderness (as measured by WBSF and trained panel analysis) between RFI groups. The present study showed that the initial and overall tenderness of samples from muscle LL and GM were judged as similar among breeds. Chambaz et al. (2003) found similar values of sensory tenderness for cooked samples from Angus and Charolais muscles. However, WBSF of LL and GM samples were significantly different among breeds. Although the results of the current study are different from Bures et al. (2006), who reported that the steaks of muscle LL from Angus were perceived as more tender than those from Charolais, both studies reflected the poor relationship between subjective and objective tenderness measurement as confirmed by poor correlation between shear force values and initial and overall tenderness in this study. As tenderness is a complex attribute, such a disagreement between tenderness determined by WBSF analysis and that evaluated by trained panel is commonly observed (Calkins and Sullivan, 2007). The significant decrease of shear force value of four muscles during aging indicated that toughness decreased during post-mortem aging. Similar results were also found by Bruce et al. (2004) and Zorzi et al. (2013), who explained that there would be an effect of proteolytic enzymes or ionic solubilisation on the strength of the myofibrillar protein structure. The large decrease in shear force values of LL steaks during aging reinforces the importance of this process in improving meat quality of this high-value beef muscle.

Tenderness is influenced by several ante-mortem and post-mortem factors, including sarcomere length, connective tissue amount and maturity, muscle chemical composition, aging and so on. Our results showed that sarcomere length of muscle TB and SM were similar among breeds, while tenderness of steaks from these two muscles were perceived with significant difference among breeds. The relationship between meat sarcomere length and tenderness is not clear (Weaver et al., 2008). Some studies even reported a poor relationship between sarcomere length and tenderness (King et al., 2003). Consistent with a previous study (Zorzi et al., 2013), no influence of RFI on sarcomere length was observed. Previous studies (Keith et al., 1985; Chambaz et al., 2003) indicated that the amount, structure and composition of connective tissue in the muscle contribute to the toughness of meat. Thus, the results of the current study that perceived connective tissue was highly correlated with initial and overall tenderness evaluated by panelists were expected.

2.5 Conclusion

The results of this study suggested that the effect of breed on beef carcass and meat sensory quality was greater than selection for RFI. The interaction between breed and RFI was not significant in most of the beef quality traits studied. Variability in carcass traits and meat quality of different muscles from different beef breeds were observed. Generally, Angus and Kinsella Composite were of "better" meat quality and sensory quality, while Charolais had better yield quality. As expected, the results of this study confirmed that increasing the aging period of the beef carcass would reduce peak shear force values of meat. However, meat quality and palatability of all breeds between two different residual feed intake levels were similar for most muscles studied, except the flavor and juiciness of TB. Thus, selection for low RFI animals may be a beneficial tool to reduce feed costs or reduce environmental impact, without compromising meat quality and palatability of most muscles, especially for high-value beef cuts.

2.6 Tables and figures

		Breed ¹			I	RFI		P-v	alue ²
	Ang	Cha	KC	SEM	Low	High	SEM	Breed	RFI
ADG, kg	1.71	1.65	1.56	0.06	1.62	1.66	0.05	0.1837	0.5989
DMI, kg/d	12.15 ^a	11.10 ^b	10.70 ^b	0.18	10.36 ^y	12.27 ^x	0.15	9.45e-07	1.76e-13
RFI ³ , kg/d	0.24	0.01	0.05	0.09	-0.82 ^y	1.01 ^x	0.07	0.1429	<2e-16
RFIf ⁴ , kg/d	0.24	0.01	-0.02	0.09	-0.83 ^y	0.98 ^x	0.07	0.0890	<2e-16
HCW, kg	753.88 ^b	841.54 ^a	666.00 ^c	11.53	755.45	752.16	9.42	1.81e-15	0.8056
AFAT ⁵ , cm	11.61ª	7.21 ^b	10.63ª	0.52	9.85	9.78	0.42	1.428e-07	0.9020
REA^6 , cm^2	74.78 ^b	93.38ª	72.67 ^b	1.67	79.91	79.97	1.38	7.07e-14	0.9744
LMY ⁷ , %	55.67 ^b	61.45 ^a	56.80 ^b	0.53	57.92	58.03	0.43	7.914e-11	0.8639
Marbling ⁸	429.38 ^a	387.91 ^b	374.17 ^b	11.35	392.92	401.39	9.3	0.0029	0.5205

Table 2-1 Effect of breed and RFI on the beef growth and carcass performances.

^{a-c} Least square means with different letters within a row differ (P < 0.05) among breeds

^{x, y} Least square means with different letters within a row differ (P < 0.05) due to RFI

¹Breed: Ang = Angus; Cha = Charolais; KC = Kinsella Composite

²P-value: significance level of main effects for Breed, RFI

 ${}^{3}RFI = residual feed intake$

 4 RFIf = residual feed intake adjusted for back fat thickness assessed at the end of the test

 5 AFAT = average backfat thickness

 6 REA = carcass rib eye area

 $^{7}LMY = lean meat yield$

⁸Marbling: 100-199 = trace marbling (Canada A grade); 200-299 = slight marbling (Canada AA grade); 300-399 = small to moderate marbling (Canada AAA grade); 400-499 = slightly abundant or more marbling (Canada Prime)

	ADG	DMI	RFI	RFIf	HCW	AFAT	CREA	LMY	Marbling
ADG	1.00	0.40*	-0.01	-0.02	0.27	-0.13	0.18	0.12	0.06
DMI		1.00	0.76*	0.76*	0.36	0.04	0.07	-0.09	0.22
Residual feed intake (RFI)			1.00	1.00*	0.00	0.03	-0.04	-0.05	0.13
RFI adjusted for backfat (RFIf)				1.00	0.02	0.02	-0.02	-0.03	0.13
HCW					1.00	-0.39*	0.73*	0.44*	0.07
Average backfat thickness (AFAT)						1.00	-0.56*	-0.89*	0.10
Carcass rib eye area (REA)							1.00	0.83*	-0.02
Lean meat yield (LMY)								1.00	-0.10
Marbling									1.00

Table 2-2 Pearson's correlations between main production and carcass measurements.

*Pearson's correlations are significant based on p < 0.0017 corrected by Bonferroni correction

Table 2-3 Effect of breed and RFI on the objective meat quality of beef steaks from m.*longissimus lumborum*, m.*tricep brachii*, m.*semimembranosus*, and m.*gluteus medius*.

		Breed ¹			R	FI		Days c	of aging			P-value ²		
	Ang	Cha	KC	SEM	Low	High	SEM	Day3	Day13	SEM	Breed	RFI	Aging	$B \times A^3$
					m.long	gissimı	ıs lum	borum						
pН	5.50	5.54	5.52	0.01	5.52	5.52	0.01	n/a	n/a	n/a	0.0792	0.9652	n/a	n/a
Temperature, °C	2.81ª	2.34 ^b	2.66ª	0.07	2.59	2.62	0.07	n/a	n/a	n/a	0.0070	0.7747	n/a	n/a
Fat, %	4.73 ^a	2.95 ^b	3.73 ^b	0.29	3.97	3.64	0.24	n/a	n/a	n/a	0.0003	0.3262	n/a	n/a
Protein, %	20.75 ^b	21.23ª	21.36 ^a	0.12	21.12	21.10	0.10	n/a	n/a	n/a	0.0024	0.8873	n/a	n/a
Moisture, %	72.02 ^b	73.42ª	72.60 ^b	0.23	72.56	72.80	0.19	n/a	n/a	n/a	0.0003	0.3842	n/a	n/a
Drip loss, mg/g	43.95	40.35	42.50	2.58	42.28	42.26	2.08	n/a	n/a	n/a	0.6238	0.9936	n/a	n/a
Sarcomere length, µm	1.74	1.72	1.73	0.03	1.75	1.72	0.02	n/a	n/a	n/a	0.8424	0.3780	n/a	n/a
Cooking loss, mg/g	203.92	220.65	209.66	5.86	211.98	210.83	4.78	207.53	215.28	4.78	0.1238	0.8646	0.2539	0.4862
Shear force, kg	3.80 ^b	4.01 ^{ab}	4.45ª	0.23	4.13	4.04	0.13	4.96 ^A	3.22 ^B	0.13	0.0196	0.6351	< 0.0001	0.1176
					m	. <i>tricep</i>	brach	ii						
pН	5.53 ^b	5.56 ^b	5.59ª	0.01	5.57 ^x	5.55 ^y	0.01	n/a	n/a	n/a	< 0.0001	0.0089	n/a	n/a
Temperature, °C	2.56ª	2.25 ^b	2.48ª	0.15	2.44	2.42	0.15	n/a	n/a	n/a	0.0008	0.8028	n/a	n/a
Fat, %	3.49	2.98	3.45	0.19	3.36	3.26	0.15	n/a	n/a	n/a	0.1117	0.6691	n/a	n/a
Protein, %	19.86	20.13	20.16	0.38	19.98	20.11	0.37	n/a	n/a	n/a	0.2957	0.4571	n/a	n/a
Moisture, %	74.31	74.35	74.47	0.16	74.37	74.38	0.13	n/a	n/a	n/a	0.7506	0.9363	n/a	n/a
Dri ploss, mg/g	38.68ª	30.32 ^b	31.05 ^b	1.56	33.96	32.74	1.26	n/a	n/a	n/a	0.0022	0.4824	n/a	n/a
Sarcomere length, µm	2.19	2.21	2.20	0.04	2.18	2.22	0.03	n/a	n/a	n/a	0.9147	0.3971	n/a	n/a
Cooking loss, mg/g	238.98	242.37	226.17	6.89	236.60	235.08	5.62	231.22	240.46	5.66	0.2228	0.8498	0.2486	0.6641
Shear force, kg	3.44	3.55	3.40	0.08	3.46	3.47	0.06	3.92 ^A	3.00 ^B	0.06	0.3713	0.9254	< 0.0001	0.0410
					m.se	emimen	nbran	osus						
pН	5.45 ^b	5.47ª	5.49ª	0.01	5.47	5.48	0.01	n/a	n/a	n/a	0.0177	0.3429	n/a	n/a
Temperature, °C	2.82	2.48	2.55	0.10	2.58	2.65	0.09	n/a	n/a	n/a	0.0616	0.5381	n/a	n/a
Fat, %	3.48 ^a	2.76 ^b	2.76 ^b	0.19	3.21	2.79	0.15	n/a	n/a	n/a	0.0129	0.0600	n/a	n/a
Protein, %	21.12	21.32	21.51	0.16	21.21	21.43	0.13	n/a	n/a	n/a	0.2433	0.2567	n/a	n/a
Moisture, %	72.70 ^b	73.30ª	73.14 ^{ab}	0.16	72.91	73.19	0.13	n/a	n/a	n/a	0.0233	0.1263	n/a	n/a
Dri ploss, mg/g	49.80ª	43.51 ^b	40.56 ^b	1.60	44.94	44.31	1.29	n/a	n/a	n/a	0.0016	0.7216	n/a	n/a
Sarcomere length, µm	1.75	1.76	1.80	0.03	1.76	1.78	0.02	n/a	n/a	n/a	0.3791	0.5163	n/a	n/a
Cooking loss, mg/g	264.94	283.35	276.44	6.67	270.99	278.83	5.45	276.86	272.96	5.45	0.1452	0.3111	0.6142	0.4984
Shear force, kg	3.47	3.71	3.67	0.10	3.60	3.63	0.08	3.89 ^A	3.35 ^B	0.08	0.2087	0.8257	< 0.0001	0.6059

