

THE EFFECTS OF THE F94L MYOSTATIN GENE MUTATION IN
BEEF-X-DAIRY CROSSED CATTLE ON MUSCLE AND CARCASS
CHARACTERISTICS, BOXED BEEF AND RETAIL YIELDS, STEAK
SHAPE, AND PALATABILITY.

By

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titled: **The effects of the F94L myostatin gene mutation in beef-x-dairy crossed cattle on muscle and carcass characteristics, boxed beef and retail yields, steak shape, and palatability.**

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ABSTRACT

In the first study, producer live performance data and carcasses from steers (n=116) resulting from the mating of four Limousin/Angus sires heterozygous for the F94L myostatin mutation to Jersey/Holstein dams were utilized to evaluate the effects of one copy of the F94L allele on live performance, carcass traits and USDA grades, and boxed beef and retail yields. Slaughter data were collected at time of harvest and carcass data was collected 48 hours postmortem. One side of each carcass was fabricated into boxed beef and retail cuts by experienced lab personnel 5-8 days postmortem. One copy of the F94L allele did not affect gestation length, birth weight, percent unassisted births, feedlot average daily gain, live weight at harvest, hot carcass weight, or dressing percentage ($P > 0.05$). Fiber type analysis indicated that the increase in muscularity by the F94L allele in the semitendinosus and longissimus was likely due to hyperplasia as there was a 19% increase in the quantity of Type IIA and IIX fibers in the semitendinosus ($P < 0.05$) and no effect to the muscle fiber size ($P > 0.05$). Carcasses from steers with one F94L allele had larger ribeye areas (99.2 versus 92.3 sq.cm.), greater ribeye width: length ratios (0.498 versus 0.479), lower USDA yield grades (2.21 versus 2.66), and lower marbling scores (438 versus 480) ($P < 0.05$). Additionally, for boxed beef yields, one F94L allele (versus zero F94L alleles) increased ($P < 0.05$) 85/15 trimmings (+3.4 kg), top round (+1.50 kg), strip loin (+0.67 kg), eye round (+0.52 kg), tenderloin (+0.35), boneless foreshank (+0.33 kg), cap/wedge (+0.32 kg), and tri-tip (+0.22 kg) per animal. Overall, carcasses from steers with one F94L allele had greater boxed beef yield (+1.06%), boxed beef plus 85/15 trimmings yield (+1.65 %), and total retail cuts plus ground beef 85/15 yield (+1.78%) than carcasses from steers with zero F94L alleles ($P < 0.05$). One copy of the F94L allele utilized in beef \times dairy breeding system had no significant impact on live performance traits but resulted in lower marbling scores

and increased muscularity as evidenced through larger, more symmetrical ribeyes, lower USDA yield grades, and higher carcass cutout yields (both boxed beef and retail yields).

In the second study, carcasses from steers (n=57) resulting from the mating of two Limousin/Angus sires heterozygous for the F94L myostatin mutation to Jersey/Holstein dams were utilized to evaluate the effects of one copy of the F94L allele on striploin dimensionality, Warner-Bratzler shear force (WBS) and slice shear force (SSF), and sensory panel ratings. Following carcass fabrication, samples of longissimus (LD), psoas major (PM), gluteus medius (GM), semitendinosus (ST), serratus ventralis (SV), triceps brachii (TB), and biceps femoris (BF) muscles from carcasses were vacuum packaged, aged until 10 days postmortem, and frozen until processed into 2.54-cm-thick steaks for sensory and shear force analysis. Carcasses from steers (n=58) from two additional heterozygous F94L sires were identified at a commercial packing plant; strip loins were excised for sensory panel analysis and shear force measurements. Individual strip steaks were imaged at a fixed height on a gridded background and processed through image analysis software capable of measuring individual pixel size (Fiji Image J) for strip loin dimensionality. Frozen steaks allocated to both sensory and shear force analyses were thawed for 24 hours at 2°C to 0-4°C prior to cooking. Steaks were cooked at 204°C with 0% relative humidity until peak internal temperature of 69°C was reached then removed from the oven and rested until the internal temperature reached 71°C. Steak sensory attributes were identified and quantified by trained panelists using the lexicon developed by Adhikari et al. (2011). In strip steaks from carcasses with one F94L allele, longissimus muscle area was increased in steaks 4, 5, 7, 8, and 9 which resulted in less angular steaks from the center and posterior end of the strip loin (steaks 6-10) ($P < 0.05$). Of the seven muscles observed, there were no differences among the WBS or SSF regardless of carcasses F94L genotype ($P > 0.05$). LD

and GM cooked steaks from carcasses with one copy of the F94L had no differences in sensory panel ratings compared to steaks from carcasses with no copies of the F94L allele ($P > 0.05$). ST cooked steaks from carcasses with one F94L rated lower in fat-like flavor compared to carcasses with no F94L allele ($P = 0.0348$). PM cooked steaks from carcasses with one F94L allele rated lower in juiciness, fat-like flavor, buttery flavor, and umami flavor compared to no copies of the F94L ($P < 0.05$). One copy of the F94L allele utilized in beef \times dairy cross steers decreased strip loin angularity and fat-like flavor attributes in the PM and ST while WBS and SSF measurements were not impacted. Use of F94L homozygous terminal beef sires would be an easily implemented strategy for dairy producers to improve steak portion size and shape in carcasses from non-replacement calves.