					m	.gluteu	s med	ius						
pН	5.45 ^b	5.47 ^b	5.51ª	0.01	5.48	5.47	0.02	n/a	n/a	n/a	< 0.0001	0.5498	n/a	n/a
Temperature, °C	2.83	2.53	2.52	0.10	2.64	2.61	0.08	n/a	n/a	n/a	0.0471	0.7913	n/a	n/a
Fat, %	3.80 ^a	2.45 ^b	2.97 ^b	0.20	3.19	2.96	0.17	n/a	n/a	n/a	< 0.0001	0.3215	n/a	n/a
Protein, %	20.36 ^b	20.85ª	20.88ª	0.13	20.78	20.62	0.10	n/a	n/a	n/a	0.0068	0.2616	n/a	n/a
Moisture, %	72.65 ^b	73.75ª	73.24ª	0.17	73.11	73.32	0.14	n/a	n/a	n/a	< 0.0001	0.2612	n/a	n/a
Drip loss, mg/g	42.98	42.16	39.66	1.69	41.88	41.32	1.37	n/a	n/a	n/a	0.3411	0.7678	n/a	n/a
Sarcomere length, µm	1.72 ^a	1.67 ^a	1.54 ^b	0.03	1.63	1.66	0.03	n/a	n/a	n/a	0.0008	0.4341	n/a	n/a
Cooking loss, mg/g	241.63	260.74	262.41	6.96	260.37	249.48	5.68	253.54	256.34	5.66	0.0685	0.1775	0.7248	0.2264
Shear force, kg	2.92 ^b	3.05 ^b	3.74ª	0.15	3.24	3.23	0.09	3.56 ^A	2.91 ^B	0.09	< 0.0001	0.9596	< 0.0001	0.4078

^{a-c} Least square means with different letters within a row differ (P < 0.05) due to breeds.

^{x, y} Least square means with different letters within a row differ (P < 0.05) due to RFI.

^{A, B} Least square means with different letters within a row differ (P < 0.05) due to Aging.

¹Breed: Ang = Angus; Cha = Charolais; KC = Kinsella Composite.

²P-value: significance level of main effects for Breed, RFI, Aging, and interaction between Breed and Aging

 $^{3}B\times A$: interaction between breed and aging.

Table 2-4 Effect of breed and RFI on the trained panel sensory attributes of beef steaks from m.*longissimus lumborum*, m.*tricep brachii*, m.*semimembranosus*, and m.*gluteus medius*.

		Breed ¹]	RFI			P-value ²	
Sensory attributes ⁴	Ang	Cha	KC	SEM	Low	High	SEM	Breed	RFI	$\mathbf{B} \times \mathbf{R}^3$
			m.loi	ngissim	us lum	borum				
Initial tenderness	6.3	6.0	6.2	0.16	6.1	6.1	0.13	0.3694	0.9421	0.0517
Initial juiciness	5.7	5.5	5.4	0.65	5.5	5.5	0.67	0.2431	0.9561	0.7587
Beef flavor desirability	5.9	5.7	5.9	0.08	5.9	5.8	0.07	0.3053	0.8931	0.7129
Beef flavor intensity	5.9ª	5.6 ^b	6.0ª	0.07	5.8	5.8	0.06	0.0002	0.9778	0.8572
Off flavor intensity	8.0	8.0	7.9	0.11	8.0	7.9	0.09	0.9937	0.4470	0.4629
Connective tissue	8.2	8.1	8.0	0.06	8.1	8.1	0.05	0.0994	0.3942	0.3260
Overall tenderness	6.5	6.3	6.2	0.15	6.3	6.3	0.12	0.2893	0.8394	0.1684
Sustainable Juiciness	5.7ª	5.4 ^b	5.6 ^b	0.08	5.6	5.5	0.07	0.0057	0.9033	0.4486
Overall Palatability	5.7	5.4	5.5	0.10	5.6	5.5	0.08	0.2160	0.6376	0.5966
			1	m. <i>tricep</i>	brach	nii				
Initial tenderness	5.4 ^b	5.2 ^b	5.9ª	0.13	5.4	5.5	0.11	0.0015	0.5137	0.0547
Initial juiciness	5.8	5.5	5.3	0.14	5.5	5.6	0.12	0.0650	0.3841	0.4295
Beef flavor desirability	5.6 ^{ab}	5.4 ^b	5.9ª	0.08	5.5 ^y	5.8 ^x	0.07	0.0022	0.0215	0.2787
Beef flavor intensity	5.6 ^b	5.4 ^b	5.9ª	0.07	5.5 ^y	5.7 ^x	0.07	4.42E-06	0.0158	0.9878
Off flavor intensity	7.8	7.8	7.9	0.10	7.8	7.9	0.10	0.8400	0.3302	0.1160
Connective tissue	7.6 ^b	7.6 ^b	8.1ª	0.08	7.8	7.8	0.08	6.76E-05	0.7752	0.2258
Sustainable Juiciness	5.8ª	5.4 ^b	5.8ª	0.07	5.6 ^y	5.8 ^x	0.07	0.0003	0.0266	0.9873
Overall Palatability	5.3 ^{ab}	5.0 ^b	5.5ª	0.07	5.2	5.3	0.07	0.0015	0.0758	0.2009
			m.,	semime	mbran	osus				
Initial tenderness	3.8 ^{ab}	3.6 ^b	4.3ª	0.18	4.0	3.8	0.15	0.0164	0.3665	0.0913
Initial juiciness	4.5	4.2	4.3	0.17	4.4	4.2	0.14	0.2720	0.2612	0.3606
Beef flavor desirability	5.1 ^b	5.3 ^{ab}	5.4ª	0.07	5.3	5.2	0.06	0.0361	0.0831	0.6258
Beef flavor intensity	5.3	5.3	5.4	0.07	5.4	5.3	0.06	0.1872	0.2072	0.7801
Off flavor intensity	7.7	7.9	7.7	0.09	7.8	7.7	0.08	0.3506	0.5860	0.6897
Connective tissue	6.2 ^b	5.7 ^b	6.8ª	0.16	6.3	6.2	0.15	< 0.0001	0.6672	0.1033
Sustainable Juiciness	4.7 ^a	4.2 ^b	4.9ª	0.11	4.7	4.5	0.10	3.44E-05	0.2229	0.4280
Overall Palatability	4.0 ^b	3.7°	4.5ª	0.08	4.6	4.4	0.09	5.27E-09	0.4219	0.0790

			n	n. <i>glutei</i>	ıs med	ius				
Initial tenderness	5.4	5.4	5.7	0.16	5.6	5.4	0.13	0.3857	0.253	0.3626
Initial juiciness	5.4ª	4.7 ^b	5.2 ^{ab}	0.17	5.1	5.1	0.15	0.0139	0.9966	0.2283
Beef flavor desirability	5.5ª	5.2 ^b	5.7ª	0.08	5.5	5.5	0.07	0.0001	0.8099	0.6358
Beef flavor intensity	5.6 ^b	5.1°	5.8ª	0.06	5.5	5.5	0.07	8.89E-11	0.9633	0.3989
Off flavor intensity	7.5 ^b	7.6 ^b	8.0 ^a	0.10	7.7	7.6	0.09	0.0016	0.4839	0.6176
Connective tissue	7.6	7.7	7.6	0.10	7.7	7.6	0.08	0.6776	0.2080	0.5612
Overall tenderness	6.0	6.0	6.1	0.12	6.1	6.0	0.10	0.8708	0.2088	0.1579
Sustainable Juiciness	5.5ª	4.9 ^b	5.6 ^a	0.11	5.4	5.3	0.10	< 0.0001	0.5292	0.3601
Overall Palatability	5.0 ^{ab}	4.7 ^b	5.3ª	0.10	5.0	4.9	0.09	7.56E-05	0.3118	0.5715

^{a-c} Least square means with different letters within a row differ (P < 0.05) due to breeds

^{x, y} Least square means with different letters within a row differ (P < 0.05) due to RFI

¹Breed: Ang = Angus; Cha = Charolais; KC = Kinsella Composite

²P-value: significance level of main effects for Breed, RFI and interaction between Breed and RFI

³B×R: interaction between breed and RFI (scale of attributes)

⁴Sensory attributes are evaluated based on the 9-point descriptive scales scale (9 = extremely tender, extremely juicy, extremely intense and desirable beef flavor, no intense off-flavor, no perceptible connective tissue, extremely palatable; 1 = extremely tough, extremely dry, extremely bland and undesirable beef flavor, extremely intense off-flavor, extremely abundant connective tissue, extremely unpalatable)

Attributes	SL	3dSF	13dSF	InitialT	CT	OT	OP
1	n. <i>long</i>	gissimus	s lumbor	rum			
Sarcomere length (SL)	1.00	-0.03	-0.08	0.06	-0.03	0.05	0.10
3 days aging WBSF (3dSF)		1.00	0.44*	-0.39*	-0.36	-0.43*	-0.24
13 days aging WBSF (13dSF)			1.00	-0.35	-0.26	-0.39*	-0.19
Initial tenderness (InitialT)				1.00	0.59*	0.90*	0.57*
Connective tissue amount (CT)					1.00	0.63*	0.29
Overall tenderness (OT)						1.00	0.67*
Overall Palatability (OP)							1.00
	m	.tricep	brachii				
Sarcomere length (SL)	1.00	-0.13	-0.16	0.19	-0.11	0.19	0.19
3 days aging WBSF (3dSF)		1.00	0.17	-0.10	-0.04	-0.13	0.07
13 days aging WBSF (13dSF)			1.00	-0.23	-0.34	-0.28	-0.45*
Initial tenderness (InitialT)				1.00	0.50*	0.83*	0.44*
Connective tissue amount (CT)					1.00	0.47*	0.22
Overall tenderness (OT)						1.00	0.47*
Overall Palatability (OP)							1.00
	m.se	mimem	branosu	IS			
Sarcomere length (SL)	1.00	-0.07	-0.02	0.04	0.19	-0.01	0.09
3 days aging WBSF (3dSF)	-	1.00	0.18	-0.15	-0.04	-0.10	0.00
13 days aging WBSF (13dSF)			1.00	-0.27	-0.31	-0.22	-0.16
Initial tenderness (InitialT)				1.00	0.76*	0.92*	0.74*
Connective tissue amount (CT)					1.00	0.82*	0.71*
Overall tenderness (OT)			-			1.00	0.83*
Overall Palatability (OP)							1.00

Table 2-5 Pearson's correlations between tenderness related traits.

	m.	gluteus	medius				
Sarcomere length (SL)	1.00	-0.42*	-0.39*	-0.05	-0.01	-0.01	-0.15
3 days aging WBSF (3dSF)		1.00	0.46*	-0.20	-0.17	-0.25	-0.04
13 days aging WBSF (13dSF)			1.00	-0.20	-0.06	-0.21	0.00
Initial tenderness (InitialT)				1.00	0.56*	0.91*	0.63*
Connective tissue amount (CT)					1.00	0.58*	0.27
Overall tenderness (OT)						1.00	0.63*
Overall Palatability (OP)							1.00

*Pearson's correlations are significant based on p < 0.0017 corrected by Bonferroni correction

	Marbling	pН	Temp	Mois	Fat	Protein	Drip loss	3dCL	13dCL	IJ	FD	FI	OffF	SusJ	OP
						m. <i>lor</i>	ngissimus lu	mborum	ı						
Marbling	1.00	-0.09	0.16	-0.65*	0.68*	-0.52*	-0.13	-0.08	-0.30	0.02	0.31	0.24	0.34	0.11	0.31
pН		1.00	-0.15	0.01	-0.04	0.15	-0.21	0.13	0.05	0.00	-0.03	-0.09	-0.01	-0.10	0.00
Temp			1.00	-0.12	0.16	-0.14	0.17	-0.09	0.03	0.15	0.00	0.07	0.02	0.29	0.06
Mois				1.00	-0.96*	0.45*	0.14	0.17	0.25	-0.19	-0.38*	-0.42*	-0.21	-0.37*	-0.35
Fat					1.00	-0.63*	-0.09	-0.21	-0.25	0.21	0.35	0.33	0.24	0.37*	0.34
Protein						1.00	0.09	0.13	0.15	-0.24	-0.06	0.05	-0.13	-0.23	-0.13
Drip loss							1.00	-0.02	0.25	-0.12	-0.25	-0.22	-0.17	-0.02	-0.22
3dCL								1.00	-0.03	0.00	-0.05	-0.05	-0.06	-0.08	-0.05
13dCL									1.00	0.09	-0.15	-0.14	-0.12	0.08	-0.10
IJ										1.00	0.25	0.03	0.04	0.79*	0.33
ID											1.00	0.77*	0.71*	0.40*	0.81*
FI												1.00	0.44*	0.28	0.57*
OffF													1.00	0.18	0.70*
SusJ														1.00	0.45*
						1	n. <i>tricep bra</i>	ichii							
Marbling	1.00	-0.33	0.05	-0.47*	0.50*	-0.27	0.03	-0.12	-0.18	0.12	0.03	-0.01	0.04	0.18	0.07
pН		1.00	-0.06	0.31	-0.08	-0.08	-0.03	0.13	-0.02	-0.12	0.16	0.22	0.06	-0.01	0.19
Temp			1.00	-0.20	0.25	0.20	0.14	0.20	-0.02	-0.09	0.21	0.45*	0.21	0.11	0.22
Mois				1.00	-0.77*	-0.01	0.11	0.06	0.03	0.00	-0.02	-0.15	0.01	-0.05	-0.03
Fat					1.00	-0.37*	0.12	-0.10	0.04	0.03	0.20	0.37*	0.04	0.28	0.20
Protein						1.00	-0.25	0.24	0.12	-0.05	-0.16	-0.06	-0.06	-0.20	-0.12
Drip loss							1.00	0.09	-0.02	0.01	-0.06	-0.09	-0.11	0.09	-0.17
3dCL								1.00	0.18	-0.08	-0.03	-0.03	0.02	-0.13	0.05
13dCL									1.00	-0.02	-0.05	-0.02	0.03	-0.12	-0.21
IJ										1.00	0.11	-0.09	-0.15	0.65*	0.10
ID											1.00	0.76*	0.72*	0.32	0.85*
FI												1.00	0.52*	0.24	0.71*
OffF													1.00	-0.07	0.62*
SusJ														1.00	0.39*

Table 2-6 Pearson's correlations between juiciness and flavor related traits.
						m.	semimemb	ranosus							
Marbling	1.00	-0.21	0.05	-0.60*	0.65*	-0.41*	-0.03	-0.04	-0.15	0.15	0.15	0.24	0.08	0.15	-0.02
pН		1.00	-0.36	0.31	-0.32	0.09	-0.46*	-0.05	0.07	-0.18	-0.07	-0.08	-0.03	0.00	0.23
Temp			1.00	-0.19	0.20	0.08	0.14	-0.12	0.03	0.08	-0.08	0.03	-0.04	0.09	-0.10
Mois				1.00	-0.92*	0.32	0.05	0.04	0.03	-0.12	-0.04	-0.12	0.04	-0.24	-0.02
Fat					1.00	-0.51*	0.02	0.02	-0.08	0.14	0.09	0.17	0.04	0.20	-0.05
Protein						1.00	0.03	-0.11	0.08	-0.05	-0.23	-0.15	-0.34	-0.04	0.03
Drip loss							1.00	0.04	-0.14	0.09	-0.06	0.05	0.05	-0.09	-0.12
3dCL								1.00	0.07	-0.08	-0.14	-0.08	-0.08	-0.03	0.04
13dCL									1.00	0.01	0.06	0.17	-0.10	-0.01	-0.01
IJ										1.00	0.23	0.27	0.01	0.79*	0.38*
ID											1.00	0.75*	0.63*	0.22	0.31
FI												1.00	0.40*	0.33	0.43*
OffF													1.00	0.03	0.09
SusJ														1.00	0.71*
						r	n. <i>gluteus n</i>	nedius							
Marbling	1.00	-0.26	0.00	-0.59*	0.62*	-0.38*	-0.17	-0.20	-0.37*	0.37*	0.08	0.06	-0.20	0.23	0.10
pН		1.00	-0.14	0.23	-0.24	0.23	0.02	-0.04	0.19	-0.15	0.15	0.30	0.24	-0.14	0.08
Temp			1.00	0.06	-0.01	-0.11	0.31	-0.04	0.06	-0.08	-0.07	0.04	0.06	-0.18	-0.19
Mois				1.00	-0.93*	0.34	0.43*	0.20	0.15	-0.44*	-0.21	-0.26	0.09	-0.44*	-0.20
Fat					1.00	-0.57*	-0.32	-0.16	-0.10	0.43*	0.21	0.24	-0.10	0.39*	0.19
Protein						1.00	0.15	0.03	0.03	-0.29	-0.04	-0.03	0.22	-0.17	-0.04
Drip loss							1.00	-0.09	0.09	-0.23	-0.04	-0.02	0.05	-0.29	-0.02
3dCL								1.00	0.02	0.00	0.14	-0.01	0.23	0.02	0.16
13dCL									1.00	-0.09	0.07	0.06	0.14	-0.08	0.09
IJ										1.00	0.37*	0.30	-0.19	0.84*	0.51*
ID											1.00	0.71*	0.65*	0.47*	0.84*
FI												1.00	0.35	0.42*	0.65*
OffF													1.00	-0.06	0.44*
SusJ														1.00	0.67*

*Pearson's correlations are significant based on P < 0.0017 corrected by Bonferroni correction

Temp = Temperature; Mois = Moisture; 3dCL = 3 days aging Cookloss; 13dCL = 13 days aging Cookloss; IJ = Initial juiciness; ID = Flavor desirability; FI = Flavor intensity; OffF = Off flavor intensity; SusJ = Sustainable juiciness; OP = Overall Palatability

Figure 2-1 Shear force (kg) of beef steaks from m.tricep brachii as affected by an interaction between breed and aging period.



^{a, b}Columns with different letter are significantly different (P < 0.05). Error bars are standard error of mean (SEM)

Figure 2-2 Overall tenderness of beef steaks from m.*semimembranosus* (left) and m.*tricep brachii* (right) as affected by an interaction between breed and RFI levels.



^{a-c}Columns with different letter are significantly different (P < 0.05). Error bars are standard error of mean (SEM). Overall tenderness is evaluated based on the 9-point descriptive scales scale (9 = extremely tender; 1 = extremely tough)

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Chapter 3: Effect of residual feed intake status, breed and post mortem aging on consumer evaluation of beef ribeye steaks

3.1 Introduction

Expenses of feed account for the largest proportion of costs in cow/ calf production in Western Canada (Council and Network, Western Canadian Feed Innovation, 2011), thus increasing the feed efficiency of animals is important to increase profits from beef production. Moreover, an efficient production system saves resources and reduces environmental impacts (Capper, 2011), such as methane emission (Fitzsimons et al., 2013).

Residual feed intake (RFI), first proposed by Koch et al. (1963), is a measure of feed efficiency of animals. Animals with negative or low RFI consume less feed than the amount predicted by their growth and maintenance requirements for a specific test period (Basarab et al., 2003). Studies showed that RFI is independent of body growth traits like growth rate and body weight (Arthur and Herd, 2008; Bezerra et al., 2013), meaning that animals selected for high feed efficiency consume less feed at the same body weight and gain compared with inefficient animals.

The influence of RFI on beef growth performance has been widely studied, and RFI has proven to be a valuable method to select feed efficient animals (Kelly, 2015). However, selecting low RFI cattle to reduce cost will be beneficial only if meat quality is not adversely influenced (Nascimento et al., 2016). Although most studies have reported no influence of RFI on beef meat quality (Baker et al., 2006; Fidelis et al., 2017), the potential effect of RFI is still controversial. Baker et al. (2006) found no difference in Warner Bratzler shear force (WBSF) between RFI groups of purebred Angus steers. However, Zorzi et al. (2013) studied the meat quality of Nellore bulls and observed a tendency for reduced instrumental tenderness of low RFI animals. Undesirable influence of RFI selection was also reported by McDonagh et al. (2001); low RFI cattle had a lower

rate of myofibrillar fragmentation index and higher calpastatin concentration, causing lower rate of proteolytic activity and tougher meat than high RFI animals.