CHAPTER 1 – LITERATURE REVIEW

Introduction

Traditionally, cattle are raised either for milk or meat, and breeds have adapted for those specific production systems. With that being said, about 22% of the U.S. beef supply originates from the dairy sector, a majority of that being steers born from dairy dams. As reproductive efficiency and cow size have improved through selective breeding over time, the contribution of beef from the dairy industry has continually increased (DelCurto et al., 2017). Milk production in the United States has more than doubled since 1961 while the total cow herd has steadily decreased with the advancement of many technologies, improved genetics, and breeding regimes (Ritchie et al., 2017; USDA, 2022). Advancements in breeding technologies, the lower value of dairy steers, and concerns over the disposal of low-value dairy bull calves have resulted in a larger portion of the dairy herd inseminated with beef semen (Berry, 2021); the resulting beef-X-dairy steers being raised and sold into the beef supply. This opportunity is possible mainly due to the advancements in sexed semen technologies. Prior to this technology, dairy producers needed to artificially inseminate their entire herd with dairy semen to ensure that they had a sufficient number of replacement heifers to maintain or grow their herd. Now, with sexed semen, dairy producers can breed a lower percentage of their cows for dairy heifer replacements. This has not only increased efficiency by improving dairy cow genetics for milk production by continually breeding superior milk producers but has created an opportunity to breed the remaining dairy herd for an additional purpose in mind, meat. By breeding a portion of the dairy herd to beef semen and reducing carcass conformation and yield deficiencies, the economic potential of these beef-X-dairy offspring is far greater than that of non-replacement dairy steers. Implementing the

proper beef sire genetics will be crucial in ensuring a sustainable and profitable long-term crossbreeding program.

Industry Overview

Economic factors that influence the production of milk and meat in the dairy and beef industries have played a significant role in the evolution of cattle type and their physiological and biochemical characteristics over time. Natural selection is the idea that an organism that is better adapted to the environment will reproduce and that those desirable traits will be passed along to their offspring. Similar to how natural selection emphasizes the evolution of a species based on traits of survival, humans have selectively bred cattle for desirable traits. For example, dairy cow milk yield and efficiency have improved by selectively breeding the top milk producing cows whereas the beef industry has focused more on genetic selection for muscling and marbling characteristics. This continual selection for desirable milk traits in dairy cattle has resulted in poor carcass conformation and decreased carcass cutout yield (Berry et al., 2021). Based on data from 2,590 Holstein sires, Berry (2021) reported a negative genetic correlation between milk production traits (yield, protein yield, and fat) and carcass conformation (-0.43 to -0.22) and carcass fat (-0.29 to -0.20). Furthermore, breeding for high milk yield has led to the development of long-bodied dairy cows with a greater hip-height known to cause physical challenges to commercial beef packers (Picard and Gagaoua, 2020). As dairy producers continue selection for milk traits the outcome will be poor muscling characteristics in offspring. Alternatively, selection for increased red meat yield has resulted in beef cows of moderate to large mature size with greater muscle deposition and higher percentages of glycolytic muscle fibers (a characteristic linked to greater muscle hypertrophy and red-meat yield). Genetic value estimates known as expected progeny differences (EPD's) are used to compare sires and select

genetics that a producer wants to implement. EPD's quantify the genetic potential of animals as parents for several phenotypic and production traits and are a tool the beef industry uses to compare animals within the same breed (Spangler, 2015). Understanding EPD's to select appropriate terminal beef sires will improve meat production traits important to dairy producers.

Myostatin

Myostatin Overview

Myostatin is an important protein translated by the myostatin gene (*MSTN*) that inhibits muscle growth and regulates the balance of protein synthesis and degradation. For the sake of this literature review, "*MSTN*" will be used when referring to the gene and "myostatin" will be used when referring to the protein. Several beef breeds carry naturally occurring mutations of the *MSTN* gene known to increase muscle growth due to greater prenatal muscle hyperplasia resulting in animals that have heavier muscled carcasses. Extreme mutations within the gene are commonly referred to as "double muscling" because of the obvious heavier muscularity (McPherron and Lee, 1997; Grobet et al., 1998). Many of the known *MSTN* mutations have minimal to no effect on myostatin functionality, but a commonly referenced mutation occurring frequently in Belgian Blue cattle causes complete dysfunction of the myostatin proteins because of a severe 11 base pair deletion within the *MSTN* gene. While the utilization of *MSTN* knockout beef sires in a dairy production system would increase muscling, it could also negatively affect calving ease. However, the conservative F94L myostatin mutation is a single nucleotide polymorphism (SNP) that positively impacts muscling, conformation, performance, and fat without severe dystocia effects (Grobet et al. 1998). Utilizing terminal beef sires with two copies

of the F94L mutation in a beef-X-dairy breeding model would ensure that one copy of the F94L allele is passed onto offspring to increase muscling, thereby increasing carcass yield and cutout in non-replacement progeny. Minimal research has observed the effect of the F94L myostatin mutation on the Beef-X-Dairy crossbred model and its impact on muscling, yield, and palatability.