The use of different breeds may be one reason for inconsistent results among studies. Studies comparing meat quality of different beef breeds have reported clear differences in meat tenderness and other meat attributes among different breeds (Pringle et al., 1997; Campo et al., 1999; Chambaz et al., 2003; Bartoň et al., 2006; Ba et al., 2013). The response of different breeds or crossbred cattle for RFI selection may not be consistent (Baker et al., 2006), therefore it is important to compare the influence of RFI selection among breeds.

Post-mortem aging is a widely used production practice to improve meat tenderness and acceptability (Jeremiah and Gibson, 2003), with the activity of several endogenous enzymes on degradation of myofibrillar structure, protein and other chemical compounds of muscles (Lamare et al., 2002; Gorraiz et al., 2002). However, the influence of aging on flavor and juiciness of meat is not conclusive with varying results reported (Jeremiah et al., 1991; Bruce et al., 2005; Monsón et al., 2005; Brewer and Novakofski, 2008). Additionally, it is important to investigate potential interactions between RFI status and post-mortem aging to ensure no unexpected influence of RFI selection during the tenderization process of aging.

Trained panels have been used to investigate the effect of RFI on beef eating quality, while consumer acceptability or preference for steaks from low and high RFI animals has received limited investigation. Sensory attribute input from consumers and their preferences are important for the meat industry as consumers' opinions are the key factor to determine meat quality (Destefanis et al., 2008). Several studies comparing consumers and trained panelists in profiling tasks found that well instructed consumers were able to consistently discriminate differences among products as well as a trained panel (Moskowitz, 1996; Husson et al., 2001; Worch et al., 2010). Consumers involved in profiling tasks can provide their preferences and other valuable information like the ideal intensity of the products (Le and Worch, 2014).

The statistical technique of Generalized Procrustes Analysis (GPA) can be used to visualize consumer perceptions of product differences, as well as the relationship between products and sensory attributes based on panelist agreements (Ferreira et al., 2008). GPA can also minimize differences among consumer evaluations (Rodrigues and Teixeira, 2013) and determine the consensus among assessors (Hunter and Muir, 1995b). Preference mapping can be performed to illustrate the relationship between consumers' perceptions and product acceptability (Costell et al., 2000).

The objective of the present study was to determine the effects of breed (Angus, Charolais and Kinsella Composite), RFI levels (negative and high) and aging times (4 and 18 days) on consumer sensory perception and preference of rib-eye steaks.

3.2 Materials and methods

Beef production was performed following the guidelines of the Canadian Council on Animal Care (1993). Approval for the use of human subjects in consumer panels was received from a Research Ethic Board at the University of Alberta following review of the study protocol for its adherence to ethical guidelines. All participants completed written informed consent.

3.2.1 Animals and meat sample management

Calves from three different cattle breeds, purebred Angus, purebred Charolais and crossbred Kinsella Composite, were born in April or May 2014 and reared at the University of Alberta Kinsella cattle herd. Kinsella Composite population was produced by crossing between Angus, Charolais or University of Alberta hybrid bulls and a hybrid dam line. The hybrid dam line was described in detail by Jiang et al. (2012). Each animal was identified by a unique ear tag. Bull calves were castrated within 8 weeks of birth. Calves were placed with their dams in the pastures and weaned at approximately 190 days of age.

Steers were placed in a feedlot pen by breed and individual feed intake and feeding frequency were monitored using GrowSafe feeding systems (GrowSafe System Inc., Airdrie, Canada) for RFI evaluation as described in Chapter 2. Angus, Charolais and Kinsella Composite were tested for 61 days, 72 days and 75 days, respectively and weighed on 2 consecutive days at the start and end of the evaluation, as well as every 2 weeks during the test period. Steers were fed a finishing ration until they reached a minimum 2 mm back fat at the 12th - 13th rib site, the minimum requirement for a carcass eligibly for Canada A quality grades.

Equations to calculate expected dry matter intake (DMI) and RFI values were as described in Chapter 2 section 2.3.2. After RFI evaluation, twelve steers from each breed were selected for sensory experiments; six negative RFI and six positive RFI. Steers were slaughtered in the federally registered Agriculture and Agri-Food Canada research abattoir in Lacombe, Alberta from July to September in 2015. On each slaughter day, 6 cattle from each breed were randomly selected and sent to the abattoir to rest with ad libitum access to water. Steers were federally inspected prior to entering the human food system. After recording live weight, steers were stunned with a captive bolt pistol and humanely slaughtered. At 72 h post-mortem, the target muscles samples (m.*longissimus thoracis*) were removed from the right sides of the carcasses and fabricated into steaks. Two steaks from each carcass were individually vacuum-packaged, one was aged at $4 \pm 1^{\circ}$ C for 4 days while the other was aged for 18 days. Therefore, a total of 72 beef rib-eye steaks were obtained from 36 cattle for this study. After aging, all samples were frozen at $-20 \pm 1^{\circ}$ C and shipped to the University of Alberta for evaluation at September in 2016.

3.2.2 Sample preparation

Steaks were removed from the freezer and thawed at 4°C in their vacuum bags for approximately 24 h before evaluation. Before cooking, the steaks were rested at room temperature for 30 min to reach an internal temperature of approximately 18°C. The weight of packaged steaks and initial raw steaks were recorded to determine the purge loss in the package, expressed in milligrams of water lost per gram of muscle. Samples were cut to the same thickness and then cooked in a clam shell grill (Model: 169232, GE, Mississauga, Ontario, Canada) according to American Meat Science Association Guidelines (2016) with some modifications. A thermocouple (ThermoWorks, Inc. UT, USA) was placed into the geometric center of each steak to monitor the internal temperature during cooking. Steaks were turned once at approximately 45°C. After cooking, individual steaks were weighed immediately to determine cooking loss, expressed in milligrams of weight lost per gram of raw steak. Cooked steaks were placed in Corning Ware dishes with lids and stored in a warming oven at 75°C to maintain sample temperatures (AMSA, 2016). Three minutes before serving, each steak was trimmed of visible fat and connective tissue, cut into bite-sized cubes (approximate 2.54 cm \times 1.27 cm \times 1.27 cm), and two cubes were placed into a 3-digit code labeled foam container with a plastic lid and served to each participant.

3.2.3 Consumer sensory evaluation

Participants were recruited and prescreened from the University of Alberta campus to ensure they were at least 18 years old and consumed "top quality" beef steaks (i.e.: Rib-eye, Tenderloin, T-bone, and Top sirloin etc.) at least once per month (Appendix 1; 2). Consumers (n=24) were selected based on their willingness to attend 3 sensory sessions.

Participants evaluated steak samples in individual booths under red lights to mask variations in meat color (Monsón et al., 2005). An incomplete block design was used in the sensory evaluation. Each consumer tasted 4 treatments randomly selected from 12 treatment combinations (3 breeds \times 2 RFI \times 2 aging) in each of three sessions, presented in a William's square design to avoid both tasting position and carry over effects (AMSA, 2016). Each steak sample was evaluated by four participants in given session. Unsalted soda crackers and distilled water were used for palate cleansing between samples. Participants followed the same sensory evaluation procedure at the second and third session.

At the first session, each participant also completed a demographic and beef steak consumption habit questionnaire and answered questions about the importance of package and eating experience on the purchase of top quality beef steaks on a 5-point scale (5 = extremely important and 1 = not at all important) based on a previous beef consumption survey (Schroeder et al., 2006) with some modifications (Appendix 4).

Consumers rated each sample for intensity of beef flavor, tenderness and juiciness on a 9-point intensity scale (9 = extremely intense, tender, juicy, respectively and 1 = not intense at all, not tender at all, not juicy at all, respectively) and overall acceptability on a 9-point hedonic scale (9 = like extremely and 1 = dislike extremely) (AMSA, 2016). After evaluating steaks at each session, they rated their ideal intensity of beef flavor, tenderness, and juiciness of rib eye steaks on the 9-point intensity scale (Appendix 5).

3.2.4 Data analysis

Descriptive statistics were performed on data from the demographic and consumption habit questionnaire and the beef survey data using R (Version 3.3.1). Beef survey data for the two lowest and the two highest levels of importance were collapsed.

Sensory data were analyzed with R statistical software (Version 3.3.1) by threeway analysis of variance (ANOVA) with breed, RFI levels, and aging time as fixed effects; evaluation sessions and participants as random effects. The effect of the three main factors and their interactions were analyzed. Least square means of each treatment were calculated and compared using the Tukey's Honestly Significant Difference test at the significance level of 0.05.

The sensory profiles of beef steaks from twelve treatment groups and the ideal rib-eye steaks were generated in a consensus plot from Generalized Procrustes Analysis (GPA) using XLSTAT software (Trial version 2017). Finally, to investigate the relationship between consumers' preference and perception of twelve beef steaks and ideal samples, preference mapping (PREFMAP) was performed with XLSTAT software (Trial version 2017) using the coordinates of steaks samples and ideal rib-eye steaks in the consensus plot of GPA results.

3.3 Results

3.3.1 Demographic information and purchase survey

The demographic profile and beef consumption habits of the consumer panelists (n=24) are shown in Table 3-1. The number of male (54%) and female (46%) participants was about equal. Almost all participants (95.8%) had completed at least some college or university education and most (87.5%) were younger than 35 years of age. For household income, 12.5% of participants had the lowest income (i.e. less than \$15,000), while 16.7% had upper income levels (i.e. more than \$80,000). About 91.7% of participants ate beef at least once per week; many (45.8%) consumed beef 2 or 3 times per week. About 87.5% of participants reported eating top quality beef steaks at least 2 or 3 times per month. Indoor (34.5%) and outdoor grilling (31.0%) were the two primary methods of steak preparation.

The results of the survey to evaluate the importance of seventeen attributes on purchasing beef steaks are shown in Table 3-2. These 17 attributes describe package and visual information, and the steak eating experience. Most consumers regarded freshness ("packaged on" date) (70.8%), marbling (87.0%), beef grades (79.2%), product color (62.5%) and product safety assurance (65.2%) as very or extremely important factors affecting their willingness to buy steaks. The distribution of participants' opinions on the other package and visual information such as nutritional information, traceability of product to farm, labeled "organic", labeled "hormone and antibiotic free", labeled "sustainable production", varied in importance among the participants. The sensory attributes of tenderness, juiciness and flavor were chosen as very to extremely important to the steak eating experience by more than 95% of participants. Preparation ease and time were rated as less important than the sensory attributes, with 25% or less of participants rating them as very and extremely important.

3.3.2 ANOVA results

The main effects of breed, RFI and aging on water holding capacity, three sensory attributes and overall acceptance of rib-eye steaks are shown in Table 3-3. In this study, purge loss of steaks was not affected by any main factor, while cooking loss of steaks was different among breeds (P < 0.01). Rib-eye steaks from Charolais had the lowest cooking loss, followed by steaks from Kinsella Composite and Angus. Similarly, breed type significantly affected sensory juiciness intensity (P < 0.01) and overall acceptance (P < 0.05). Steaks from Charolais were significantly juicier than those from Kinsella Composite, while Angus samples were not different from either. Overall acceptance ratings were greater for rib-eye steaks from Angus than Kinsella Composite, while steaks from Charolais were similar to steaks from both breeds. There was no significant effect of any main factor on beef flavor intensity of steaks (P > 0.05). However, there was a trend for steaks aged for 18 days to have more intense beef flavor than 4-day aged steaks (P = 0.1068). The longer post-mortem aging period positively influenced overall acceptance (P = 0.0075) and tended to affect juiciness (P = 0.0832) of beef rib-eye steaks; rib-eye steaks with longer aging time (18 days) were more acceptable and tended to be juicier than steaks with shorter aging time (4 days). No RFI effect was observed on any sensory descriptor or overall acceptance by consumers. A significant breed, RFI and aging interaction (P < 0.05) was observed for the tenderness of beef steaks (Figure 3-1). No significant improvement of tenderness of steaks was observed by aging (from 4 days to 18 days) on negative RFI animals of any breed, while there was a trend for tenderness of steaks from high RFI Charolais (P< 0.05, data not shown) and Kinsella Composite cattle (P=0.0731, data not shown) to increase during aging period. Moreover, steaks from high RFI Kinsella Composite animals tended to be more tender (P = 0.0534, data not shown) than steaks from low RFI Kinsella Composite after 18 days of aging.

The comparison between consumers' ideal rib-eye steaks and tested samples is shown in Table 3-4. The intensity of the sensory attributes (beef flavor, tenderness and juiciness) of steaks from some treatments was significantly different (P < 0.01) from

consumers' ideal products. Participants rated beef flavor intensity of the samples from carcasses of Angus and 18-day aged carcasses from low RFI Charolais and Kinsella Composite as not different from their ideal intensity. All steaks aged for 18 days, except those from low RFI Kinsella Composite, were not different from the ideal tenderness. The juiciness of steaks from Kinsella composite aged for 4 days, low RFI Kinsella composite and Angus aged for 18 days were significantly different from ideal rib-eye steaks, while the juiciness of steaks from Charolais were similar to the ideal. Steaks from Kinsella Composite, except those from high RFI beef aged for 18 days, were significantly different from participants' ideal degree of both tenderness and juiciness.

3.3.3 GPA and preference mapping

Approximately 80.29% of the total variation among samples was explained by the first two main dimensions of the GPA consensus configuration (Figure 3-2), while the third dimensions explained only 19.71% of the variation. GPA results of this study can be used to interpret the relationships between samples and sensory attributes, as well as the sensory product space. In the consensus plot for rib-eye steaks, there is a tendency for beef steaks with 4 days of aging and 18 days of aging to group separately. Moreover, the first dimension was negatively loaded with most of the steaks with shorter aging time (except the Angus with high RFI steaks), but positively correlated with consumers' ideal rib-eye steaks and most of the steaks with longer aging time (except the steaks from Kinsella Composite with low RFI). Therefore, aging was responsible mainly for clustering samples in the first dimension. The correlation between sensory attributes and GPA factors shown in Figure 3-2 reveals that all sensory attributes are highly and positively correlated with the first dimension, while beef flavor intensity is also highly but negatively correlated with the second dimension. Beef rib-eye steaks aged for 18 days and ideal steaks are characterized as juicer, and more tender and with more intense beef flavor in this consensus configuration.

Following GPA analysis, preference mapping was performed to obtain additional information from consumers' preference data of beef steaks (Figure 3-3, left).

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Consumers' preferences of twelve samples and ideal steaks are presented, with each point representing a sample and each vector representing a participant's preference level. As shown in the preference map, all the participants appreciated the samples located in the positive region of the first dimension. The contour plot (Figure 3-3, right) calculates the percentage of consumers with above mean preference in a given area of the preference map (XLSTAT, 2017). The greater the proportion of consumers with high preference scores, the hotter (more red) the region color. In this case, samples in the region with the red color received the greatest number of high preference scores from consumers (XLSTAT, 2017). When the preference map and contour plot map are superimposed, the preference ranking for samples can be concluded. More than 90% of consumers rated the ideal steaks and AH18 and CL18 samples with above average scores, 80% to 90% of them did the same for AH4 steaks, and 70% to 80% for CH 18 and AL18 steaks. Fifty to sixty percent of consumers gave a higher score to KH18 steaks, while 40% to 50% did the same for CL4; 30% to 40% did for KL18; 20% to 30% for AL4; 10% to 20% of consumers gave above average scores for KH4 and CH4, and less than 10% gave to KL4. As found from the contour plot, steaks from high RFI Angus and Kinsella Composite animals aged for the same number of days (4 or 18 days) always received higher consumer acceptance scores than steaks from those animals with low RFI. However, steaks from low RFI Charolais were rated with higher overall acceptance than those from high RFI Charolais.

3.4 Discussion

The high frequency of consumption of beef products and top quality beef steaks among participants in the current study confirmed their familiarity with the eating quality of meat cuts such as rib-eye steaks and suitability for participation in the study. The importance of attributes affecting beef purchase in the present study agreed with the original survey of consumer attitudes about beef products (Schroeder et al., 2006), in which product freshness and color were among the top five determinants of beef purchase among Canadian consumers. Similarly, participants in both surveys indicated that information such as organic labels, nutritional information, product traceability, and preparation ease and preparation time were not considered important factors when purchasing beef products. However, in the current survey, most participants regarded the three sensory attributes of the steak eating experience (i.e. juiciness, flavor and tenderness) as very or extremely important factors, while Canadian consumers in the previous survey did not (Schroeder et al., 2006). This may be attributed to the focus on beef steaks in the current study, the sensory attributes of which are the most important motivators for their purchase (Verbeke and Viaene, 1999), while beef products in general were the focus of the previous survey (Schroeder et al., 2006). Thus, the evaluation of the sensory attributes (tenderness, juiciness, and beef flavor intensity) of rib-eye steaks in this study was straightforward for participants, potentially increasing the validity and reliability of their sensory judgments.

Through ANOVA results, selection of animals with high feed efficiency using RFI did not influence meat palatability and overall acceptability, as the evaluations of meat juiciness, flavor intensity and overall acceptance were similar among the two RFI groups according to the participants. Several studies investigating the association of RFI with meat sensory qualities have reported similar results. Both Baker et al. (2006) and Ahola et al. (2011) observed no significant differences in the meat tenderness and beef flavor among different RFI groups of Angus using trained panels. Baker et al. (2006) even found that the steaks from low RFI animals tended to be juicier, suggesting better sensory quality. In addition to sensory studies, some studies investigating the relationship between RFI and other meat quality parameters observed no significant effect of selection for reduced RFI among different beef breeds (Baker et al., 2006; Ahola et al., 2011; Fidelis et al., 2017). In a study investigating the effect of RFI on meat quality in Nellore cattle, Fidelis et al. (2017) observed no significant differences in shear force value and myofibrillar fragmentation index, two quality traits related with tenderness; or in final pH and cooking loss, two traits related with juiciness; or in meat color and chemical composition. Similarly, no differences in shear force values of steaks from pure Angus

breed (Baker et al., 2006; Ahola et al., 2011) and other crossbred cattle (McDonagh et al., 2001) were found.

Although the results of these studies and the present study support the statement that RFI selection of animals with improved efficiency has limited influence on meat quality assessed, the results of GPA and preference mapping in the current study suggest a larger degree of influence. More consumers in this study preferred the rib-eye steaks from carcasses of high RFI Angus or Kinsella Composite aged for same amount of time compared to the steaks from low RFI animals, indicating the possible adverse relationship between RFI selection and consumer preference in some breeds. However, given the lack of understanding of the effect of RFI selection from the consumer perspective and the limited research conducted so far, no firm conclusion can be made about the effect of RFI selection on consumer preference.

Steaks from low RFI Charolais were rated with higher acceptability by consumers than steaks from high RFI animals. Additionally, the different steak tenderness between two RFI animal groups was observed only on Kinsella Composite cattle after aging. These results agree with Baker et al. (2006) that the effect of RFI on meat quality may not be consistent across breeds. Therefore, meat quality studies should be performed in all breeds to ensure RFI selection does not influence meat quality in a specific breed. Moreover, the results of consumer studies may provide a different perspective compared to objective meat quality analysis and trained panel analysis. Multivariate analysis of consumer data can provide valuable information regarding consumer preference of beef steaks. Therefore, more consumer studies should be performed to understand the effect of RFI on consumers' meat eating experiences.

Post-mortem aging of animal carcasses at cold temperature is a well-accepted method to tenderize meat (Jiang et al., 2010). The activity of endogenous enzymes, especially the calpains (Lamare et al., 2002) in muscle during post-mortem is associated with meat tenderness (Smith et al., 1978; McDonagh et al., 2001). Both univariate and multivariate analysis of the current study confirmed the importance of aging on meat tenderness; rib-eye steaks aged 18 days were perceived by consumers as more tender than steaks aged 4 days.

The three-way interaction of breed, aging and RFI on tenderness in this study showed that post-mortem aging did not improve the perceived tenderness of steaks from any breed with low RFI, indicating the negative influence of RFI selection on tenderization of meat during the post-mortem aging period. In agreement with the current study, several studies also found an adverse influence of RFI selection. Zoizi et al. (2013) observed that the steaks from low RFI Nellore bull carcasses aged for 0, 7 and 14 days had higher shear force values than their high RFI counterparts. The study conducted by McDonagh et al. (2001) on Angus and other cross-bred cattle observed a lower index of myofibrillar fragmentation and higher concentration of calpastatin in the low RFI animals compared to high RFI animals, indicating the potential for less tender meat from low RFI animals as these two measurements are related to proteolytic activity (Zorzi et al., 2013). Apart from these objective meat quality measurements, a trained panel used by Kelly (2015) also detected the undesirable influence of selection for low RFI Angus \times Simmental steers on three meat texture attributes (softness, tenderness and rate of breakdown). Although the results of the current study and other studies suggest negative responses in meat tenderness and other meat quality traits with selection for low RFI cattle, the reason for the negative influence on tenderization of meat during post-mortem aging is not clear, due to a lack of study and knowledge of a potential genetic relationship with tenderness. Future research on the relationship between RFI selection and postmortem aging with more aging periods, as well as other typical industry practices, should be conducted to ensure no adverse influence of selection for low RFI on these techniques normally considered to improve meat quality.