Myostatin Gene

Located in the muscular hypertrophy (*mh*) locus at the centromeric end of bovine chromosome 2 (BTA2), Myostatin (*MSTN*), or GDF8 (growth and differentiation factor 8), is part of the Transforming Growth Factor-Beta (TGF- β) superfamily responsible for negatively regulating skeletal muscle cell proliferation (Grobet et al., 1997; McPherran et al., 1997; Smith et al., 1997). The myostatin protein is synthesized by *MSTN* and is responsible for inhibiting muscle cell growth and differentiation in fetal development and regulates protein synthesis and degradation in adult skeletal muscle tissue. The myostatin protein is synthesized in a precursor form (inactive) by the *MSTN* gene, consisting of a N-terminal (propeptide) and C-terminal (active myostatin) referred to as the latent complex (Lee, 2004). The latent complex resides in the extracellular matrix (ECM), activation of myostatin occurs when proteolysis cleaves the N-terminal. Activated myostatin (C-terminal) binds to its own receptor on the plasma membrane causing multiple downstream events that inhibit myoblast differentiation by downregulating MyoD expression mediated through the Smad 2/3 pathway suppressing myogenesis. The myostatin-smad 2/3 pathway acts as a negative regulator of skeletal muscle growth by inhibiting MyoD expression needed for myoblast differentiation. Protein synthesis and degradation are occurring simultaneously within skeletal muscle where the net result is the amount of growth. To maintain the balance of protein synthesis and degradation in skeletal muscle, myostatin binds to

its transducing receptors activating SMAD 2/3, P38, Akt, and Pi3K pathways causing downstream events that ultimately results in the inhibition of muscle regeneration and induction of fibrous (connective tissue formation) (Aiello et al., 2018; Langley et al., 2002; Lee et al., 2019). There is an intricate relationship between these pathways; mutations within the *MSTN* have been identified to cause various levels of disruptions in the functionality of myostatin resulting in varying degrees of increased muscularity as growth goes unchecked.

Mutations

The first recorded identification of double-muscled animals was documented by Culley in 1807, and for over 200 years, researchers have studied the phenotypic expression of double-muscled animals. McPherran et al. (1997) first identified the gene, *MSTN*, responsible for double-muscle syndrome in mice and soon after found that changes in *MSTN* were also responsible for the increased muscle mass that characterizes Belgian Blue and Piedmontese cattle (McPherran and Lee, 1997). Polymorphisms of *MSTN*, ranging from deletions, insertions, or substitutions occur in several European cattle breeds resulting in double-muscling phenotype; this is observed in additional beef breeds like Blond 'Aquitaine, Limousin, and others. In severe *MSTN* null cattle, muscle growth goes unchecked producing animals with as much as 20% more muscle mass compared to their normal *MSTN* counterparts (Swatland and Keiffer, 1974; Grobet et al., 1997) resulting in higher carcass yields, lower collagen, and connective tissue (leaner more tender meat) (Cieplocch et al., 2017). Additionally, *MSTN* mutations in cattle have been associated with poor fertility, lowered stress tolerance, decreased calf viability, and increased rates of dystocia (Arthur et al., 1988).

The “double-muscling” phenotype is not the result of twice as many muscles, nor the increase in size of muscle fibers (hypertrophy), but due to an increase in the number of muscle

fibers, or hyperplasia (Charlier et al., 1995; Lee et al., 2019). Myostatin's role in prenatal protein synthesis impacts muscle fiber differentiation and proliferation causing an increase in the number of muscle fibers as protein degradation is decreased due to the inactivation of *MSTN* or effectiveness of the myostatin protein. Of the 20 identified *MSTN* haplotypes, many are conservative SNP's that cause minor if any disruption to myostatin. The main mode of inheritance has been identified as "partially recessive" as animals that are heterozygous for some of these mutations reap the benefits of increased muscling and carcass leanness. Of the many possible *MSTN* mutations, six are known to be responsible for the double-muscled phenotype (Dunner et al., 2003) that is the result of complete inactivation of myostatin. One of the most recognized mutations, nt821, occurs most frequently in Belgian Blue cattle as they have been systematically selected for the double muscling phenotype; an 11 base pair deletion in the third exon of the *MSTN* gene prompts the premature termination of translation. Additionally, two nucleotide substitutions in exon one and exon three of the *MSTN* gene are likely responsible for complete loss of function of myostatin in Piedmontese cattle generated by the repeated selection of the double-muscle phenotype (McPherron and Lee, 1997; Cieplocch et al., 2017).

Interestingly, the F94L myostatin gene variant in Limousin cattle produces an intermediate muscle mass gain compared to the two previously discussed variations found in Belgian Blue and Piedmontese cattle. Identifying the F94L gene and other *MSTN* mutations in terminal sires could be a useful tool to improve efficiency by increasing the pounds of meat produced per pound of feed consumed; however, all mutations represented in beef cattle may not be suitable for a beef-x-dairy production system.

Muscle Fiber Type

Skeletal muscle is converted to meat after the slaughter of meat animals. Muscle cells, also referred to as ‘muscle fibers,’ compose up to 90% of muscle volume and are demarcated by the sarcolemma (plasma membrane) and composed of myofibrils. Successions of sarcomeres (smallest unit of skeletal muscle responsible for contraction) make up the elongated myofibrils. In short, sarcomeres consist of thin myofilaments comprised of mostly actin, and thick myofilaments comprised of myosin formed from two myosin heavy chains and four myosin light chains. Identification of myosin heavy chain isoforms can establish the type of contraction and metabolism of the muscle fiber. Myosin heavy chain isoforms are classified as I, IIa, IIb, and IIx where muscle fiber types I (slow oxidative), IIA (fast oxidative glycolytic), IIX (fast glycolytic), IIB (fast glycolytic) contain the respective isoform. Contraction speed is ultimately delineated from differences in muscle fiber ATPase activity commonly referred to as slow-twitch or fast-twitch. Muscle cell metabolism is characterized by the preferred glycolytic or oxidative pathways in the organelle, substrate, or enzyme activities of the cell. In bovine, type I, IIA, and IIX fibers are mostly identified due to the rarity of type IIB fibers (Austruc and Venian, 2017; Schreurs et al., 2011).

The number of muscle fibers is determined at birth; MSTN mutations are known to increase the number of muscle fibers (hyperplasia) during fetal development. Muscle fiber types have been closely related to meat quality, and composition of different fiber types vary greatly depending on the overall functionality of the muscle. For instance, deep postural muscles like the psoas major that runs along the lumbar vertebrae of beef consist of a greater proportion of type I fibers and reduced proportions of type II fibers (Klont et al., 1998; Song et al, 2020; Zou et al., 2023). The increased proportion of type I fibers increased tenderness and palatability because of

the smaller fiber diameter and reduced connective tissue. Alternatively, the semitendinosus, a more superficial muscle on the round of the beef carcass, tends to have a higher proportion of type II fibers (Kirchofer et al., 2002; Listrat et al., 2020), resulting in a muscle that tends to be more tough and lean; this is partially due to increased proportions of connective tissue and larger fiber diameter.

MSTN mutations are known to alter the normal distribution of fiber types in many species. In *MSTN* null mice, the proportion of type II fibers was significantly higher than in wildtype mice (Girgenrath et al., 2004). These type II fibers are characteristically larger in diameter, faster, and utilize glycolytic metabolism. The greater proportion of fibers with a larger diameter lend to a greater potential for increased muscling or muscle growth. Limited research has established in detail the effects of specific *MSTN* mutations on fiber type distribution in beef cattle. Round muscles in normal and double muscled cattle were evaluated during gestation for percentage and diameter of muscle fiber types, (Martyn et al., 2004) discovering that the proportion of type I muscle fibers were lower in double muscled cattle compared to normal cattle. Cullen et al. (2009) established that the F94L mutation had an impact on muscle fiber distribution of Jersey-Limousin cross cattle by increasing the proportion of type II fibers in the longissimus. While the increased proportion of type II muscle fibers in double-muscled animals increases muscularity and meat yields, it may affect meat quality characteristics, potentially impacting palatability positively or negatively.

F94L Mutation

F94L Overview

Shortly after the discovery of *MSTN*, Grobet et al. (1997) characterized the F94L substitution as a conservative single nucleotide polymorphism where the double-muscling effect is less severe; they originally suggested that the increased muscling in Limousin cattle may not even be due to a change in the *MSTN* genotype but perhaps due to their environment because of the moderate nature of phenotypic differences compared to other mutations. Due to the nature of the F94L substitution in *MSTN*, the double-muscling phenotype is less severe in homozygous beef cattle compared to more severe variants, but researchers have since established the positive impact on growth and muscling traits (Cieploch et al., 2017; Esmailizadeh et al, 2008; Lee et al., 2015; Sellick et al., 2007). The F94L *MSTN* mutation is characterized by a single amino acid substitution at nucleotide position 282 of cytosine to adenine (c.282C>A) in the first exon of the *MSTN* gene that results in translation of leucine instead of phenylalanine at the 94th protein sequence. The myostatin protein is synthesized in a precursor form (inactive) by the *MSTN* gene, consisting of a N-terminal (propeptide) and C-terminal (active myostatin) referred to as the latent complex. Furthermore, the F94L substitution occurs in a conserved region of the N-terminal propeptide impacting the tertiary structure of the C-terminal by reduction of loops. Proteolytic cleavage of the N-terminus results in activation of latent myostatin. It is suggested that changes to the cystine knot structure of the C-terminal caused by the F94L mutation negatively impacts the active myostatin binding ability with the receptors on plasma membrane. The inability to bind to its receptors force myogenesis to continue without balancing protein synthesis and degradation (Lee et al., 2004). Based on data from 1,140 cattle across 15 different beef and dairy breeds, Vankan et al. (2010) determined that the allelic frequency of at least one F94L allele in

Limousin cattle was 94.2% with the nearest frequency observed in Droughtmaster at 4%.

Multiple studies have determined that the F94L mutation occurs in high frequencies in Limousin cattle, but have also been observed in Angus, Charolais, Blonde'd Aquitaine, Droughtmaster, Piedmontese, and Simmental (Dunner et al., 2003; Sellick et al., 2006; Vankan et al., 2010). The frequency to which the F94L allele is found in Limousin cattle and conservative nature of the mutation lends itself to be easily implemented in beef and dairy production systems.