The results of this study showed that post-mortem aging did not affect the juiciness and flavor intensity of rib-eye steaks. However, the effect of aging on beef flavor and juiciness is still debated as some studies have observed no effect of aging on these two attributes (Monsón et al., 2005; Brewer and Novakofski, 2008), while others have reported different results (Gorraiz et al., 2002). Monson et al. (2005) found the beef

flavor intensity did not change significantly throughout the aging time (1, 3, 7, 14, 21, and 35 days) according to a trained panel. From a consumer panel, Brewer et al. (2008) also found no effect of aging on beef flavor intensity and juiciness. However, aging theoretically affects beef flavor intensity and steak juiciness, because flavor-related chemical components of muscle, like protein, peptide composition (Spanier and Miller, 1993) and fatty acids (Gorraiz et al., 2002) are affected by post-mortem aging due to the activity of endogenous enzymes. Moreover, post-mortem aging causing the degradation of cytoskeletal proteins results in the shrinkage of muscle cells, subsequently affecting water-holding capacity of meat (Huff-Lonergan and Lonergan, 2005). A challenge to the sensory evaluation of beef flavor is the complexity of this attribute and the lack of a meaningful definition (Kerth and Miller, 2015).

Multivariate analysis of consumer sensory panel data can be a valuable tool to generate a map of sensory attributes and product coordinates, tracing the development of characteristics in meat products (Spanier and Miller, 1993). The GPA consensus plot of the current study separated two groups of animals according to days of post-mortem aging; most of the steaks aged for 18 days and ideal rib-eye steaks clustered on the positive part of first dimension, on which all sensory attributes (beef flavor intensity, juiciness, and tenderness) were highly positively associated, indicating the improvement of the rib-eye steak beef flavor intensity and juiciness throughout 18 days of aging. In agreement with the current study, the trained panel evaluation conducted by Gorraiz et al. (2002) reported that beef characteristic flavor increased during 7 days of aging.

The significant influence of breed effect on sensory quality of rib-eye steaks was observed in the current study and agrees with previous studies. The rib-eye steaks from Angus and Charolais were perceived with similar intensity of juiciness, beef flavor, tenderness and overall acceptance. Similar results were reported by Chambaz et al (2003) that meat samples from both breeds had similar shear force values, myofibrillar fragmentation index, total and soluble collagen content and sensory tenderness. Even though most studies agree upon the higher meat quality of Angus than Charolais (Sinclair et al., 2001; Bures et al., 2006), the current study suggests that differences among Angus

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and Charolais steaks, both belonging to *Bos taurus* breed, were not significant for consumers.

The main limitation of the current study is the relative small size of the consumer panel due to the limited number of steaks available. However, we recruited consumers who frequently consumed top-quality steaks, and this familiarity with the product enhanced the validity of the consumer assessments. Additionally, each consumer attended all 3 sessions to complete evaluations on samples from all treatment groups to reach a balanced experimental design. GPA was performed to reduce scale effects and obtain a census configuration among consumers; Preference Mapping based on GPA results correlated consumer preference with intensity of sensory attributes, and then provided complete information of the effect of RFI selection from the consumer perspective.

3.5 Conclusion

The results of the present study demonstrated a significant effect of breed and aging on consumers' perceptions of rib-eye steaks. No influence of RFI on meat palatability and overall preference was observed based on ANOVA, nevertheless, the GPA and Preference Mapping of the data suggested a possible adverse influence of selection for reduced RFI on consumers' preference of beef steaks, indicating the importance of multivariate analysis for studying the relationship among products, sensory attributes and consumer preference. This study supports the notion that consumers recruited from a specific product group can perform product profiling by adopting multivariate data analysis techniques such as GPA. Due to the limited influence of RFI on the sensory attributes of rib-eye steaks, RFI can be a beneficial tool to reduce feed cost and environmental impact of beef production. However, selection of feed efficient animals using RFI should be accompanied by beef palatability assessments to minimize undesired consequences on meat quality and consumer acceptance.

3.6 Tables and figures

	Characteristics	% (n)
Gender	Male	54.2 (13)
	Female	45.8 (11)
Age	18-24	20.8 (5)
-	25-34	66.7 (16)
	35-44	4.2 (1)
	45-54	8.3 (2)
Education	Some collage, university	4.2 (1)
	collage/university or graduate	37.5 (9)
	Post-graduate degree	58.3 (14)
Household income	<\$15000	12.5 (3)
(\$CAD)	\$15000-\$34999	50 (12)
	\$35000-\$59999	12.5 (3)
	\$60000-\$79999	8.3 (2)
	>\$80000	16.7 (4)
Frequency of consuming	2-3 times per month	8.3 (2)
beef	Once per week	33.3 (8)
	2-3 times per week	45.8 (11)
	4 or more times per week	12.5 (3)
Frequency of consuming	Once per month or less	12.5 (3)
top beef steak	2-3 times per month	70.8 (17)
-	Once per week	12.5 (3)
	2-3 times per week	4.2 (1)
Usual methods for	Frying	17.2 (5)
cooking beef steak*	Grilling-indoor	34.5 (10)
-	Grilling-outdoor (BBQ)	31.0 (9)
	Roasting	17.2 (5)

Table 3-1 Demographics and beef consumption habits of the consumer panelists (n=24)

*Participants checked more than one option

Table 3-2 The importance of package, visual information and steak eating experience on purchasing top quality beef steaks among the consumer panelists $(n=24)^a$

Traits	Not at all & Slight important %(n)	Moderately important %(n)	Very & Extremely important %(n)
Package and visual			
information:			
Price	20.0 (4)	50.0 (10)	30.0 (6)
Freshness (i.e. 'packaged on' date)	12.5 (3)	16.7 (4)	70.8 (17)
Steak marbling	4.3 (1)	8.7 (2)	87.0 (12)
Beef grades	0 (0)	20.8 (5)	79.2 (19)
Product color	4.2(1)	33.3 (8)	62.5 (15)
Product food safety assurance	8.7 (2)	26.1 (6)	65.2 (15)
Nutritional information	70.8 (17)	16.7 (4)	12.5 (3)
Traceability of product to farm	50 (12)	25.0 (6)	25.0 (6)
Labeled organic	65.2 (15)	17.4 (4)	17.4 (4)
Labeled hormone free	56.5 (13)	17.4 (4)	26.0 (6)
Labeled antibiotic free	43.4 (10)	26.1 (6)	30.4 (7)
Labeled sustainable production	41.6 (10)	41.7 (10)	16.7 (4)
Steak eating experience:			
Juiciness	0 (0)	4.2 (1)	95.8 (23)
Tenderness	0 (0)	4.2 (1)	95.8 (23)
Flavor	0 (0)	0 (0)	100 (23)
Preparation ease	45.8 (11)	29.2 (7)	25.0 (6)
Preparation time	54.2 (13)	29.2 (7)	16.7 (4)

^aNot all participants responded to each question

		Breed			RFI				Aging			p-Value ³					
		(n=12) ¹	(n=18)				(n=	36) ²		p-value							
	Ang	Cha	KC	SEM	High	Low	SEM	D4	D18	SEM	Breed	RFI	Aging	B×R ⁴	B×R×A ⁵		
Purge loss (mg/g)	62.1	52.0	63.4	4.9563	56.3	62.0	4.1854	58.5	59.8	4.1870	0.1944	0.3039	0.7864	0.8044	0.6553		
Cooking loss (mg/g)	194.8a	162.3b	172.8ab	9.1493	172.1	181.1	8.1289	180.7	172.5	8.1330	0.0084	0.3250	0.3909	0.1854	0.6647		
Juiciness ⁶	5.8ab	6.2a	5.5b	0.2427	5.9	5.7	0.2264	5.7	6.0	0.2264	0.0070	0.1729	0.0832	0.8839	0.3280		
Beef flavor	6.2	6.0	6.0	0.2104	6.0	6.1	0.1984	6.0	6.2	0.1984	0.3613	0.5920	0.1068	0.5411	0.7741		
Tenderness	6.1a	5.8ab	5.5b	0.2368	5.9	5.7	0.2209	5.4b	6.2a	0.2209	0.0173	0.2026	< 0.0001	0.0028	0.0126		
Overall acceptance ⁷	6.7a	6.5ab	6.1b	0.1856	6.5	6.3	0.1691	6.2b	6.7a	0.1691	0.0149	0.1904	0.0075	0.2215	0.1103		

Table 3-3 Effect of breed, RFI and aging time on water-holding capacity and sensory attributes of beef rib-eye steaks assessed by a consumer panel (n=24)

^{a-c} Least square means with different letters within a row differ (P < 0.05) among breeds

¹Breed: Ang = Angus; Cha = Charolais; KC = Kinsella Composite

²Aging: D4 = 4-day port-mortem aging; D18 = 18-day post-mortem aging

³p-Value: significant level of main effects for Breed, RFI, Aging and interaction among them

⁴B×R: p-Value of interaction between breed and RFI effects

⁵B×R×A: p-Value of interaction between breed, RFI and aging effects

⁶Juiciness: Sensory attributes of juiciness, beef flavor intensity and tenderness are evaluated based on the 9-point descriptive scales (9 extremely, 1 not at all juicy/intense/tender)

⁷Overall acceptance: Overall acceptance are evaluated based on the 9-point hedonic scales (9= like extremely to 1 = dislike extremely)

Breed (n=12)		An	gus			Cha	rolais			Kin	sella		_		
RFI	High	RFI	Low RFI		High RFI		Low RFI		High RFI		Low RFI		Ideal ¹	SEM	P-Value
Days of PM ² aging	4	18	4	18	4	18	4	18	4	18	4	18			
Beef flavor ³	6.2ab	6.4ab	6.2ab	6.1ab	5.8b	6.1b	5.9b	6.4ab	5.7b	6.0b	6.0b	6.2ab	7.2a	0.2939	0.0057
Tenderness ³	6.2abc	6.6ab	5.1cd	6.3abc	4.7d	6.3abc	5.9bcd	6.4abc	5.1cd	6.4abc	5.3bcd	5.0cd	7.4a	0.3428	< 0.0001
Juiciness ³	5.8ab	6.0ab	5.8ab	5.7b	6.2ab	6.4ab	5.9ab	6.2ab	5.1b	6.2ab	5.3b	5.3b	7.2a	0.3529	0.0001

Table 3-4 Sensory attributes intensities of beef rib-eye steaks and ideal rib-eye steaks assessed by a consumer panel (n=24)

^{a-c} Least ^{a-c} square means with different letters within a row differ (P < 0.05) among breeds

¹Ideal: Means of ideal attribute intensities of three sessions were included into ANOVA model to calculate least square mean

²PM aging = post-mortem aging

³Beef flavor; tenderness; juiciness: Sensory attributes of beef flavor, tenderness and juiciness are evaluated on 9-point intensity scales (9 extremely, 1 not at all intense/juicy/tender)



Figure 3-1 Tenderness of beef rib-eye steaks as affected by an interaction between breed, RFI and aging period

^{a, b, c}Columns with different letter are significantly different (P < 0.05). Error bars are standard error of mean (SEM). Ang = Angus, Cha = Charolais, KC = Kinsella Composite. Tenderness is evaluated on 9-point intensity scales (9 extremely, 1 not at all tender)

Figure 3-2 Consensus coordinates for consumer panel sensory attributes and meat samples derived from generalized Procrustes analysis (GPA) for first two dimensions



Biplot (axes F1 and F2: 80.29 %)

F1= First principal component of GPA; F2= Second principal component of GPA; A = Angus; C = Charolais; K = Kinsella Composite; H = High RFI; L = Low RFI; 4 = 4 days of aging; 18 = 18 days of aging

Figure 3-3 PREFMAP visualization of the relationship between assessors' preference and steaks from each group of animals (left) and Contour plot of preference on steak samples from each group of animals (right).