Production

In a study measuring traits from birth to slaughter of Hereford (normal muscling), Limousin (intermediate increase in muscling) and Piedmontese (heavy increase in muscling) cattle, Limousin showed to have heavier birth weights without impacting dystocia compared to Hereford. Further analysis of this data would show that the effects of birthweight in Limousin (F94L genotype unknown) and the Piedmontese calves (two inactive *MSTN* alleles) were similar but the % dystocia was greater in Piedmontese with the overall greatest live weight and carcass weight being that of the Limousin calves (Short et al, 2002). When measuring the effects of the F94L mutation on birth weight, HCW, and pelvic area of Limousin-Jersey backcross calves, they were unaffected (Esmailizadeh et al., 2007). Consequently, making implementation of the F94L mutation a fitting choice for dairy producers to increase the value of non-replacement calves without the negative reproductive impact of more severe *MSTN* mutations.

Muscle, Fat, and Bone

With the discovery of *MSTN*, McPherran et al. (1997) reported 2-to-3 times more muscle mass and decreases in intramuscular fat and connective tissue in *MSTN* null mice compared to the wildtype. They indicated that the increase in muscling was likely due to hyperplasia.

Mutations in the same gene were soon reported to be responsible for the double-muscling phenotype experienced in some beef cattle that results in more muscle mass (Lee et al., 1997; Grobet et al., 1997). Arnold et al. (2001) describes the physiological characteristics that comprise cattle with severe *MSTN* mutations having increased proportions of muscle with simultaneously lower proportions of fat while bone tends to remain relatively constant and unaffected. Few studies have reported the effects of the conservative F94L variant on muscling and carcass characteristics. Sellick et al. (2007) utilized three Jersey-X-Limousin (X) sires backcrossed to Limousin (L) or Jersey dams (J) and 784 XJ and XL progeny were analyzed for the effects of the F94L single nucleotide polymorphism (SNP) and three additional mutations; results determined the F94L variant produced a significant additive effect for hot carcass weight (HCW) (+9.61 kg), meat % (+1.57), ribeye area (REA) (+4.15 cm²), and silver side % (+0.33) but not for longissimus dorsi % compared to two other SNPs observed. Esmailzadeh et al. (2008) documented the additive effects of the F94L substitution on several traits including muscle mass, carcass fat mass, and bone weight of carcasses from offspring of similar crosses described above in Australia and New Zealand. The results of this study indicated an increase in meat weight from homozygous F94L Limousin backcross calves (+34.5 and +18.3 kg in Australia and New Zealand, respectively) as well as a decrease in carcass fat mass (-33 and -16% in Australia and New Zealand, respectively) with no effect in bone weight. Bennett et al. (2019) further demonstrated the additive effect of one copy of the F94L allele in Limousin cattle (+6.6 cm² for ribeye area and -2.06mm adjusted fat thickness) alluding to the overall increase in lean meat yield with at least one copy of the F94L SNP. The results from these studies indicate that the introgression of the Limousin F94L allele could be a useful tool to increase lean meat yield without the dystocia effects that occur with other *MSTN* mutations.

Meat Quality

The nature of *MSTN* mutations to increase muscling without inherently increasing lipid accumulation has led to the examination of the impact on meat quality. Bailey et al. (1982) reported a negative effect of double-muscling in Charolais cattle on color, taste, and water holding capacity but a positive effect on longissimus tenderness and suggested that more tender meat could be the result of decreased amounts of connective tissue, reduced collagen cross-linking, and smaller muscle fibers. Wheeler et al. (2001) reported through trained sensory panels that at least one inactive myostatin allele in Piedmontese cattle increased tenderness and reduced the amount of connective tissue in the longissimus, gluteus Medius, semimembranosus, and biceps femoris muscles compared to Piedmontese cattle with normal *MSTN*. Lines et al. (2009) evaluated the semitendinosus muscle for tenderness and collagen content to determine the effects of the F94L mutation in Jersey-Limousin sires backcrossed to Limousin or Jersey dams; the results showed that Warner-Bratzler shear force was reduced by 15.2% in the semitendinosus muscle of homozygous F94L carcasses likely due to the 14.3% decrease in collagen content measured by hydroxyproline concentrations compared to the wild-type genotype. Additionally, Bennet et al. (2019) reported additive effects of one F94L allele resulting in higher meat yield and more tender meat evidenced through decreased adjusted fat thickness (-2.06 mm), increased ribeye area (+6.6 cm²), and decreased longissimus slice shear force (-1.7 kg) but also resulted in a decrease in marbling score (-25.4 points). There is limited literature that details the effects of the F94L myostatin variant on meat quality and further research is needed to corroborate the impact of the F94L allele on additional muscles of importance.

Conclusion

The use of beef genetics in dairy production systems is becoming more systematically utilized in order to reduce deficiencies of dairy-type carcasses' lack of muscularity and low cutout yields. The F94L substitution in the *MSTN* gene has been shown to increase muscularity without increasing dystocia compared to more extreme *MSTN* mutations. Implementing the F94L myostatin mutation as a terminal genotype may further mitigate muscling deficiencies adding value to beef-x-dairy crossed cattle. With that being said, there has been limited research comparing the impact of the F94L myostatin genotype among beef-X-dairy crossbred cattle.

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CHAPTER 2: EFFECTS OF THE F94L MYOSTATIN GENE MUTATION IN
BEEF-X-DAIRY CROSSED CATTLE ON MUSCLE FIBER TYPE, LIVE
PERFORMANCE, CARCASS CHARACTERISTICS, AND BOXED BEEF AND
RETAIL CUT YIELDS.

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Abstract

Producer live performance data and carcasses from steers (n=116) resulting from the mating of four Limousin/Angus sires heterozygous for the F94L myostatin mutation to Jersey/Holstein dams were utilized to evaluate the effects of one copy of the F94L allele on live performance, carcass traits and USDA grades, and boxed beef and retail yields. Slaughter data were collected at time of harvest and carcass data was collected 48 hours postmortem. One side of each carcass was fabricated into boxed beef and retail cuts by experienced lab personnel 5-8 days postmortem. One copy of the F94L allele did not affect gestation length, birth weight, percent unassisted births, feedlot average daily, live weight at harvest, hot carcass weight, or dressing percentage ($P > 0.05$). Fiber type analysis indicated that the increase in muscularity by the F94L allele in the semitendinosus and longissimus was likely due to hyperplasia as there was a 19% increase in the quantity of Type IIA and IIX fibers in the semitendinosus ($P < 0.05$) and no effect to the muscle fiber size ($P > 0.05$). Carcasses from steers with one F94L allele had larger ribeye areas (99.2 versus 92.3 sq.cm.), greater ribeye width: length ratios (0.498 versus 0.479), lower USDA yield grades (2.21 versus 2.66), and lower marbling scores (438 versus 480) ($P < 0.05$). Additionally, for boxed beef yields, one F94L allele (versus zero F94L alleles) increased ($P < 0.05$) 85/15 trimmings (+3.4 kg), top round (+1.50 kg), strip loin (+0.67 kg), eye round (+0.52 kg), tenderloin (+0.35), boneless foreshank (+0.33 kg), cap/wedge (+0.32 kg), and tri-tip (+0.22 kg) per animal. Overall, carcasses from steers with one F94L allele had greater boxed beef yield (+1.06%), boxed beef plus 85/15 trimmings yield (+1.65 %), and total retail cuts plus ground beef 85/15 yield (+1.78%) than carcasses from steers with zero F94L alleles ($P < 0.05$). One copy of the F94L allele utilized in beef \times dairy breeding system had no significant impact on live performance traits but resulted in lower marbling scores and increased

muscularity as evidenced through larger, more symmetrical ribeyes, lower USDA yield grades, and higher carcass cutout yields (both boxed beef and retail yields).

Introduction

Beef from dairy herds accounts for over 20% of U.S. beef production (DelCurto et al., 2017). Use of semen from beef breeds to inseminate dairy cows has increased substantially in recent years, evidenced by an increase in annual U.S. beef semen sales of 6.15 million units from 2017 to 2021 and a concurrent decrease in annual U.S. dairy semen sales of 6.21 million units during the same time frame (NAAB, 2022). This increase in utilizing beef sires on dairy cows is due to a combination of factors including: 1) a reduced requirement for dairy heifer replacements resulting from advances in reproductive performance and a slowed global dairy herd expansion, 2) increased use of sexed semen to produce dairy heifer replacements, 3) a desire to increase the value of non-replacement calves, and 4) mounting concerns about the disposal of calves shortly after birth due to lack of market value (Berry, 2021). However, limited research has been conducted on the carcass and meat characteristics from beef-on-dairy systems and much of this limited research is from the 1960s. In an extensive review on the subject, Berry (2021) concluded “Many of the studies that have compared the performance characteristics of dairy x dairy versus beef x dairy are now dated, and a description of the dairy and beef germplasm relative to the breed as a whole is not well defined.”

One of the greatest disadvantages of producing beef from dairy cattle is the lack of muscularity in dairy cattle breeds resulting in poor carcass conformation, ill-shaped ribeyes, and poor carcass cutout yields. In fact, there is a negative genetic correlation between milk yield and carcass conformation where animals that are selected to produce more milk also tend to be more angular and less muscular (Berry et al., 2004). Based on data from 2,590 Holstein sires, Berry

(2021) reported negative genetic correlations of milk traits (milk yield, protein yield, fat yield) with carcass conformation (-0.43 to -0.22). It is therefore likely that as selection continues for increased milk production, carcass conformation and carcass cutout yield of straight dairy steer carcasses will continue to decrease. Using beef sires on dairy cows could be an efficient and effective method of reducing these carcass conformation/yield deficiencies; however, it will be critical for the dairy industry to identify the correct sires.

Myostatin is a protein produced and released by muscle cells to control the rate of muscle growth by inhibition. There are several known mutations in the myostatin gene that result in varying degrees of increased muscle growth through hyperplasia, an increase in the number of muscle fibers (Cieploch et al., 2017). Some of these mutations, such as those in Belgian Blue and Piedmontese cattle, result in extreme muscle growth sometimes termed “double muscling”. These potent mutations dramatically improve carcass conformation and cutout yield in cattle; however, these mutations have been shown to increase dystocia, negatively impacting one of the top considerations when selecting beef bulls for beef-x-dairy systems (Halfman and Sterry, 2019). Another myostatin gene mutation, termed the F94L substitution, has a less severe effect on muscling phenotype when compared to other mutations (Esmailizadeh et al., 2008). This F94L substitution has been found in several breeds of cattle, most frequently in Limousin cattle, but also present in Charolais, Blonde d’Aquitaine, Angus, Droughtmaster, Piedmontese, and Simmental (Dunner et al., 2003; Sellick et al., 2006; Vankan et al., 2010).