F1= First principal component of GPA; F2= Second principal component of GPA; A = Angus; C = Charolais; K = Kinsella Composite; H = High RFI; L = Low RFI; 4 = 4 days of aging; 18 = 18 days of aging.

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Chapter 4: Conclusions and recommendations

4.1 General summary

Genetic background, animal selection and production practice are important factors affecting beef quality. The objectives of this study were to determine the effects of selecting feed efficient animals using residual feed intake (RFI) on carcass quality, meat quality, sensory palatability and consumer preferences of steaks of major beef muscles from 3 breeds of cattle and different postmortem aging times. Negative (or low) RFI animals utilize feed more efficiently by consuming less feed than expected, while positive (or high) RFI animals are considered as less feed efficient cattle by consuming more feed than expected. Clear differences of carcass quality and meat quality exist among different beef breeds, thus 3 breeds were included in the current study to investigate the influence of RFI selection on beef quality among different breeds. Postmortem aging is a widely-used practice to improve meat quality, thus this study also investigated the interaction between RFI and aging to determine if postmortem aging is influenced by RFI selection.

The results of the current study indicated that selection for reduced RFI animals successfully reduced dry matter intake while achieving similar carcass weight gain performance compared with high RFI animals, substantiating the high feed efficiency of low RFI cattle. The results of the first study (Chapter 2) suggested the limited influence of RFI selection on meat quality and sensory palatability on most of the muscle studied, except TB. The pH, flavor intensity and desirability and sustainable juiciness of TB were negatively affected by RFI selection. The results of the second study (Chapter 3), however, revealed a possible undesirable influence of RFI selection on consumer preference of ribeye steaks. These results suggest that breeding for feed efficient cattle using RFI values may affect meat quality and consumer acceptability of beef steaks, despite the limited influences observed on the major of muscles evaluated in the first study.

Animal breed significantly influenced most traits studied in this research. In both studies, steaks from Angus had better meat quality and eating experiences rated by both trained and consumer panels. However, beef quality results for Charolais and Kinsella Composite were different in the two studies. Rib-eye steaks of Charolais were juicier, more tender and had higher acceptance than steaks from Kinsella composite when evaluated by the consumer panel (chapter 3), while steaks from LL muscles of Kinsella composite received higher scores than Charolais for the sensory attributes detected with differences among breeds for trained panel (chapter 2). Possible reasons for these different type of sensory evaluation panels used and different beef cuts evaluated in these two studies. Charolais cattle had better yield quality with better hot carcass weight (HCW), carcass ribeye area (REA) and lean meat yield (LMY) compared with the other two breeds.

Both objective measurement and consumer sensory evaluation observed significant improvement of tenderness for all muscles after postmortem aging for longer times (18 versus 3 days), indicating the importance of aging for the beef industry. This is in agreement with previous research. However, an interaction among postmortem aging, breed and RFI selection on consumer perceived tenderness of ribeye steaks suggests that aging did not improve the tenderness of steaks from low RFI animals. This result may suggest a possible negative influence of selection for reduced RFI animals on the tenderization function of aging.

In conclusion, RFI can be a beneficial tool to select feed efficient animals that could reduce feed consumption and have limited impact on meat quality of most muscles. However, its possible negative influences on meat quality and consumer preferences of some beef muscles such as TB and LT, and on tenderization during post-mortem aging should be considered during breeding programs that select for feed efficient animals.

A unique aspect of this thesis research is that it is the first work to explore the influence of RFI on meat quality by both consumer and trained sensory panels, providing a comprehensive perspective of the influence of RFI on sensory meat quality. It is also
the first work to evaluate the effect of RFI selection on a variety of beef cuts, with both high-value cuts (rib eye and loin eye) and tougher cuts (cross rib and inside round).

4.2 Future work and study limitations

In this thesis research, RFI selection was shown to negatively influence consumer-perceived tenderness of muscle during post-mortem aging of ribeye steak. Further studies would be beneficial to understand the relationship between RFI selection and postmortem aging with more aging periods, as well as other typical industry practices, such as hormonal growth implant, ractopamine hydrochloride supplement and electrical stimulation, to ensure that meat quality is not affected by interaction between RFI and these practices.

The results of the second study (Chapter 3) showed that RFI selection for feed efficient animals may negatively influence consumer acceptance of ribeye steaks from Angus and Kinsella Composite, while the first study (Chapter 2) did not find any undesirable influence on sensory quality using a trained panel, indicating the different perspectives from these two sensory panels on beef steaks. As consumer acceptance of a product is vital to the success of beef industry, futures studies can be conducted to investigate influence of RFI selection on consumer acceptance of more beef cuts, such as top sirloin, strip loin.

Several studies comparing consumer and trained panels showed that both had similar discriminatory ability and reproducibility in the profiling tasks for several products, such as a beverage product (Husson et al., 2001) and a perfume product (Worch et al., 2010). Panelists from the consumer panel in the second study (Chapter 3) demonstrated their ability to discriminate among samples. However, limited studies have been performed to compare consumer and trained sensory panels on beef steak evaluation. Future studies can be conducted to compare consumer and expert profiles for beef steaks using ANOVA, correlation test and multiple factor analysis to test discriminatory ability, consensus and reproducibility (Husson et al., 2001; Worch et al., 2010).

In the second study (Chapter 3), due to the limited number of steaks available, only 24 consumers were recruited for the consumer study, resulting in reduced power for statistical analysis. However, we recruited beef consumers who frequently consumed top quality steaks to ensure the validity of their sensory assessments, and each participant evaluated all treatments. In future studies, sufficient steaks should be collected for at least 65 consumers. Additionally, steaks should be cut to a uniform thickness to facilitate consistent cooking time and final degree of doneness of the steaks.

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Appendices

Appendix 1: E-mail recruitment text for rib-eye steaks consumer panel

E-mail subject line: Beef steak lovers needed!

Hello,

I am a graduate student working with Dr. Wismer and Dr. Bruce. I'd like to post an invitation for students and staff of our department who enjoying eating beef steaks to our sensory panel to taste steaks from different breeds.

Could you please help me to spread the recruitment information? Thank you very much and have a good day.

Thanks,

Zhiqiang Jiu

Beef Steak Lovers Needed

We are from the department of AFNS and looking for volunteers to participate in a taste study of **rib-eye steak** eating quality from three beef breeds.

Volunteers will...

- ★ attend three 15-minute tasting sessions in AF233 (a) Ag/For Building
- *taste and rate 4 small beef steak samples in a session*
- Complete a demographic and beef consumption survey

To be eligible: you must...

- ✤ Be 18 years or older
- *Enjoy eating beef steaks*
- Eat 'top quality' beef steaks at least once per month (e.g. Rib eye, Tenderloin, Tbone, Top sirloin)

You will receive *a pen for first attendance and* \$5 *gift card each time for participating in the following two sessions.*

For more information or to register for a taste session, please contact Zhiqiang Jiu (MSc student) at the following email address: jiu@ualberta.ca

The plan of the study has been reviewed for its adherence to ethical guidelines and approved by the Research Ethics Board at the University of Alberta.

Appendix 2: Poster advertisement for rib-eye steak consumer panel

Beef Steak Lovers Needed!!

Researchers from the department of AFNS need volunteers to participate in a taste study of **rib-eye steak** eating quality from three beef breeds.

Volunteers will...

- attend three 15 minute tasting sessions in AF235 @ Ag/For Building
- taste and rate 4 small beef steak samples in a session
- complete a demographic and beef consumption survey

To be eligible: you must...

- be 18 years or older and enjoy eating beef steaks
- eat 'top quality' beef steaks at least once per month (e.g. Rib eye, Tenderloin, T-bone, Top sirloin)

You will receive a pen for first attendance and \$5 gift card each time for participating in the following two sessions.

Contact Zhiqiang Jiu @ jiu@ualberta.ca for more information or register for a taste session.

The pronor the seady has been	enered for its danier enere to ennear Baraer	ines and approved by the nesed en entrant	board at the oniversity of Alberta.
study: jiu@ualberta.ca Beef steak taste study: jiu@ualberta.ca	u@udy u@	Beef steak taste study: jiu@ualberta.ca Beef steak taste study: jiu@ualberta.ca	Beef steak taste study: jiu@ualberta.ca Beef steak taste study: jiu@ualberta.ca

Appendix 3: Information sheet and consent form

Research Investigators:

Zhiqiang Jiu AF 311D University of Alberta Edmonton, AB, T6G 2P5 jiu@ualberta.ca 7807093680 Dr. Wendy Wismer AF 318C University of Alberta Edmonton, AB, T6G 2P5 <u>wwismer@ualberta.ca</u> 7804922923 Dr. Heather Bruce AF 318E University of Alberta Edmonton, AB, T6G 2P5 <u>hbruce@ualberta.ca</u> 7804929871

Background

You are being asked to participate in a sensory panel to provide your opinion on the eating quality of beef steak, because you have indicated that you like and regularly consume beef steak. The results of this study will be used in support of my master's degree thesis and project report for the funding agency.

Purpose

The intent of this project is to evaluate the consumers' preference on different attributes of beef steaks from three different beef breeds, raised at the University of Alberta Kinsella Ranch.

Sensory Study Procedures

Sensory recruitment: An online registration form will be sent to you after you agree to participate in this study. You can select 3 sessions from the sensory experiment schedule provided in the form. The 3 evaluation sessions will occur at the same time & day of week over 3 consecutive weeks, or at the same time 3 days a week. Then we will send you a reminder of the sessions you selected. The sensory panel will take place in the Food Lab AF235 in the Agriculture/Forest Centre in October, 2016.