Cattle with the F94L myostatin gene have a higher average daily gain and likely have a lower maintenance energy requirement (Hales et al., 2020). The F94L substitution has been reported to increase ribeye area and decrease fat thickness without affecting carcass weight (Esmailizadeh et al., 2008; Alexander et al., 2009; Bennet et al., 2019). Consequently, the F94L

substitution has been shown to increase meat percentage in a carcass cutout study (Sellick et al., 2007) and increase tenderness in both longissimus (Bennet et al., 2019) and semitendinosus (Lines et al., 2009) muscles, but has been associated with decreased marbling scores (Bennet et al., 2019). Aside from the decrease in marbling, all other F94L effects appear beneficial for carcasses from a beef-on-dairy system. The increase in muscling and carcass cutout yields from the F94L substitution could especially benefit the value of beef-dairy cross carcasses. Our objective was to determine the effect of the F94L substitution in the myostatin gene when utilized in a beef-on-dairy mating system on muscle fiber type distribution and size, live animal weights, carcass characteristics, and carcass cutout yields for boxed beef and retail.

Materials and Methods

Animal care and use committee approval was not required for the methods used in this study as animal procedures were confined to commercial carcasses and retrospective evaluation of existing records; no live animals were used.

Phase I - Cattle Selection and DNA Testing

Carcasses (n=58) from steers resulting from the mating of two Limousin/Angus sires heterozygous for the F94L myostatin mutation to Jersey/Holstein dams were utilized in this research. As indicated by DNA analysis, 30 carcasses were from steers with one copy of the F94L allele and 28 carcasses were from steers with zero copies of the F94L allele. The two sires were proven sires that were somewhat divergent in their genetic makeup (Table 1). Sire A was a higher-percentage Limousin bull, whereas Sire B was an Angus-Limousin cross. Sire A's EPD's indicated that he would sire calves born with less dystocia than Sire B. Sire A had carcass genetics for higher cutability (higher REA EPD and lower YG EPD), whereas Sire B had a higher marbling EPD.

Phase II – Cattle Selection and DNA Testing

Carcasses from 58 other steers, sired by three bulls heterozygous for the F94L allele were identified in a commercial packing plant for additional carcass data. One of these three sires was also represented in Phase I (Table 1). Muscle tissue samples from these 58 steers were DNA tested for F94L genotype and breed composition.

Cattle Feeding, Management, and Slaughter

Calving data was obtained from the cooperating dairy producer and feedlot performance was obtained from the cooperating feedlot. All steers were transported from their geographic

origin to feedlot facilities in Happy, TX. Early in the feeding period, steers were given an implant containing 200 mg of trenbolone acetate and 28 mg of estradiol benzoate. Later in the finishing period, steers were reimplanted with 100 mg of trenbolone acetate and 14 mg of estradiol benzoate. Cattle were fed a finished diet that consisted of approximately 66% wheat, 27% wet gluten feed, 5% cornstalks, and less than 1% of added fat as-fed. Additionally, the finished diet was supplemented with vitamins A, D, and E, monensin, a probiotic, and tylosin phosphate. Standard feedlot procedure was modified so that the cattle used in this study were not supplemented with beta-agonists. Cattle were individually weighed at an average of 96 days of age (arrival to the feedlot), 285 days of age, 457 days of age, and 524 days of age (end of feeding period) to calculate average daily gain. Phase I cattle were transported 11 hours to the University of Arizona Food Product and Safety Lab where they were harvested in two random groups using standard U.S. beef industry practices and inspected by the USDA-FSIS. Electrical stimulation was not used on these carcasses.

Fiber Cross-Sectional Area and Myosin Heavy Chain Determination (Phase I carcasses only)

Longissimus and semitendinosus muscle samples were excised from the left side of each carcass and fixed for muscle fiber type analysis shortly after slaughter. Immunohistochemical analysis was performed at Texas Tech University according to the procedures of Hergenreder et al., 2016. Sections of meat sample were mounted on slides for staining. Slides were then incubated in a series of primary and secondary antibodies. Following this, slides were cured at 4°C for 24 h in the dark. All slides were imaged within 48 h of curing using 200X working difference magnification of an inverted fluorescence microscope (Nikon Eclipse, Ti-E; Nikon Instruments, Inc., Mellville, NY) with a UV light source (Intensilight C-HGFIE; Nikon Instruments, Inc.) and a CoolSnap ES2 monochrome camera (Photometrics, Tucson, AZ).

Images were artificially colored and analyzed using NIS Elements Imaging software (Nikon Instruments, Inc.). A total of 5 random images were taken of cryosections from each slide. All muscle fibers were counted and enumerated as a percentage of the total number of muscle fibers. The cross-sectional area of each fiber in each image was measured and expressed on a square μm basis.

Carcass Data Collection

Kidney, pelvic and heart fat (KPH) were removed hot after evisceration, weighed, and reported as a percentage of hot carcass weight (which included KPH weight). Carcasses were ribbed (cut between the 12th and 13th rib bones to expose the longissimus muscle for grading) 48 hours postmortem and carcass data were collected by experienced personnel. In phase I and video image analysis in phase II carcass data included adjusted fat thickness, ribeye area, overall maturity, dark cutting discount, marbling score, and USDA Quality and Yield grades. Additional carcass data collected in phase I only included lean and fat color readings in the L-a-b color space using a Konica Minolta CR-310 chroma meter (Konica Minolta Inc, Ramsey, NJ), ribeye measurements, marbling, and lean color measured on both right and left sides and averaged for each carcass.