First session of consumer sensory evaluation of beef: You will participate in an evaluation session to taste rib eye beef steaks from different breed and feed efficiency animals. Before the evaluation, you will complete a demographic and steak consumption habit questionnaire. Then you will taste and evaluate 4 beef steak samples for their tenderness, juiciness, flavor and overall liking. Your participation for this session is expected to last about 15 minutes.

Second and third session of consumer sensory evaluation of beef steak: In each of the following two sessions, you will evaluate 4 beef steak samples for their tenderness, juiciness, flavor and overall liking. Each of these sessions is expected to last about 10 minutes.

Benefits

After the first session, you will receive a pen with the University of Alberta logo. After each of the second and third sessions, you will receive a \$5 gift card in acknowledgement of your time and contribution to the study. You will not benefit directly from being in this study. The results of this study will be used in support of my master's degree thesis and project report for the funding agency. The results of this study will help the Canadian Beef Industry to determine which beef breeds have better eating quality.

Potential Risks

There are no risks other than the everyday risks of consuming beef, water and unsalted crackers. The animals are from the University of Alberta Kinsella cattle herd and slaughtered at Agriculture and Agri-Food Canada in Lacombe, a federally registered and inspected facility. The meat samples will be prepared under food safe condition. If you have any allergies, sensitivities or intolerances to foods used in the study, you should not participate in this study.

Voluntary Participation and Withdrawal from the Study

Participation in this study is completely voluntary and you are under no obligation to participate. Even after you have agreed to participate in the sensory panel, you may withdraw from the sessions at any time. If you withdraw, we will continue to use your data unless you ask us not to. After the sensory panel is completed we will not be able to withdraw your data as we will destroy the participant list and the data will become anonymous.

Confidentiality

Your anonymity cannot be guaranteed in the sensory panels as several people will participate at one time and be visible to one another. A participant number assigned to you will link your evaluations from all three sessions. We will have a list with your name and your participant number in case you forget your participant number. Also, we will record your email address in the list to send you an attendance reminder before each evaluation session. When the study ends, the list will be destroyed. You will not be personally identified in the results of this study; we present our results in aggregate form.

All study documents will be kept on file in a locked cabinet at the University of Alberta for a minimum of 5 years. Computer files will be encrypted. The final result and statistical data of this project may be reported in scientific journal publication by the study team. Individual responses will be kept confidentially. We may use the data from this study in future research, but if we do this it will have to be approved by a Research Ethics Board.

Further information

If you have any concerns about this study, you may contact the Research Ethics Office at 780-492-2615. This office has no direct involvement with this project.

The plan of the study has been reviewed for its adherence to ethical guidelines and approved by the Research Ethics Board at the University of Alberta.

The food products and ingredients of some food products in this study are listed

below. Do you have any allergies, intolerances, or sensitivities to any of the following <u>food or ingredients?</u>

Distilled water

Unsalted cracker: Enriched Wheat Flour, Soybean Oil, Baking Soda, Salt, Malted Barley Flour, Yeast Amylase Protease, Sour Dough Culture. Contains: Wheat, Barley Beef steak

If you have answered "yes", please stop and tell us immediately.

Consent Statement

I have read this form and the research study has been explained to me. I have been given the opportunity to ask questions and my questions have been answered. If I have additional questions, I have been told whom to contact. I agree to participate in the research study described above and will receive a copy of this consent form. I will receive a copy of this consent form after I sign it.

Participant's Name (printed) and Signature

Date

Date

Name (printed) and Signature of Person Obtaining Consent

Appendix 4: Participant demographics and beef consumption habits

1. What is your gender?

Male
Female

2. What age group do you belong to?

18-24 years
25-34 years
35-44 years
45-54 years
55-64 years
Over 64 years

3. What is your highest education level achieved?

Some high school
High school graduate
Some collage, technical or university
University/college bachelor's graduate
Post-graduate degree

4. What is your annual household income?

Less than \$15,000
\$15,000 - \$34,999
\$35,000 - \$59,000
\$60,000 - \$79,000
More than \$80,000

5. How frequently do you typically consume beef at any meal including both home and away from home?

Never
Once per month or less
2-3 times per month
Once per week
2-3 times per week
4 or more times per week

6. Please provide the approximate percentage of your beef consumption over the past year that would include the following beef products (your best guess is fine, they should add to 100%.

_____ ground and minced (e.g., hamburger)

roasts

_____ steaks

- _____ sausage, brats, hotdogs, beef luncheon meats, deli meats
- _____ organ meats (e.g. liver, tongue, tripe, etc.)

_____ others (please list_____)

100% = sum total

7. Which type of steak do you usually consume? (please check all that apply)

🗆 Rib eye	□ Top sirloin	□ T-bone
□ Flank	□ Flank	□ Sirloin tip
□ Tenderloin	□ Strip loin	
□ Tenderloin	Round	
Others:		<u>.</u>

8. How frequently do you consume "top quality" beef steaks (e.g., Rib eye, Tenderloin, T-bone, Top sirloin)?

Never
Once per month or less
2-3 times per month
Once per week
2-3 times per week
4 or more times per week

9. Which method do you usually choose to cook a "top quality" steak?

	Frying
	Grilling - indoor
	Grilling – outdoor (BBQ)
	Broiling
	Roasting
Other	r methods:

10. How important are each of these factors when you purchase "top quality" beef steaks (e.g., Rib eye, Tenderloin, T-bone, Top sirloin)? Please **circle the value** that represents the importance of each factor based on the **scale** below, where 1 indicates "not at all important" and 5 indicates "Extremely important".

		Not at all	Slightly	Moderately	Very	Extremely	
		important	important	important	important	Important	
	Price	1	2	3	4	5	
	Freshness: (i.e. 'packaged on' date)	1	2	3	4	5	
	Steak marbling	1	2	3	4	5	
	Beef grades	1	2	3	4	5	
	Product colour	1	2	3	4	5	
Package and visual	Product food safety assurance	1	2	3	4	5	
information:	Nutritional information:	1	2	3	4	5	
	Traceability of product to farm:	1	2	3	4	5	
	Labeled organic:	1	2	3	4	5	
	Labeled hormone free:	1	2	3	4	5	
	Labeled antibiotic free:	1	2	3	4	5	
	Labeled sustainable production:	1	2	3	4	5	
	Juiciness	1	2	3	4	5	
Staals active	Tenderness:	1	2	3	4	5	
Steak eating	Flavour:	1	2	3	4	5	
experience:	Preparation ease:	1	2	3	4	5	
	Preparation time:	1	2	3	4	5	
Thank you for completing the questionnaire							

Thank you for completing the questionnaire.

Appendix 5: Sensory evaluation questionnaire for rib-eye

steaks consumer panel

- You have 4 samples in front of you, each coded with a 3-digit number. Evaluate the samples in the order they are presented,
- Please cleanse your palate between each sample by taking a bite of cracker and a sip of water.

Sample	numbe	er						
• Fl	lavour							
How inte	ense is the	BEEF F	LAVOU	JR of the	sample	?		
Not intense at all								Extremely intense
• <i>Te</i>	exture							
How TE	NDER is	the samp	le?					
Not tender at all								Extremely tender
• Ju	iiciness							
How JUI	ICY is the	e sample?						
Not juicy at all								Extremely juicy
• 0	verall of	oinion						
	-	ERALL O	PINION	l of this b	beef stea	ak?		
Dislike Extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like v muc	-
_	_					<i>(</i>		

• Do you have any comments on this steak? (optional)

Ideal Sample

• <i>Fl</i>	avour	hink abo	-				
How inte							Extremely
intense at all							intense
• <i>Te</i> How TEN	x<i>ture</i> NDER is	your ide	al rib ey	e steaks	?		
Not tender at all							Extremely tender
• <i>Ju</i> How JUI	iciness CY is yo		rib eye s	teaks?			
Not juicy at all							Extremely juicy

Appendix 6: Sensory schedule for rib-eye steaks consumer panel

Date	Time	Activity	Person responsible
The day before sensory panel	9:30 am	*Remove 8 experimental steaks from -20°C storage to 4°C fridge to thaw at sensory lab #235 for Tuesday Session.	Zhiqiang Jiu
	4:00 pm	*Prepare 8 sensory plates each with 4 pre-labeled containers, 1 fork, 1 cup for water, and napkins. *Also prepare 2 cutting board, 2 knifes, 8 pre-labeled corningware dishes for Tuesday sessions.	Zhiqiang Jiu
	9:00 am - 9:30 am	Confirm all the stuff of the sensory panel of the day, including: *checking the consent form and questionnaires; *sending the reminder to the panelists; *prepare 3 crackers to each plate; *remove 8 experimental steaks from -20°C storage to 4°C fridge to thaw at sensory lab #235 for Wednesday Session.	Zhiqiang Jiu
	9:30am – 9:45 am	*prepare and clean the sensory booth; *turn on the oven at 75°C at 9:45 am; *turn on the clam shell at 9:50 am.	Zhiqiang Jiu
	9:45 am - 10:10 am	Start cooking first 2 steaks following the procedure, including *weigh the plate, sample with package, sample with	Zhiqiang Jiu
Tuesday		plate; *insert the thermocouple in to the center of the steaks;	
Or	10:00 am	*put two steaks into the clamshell and start cooking; Start cooking next 2 steaks following the procedure,	Helper
Wednesday Or	- 10:15 am	including *weigh the plate, sample with package, sample with plate *insert the thermocouple in to the center of the steaks *put two steaks into the clamshell and start cooking	
Thursday	10:10 am – 10: 20	* record the weight of corningware dishes; * finish cooking	Zhiqiang Jiu or
Or Friday	am	*weigh the sample weight with dishes *put two steaks into the corningware dishes and keep warm in the oven.	Helper
,	10:20- 10:30 am	*Seat panelists, introducing the experiment, ask them to finish the consent form and demographic questionnaire.	Zhiqiang Jiu or Helper
	10:20 am- 10:30 am	*Start cut each steak into 8 2.54cm*1.27cm*1.27cm cubes *and put them into the foam containers according to the serving order. *turn off the oven	Helper or Zhiqiang Jiu
	10:30 am	Serving steaks to panelists.	Zhiqiang Jiu or Helper
	10:40 - 11:00 am	*Clean up the panel *and prepare for the second seating	Zhiqiang Jiu
	11:00 - 11:40 am	Lunch	

	11:50 am -1:00 pm	Repeat the procedure of 9:50 to 11:00 am	Zhiqiang Jiu & Helper
	1:00 pm	Clean up the kitchen; prepare 8 sensory plates each with 4 pre-labeled containers, 1 fork, 1 cup for water, and napkins. Also prepare 2 cutting board, 2 knifes, 8 pre- labeled corningware dishes for Tuesday sessions.	Zhiqiang Jiu
	3:00 pm	Input all collected data into excel sheet	Zhiqiang Jiu