Boxed Beef and Retail Yields (Phase I carcasses only)

Five to eight days postmortem, the right side of each carcass was fabricated at the University of Arizona by nine experienced meat cutters (mean = 23.1 years of experience) to determine boxed beef and retail yields. As much as possible, the same meat cutter fabricated the same cuts throughout the study. Carcass sides were fabricated first into beef subprimals to determine boxed beef yield, and subsequently into retail cuts to determine retail yield. Boxed

beef fabrication style was chosen based on the highest volume traded cuts according to the USDA report “National Weekly Boxed Beef Cutout And Boxed Beef Cuts - Negotiated Sales” (USDA-AMS, 2021). For boxed beef yield, the brisket was trimmed to 25-mm maximum fat and the other subprimals were trimmed to either 6-mm fat or 0-mm fat depending on the most common industry practices (see Table 7 for complete list of boxed beef cuts and fat trim levels). For retail yields, brisket flat and point were trimmed to 6-mm fat, strip steaks and ribeye steaks were trimmed to 3-mm fat, and all other retail cuts were trimmed to 0-mm fat (see Table 9 for complete list of retail cuts and fat trim levels). Lean trimmings for each carcass were individually course-ground, mixed, and analyzed separately for total fat analysis, determined by AOAC method 960.39, to adjust each carcass’s trimmings to a standard 15% fat.

Statistical analysis

Data were analyzed to determine the effect of one copy of the F94L myostatin gene on fiber type distribution and size, carcass data, and boxed beef and retail yields. The PROC Mixed procedure in SAS Studio (SAS Institute, Cary, NC) was used to analyze data as a mixed model with F94L genotype and sire as fixed effects, slaughter group as a random effect, and percent jersey of dam as a linear covariate, with animal serving as the experimental unit. Treatment comparisons were tested for significance at $\alpha = 0.05$.

Results and Discussion

Cattle Performance Traits

The F94L allele did not affect gestation length, birth weight, or percent unassisted births ($P > 0.05$, Table 2.2). This is very important because dairy producers cite calving ease as a high priority when selecting beef sires to mate with dairy cows (Halfman and Sterry, 2019). Other myostatin mutations are known to significantly increase dystocia; therefore, the lack of increased dystocia from the F94L gene is good news for dairy producers.

There was a large sire effect on birth weight (Table 2.2) as expected based on the sires' EPDs (Table 2.1). Jersey dams had a longer gestation length, calves with lower birth weights, and a higher percentage of unassisted births compared to Holstein dams (Table 2.2). The F94L allele did not affect average daily gain in the feedlot, live weight at harvest, hot carcass weight, or dressing percentage ($P > 0.05$, Table 2.2). Calves from Jersey dams had lower feedlot gains, live weights, and hot carcass weights than calves from Holstein dams.

Abattoir Yield

The F94L allele increased tongue weight and tongue percentage as a percent of the live weight (Table 2.3). Previous studies have cited that a characteristic of double-muscled cattle is having a larger or thicker tongue compared to normal cattle (Arthur, 1995; Kieffer and Cartwright, 1972; West, 1974) which agrees with our observation of increased tongue weight in carcasses from steers with one F94L allele. Another characteristic of double-muscled cattle is a reduction in the size of heart, lungs, and kidney (Cieploch et al., 2017) contrary to what was observed in the present study. The F94L mutation results in a less severe muscling increase

compared to other myostatin mutations, which could explain why we did not observe any major reduction in organ weights.

Sire B had greater head, tongue, liver, and heart weights and a higher liver abscess score compared to sire A (Table 2.3). Steers from Jersey dams had a heavier hide, spleen, and kidney as a percentage of live weight compared to Holstein dams (Table 2.3).

Muscle Fiber Type

Increased muscularity can be the result of hyperplasia (increased number of muscle fibers) and/or hypertrophy (increased size of muscle fibers). Hyperplasia mostly occurs prenatally, whereas most hypertrophy occurs after birth. Beef muscles are a mixture of three fiber types: Type I (slow oxidative), Type IIA (fast oxidative), and Type IIX (fast glycolytic). The F94L allele increased cross-sectional muscle area of both the longissimus and semimembranosus muscles (Table 2.4). The fiber size was not affected by F94L; however, F94L caused a greater number of Type IIA and Type IIX fibers in the semitendinosus muscle. Several studies have indicated that double-muscling cattle have higher proportions of type II fibers and lower proportions of type I fibers compared to normal cattle (Ashmore et al., 1974; Stavaux et al., 1994; Martyn et al., 2004). Cullen et al. (2009) reported that the F94L mutation in Jersey-Limousin backcross cattle increased the proportion of type II fibers in the longissimus contrary to the lack of effect we observed. It appears that the increased muscularity caused by F94L is a result of increased hyperplasia, and not increased hypertrophy.

In contrast with the F94L hyperplasia effect, increased hypertrophy caused greater muscularity of the longissimus in the offspring of Sire A compared to the offspring of Sire B (Table 2.4 and Fig. 2.1). In a review on breed effects on muscle fiber proportions, Picard and

Gagaoua (2020) indicated that Limousin cattle produce higher percentages of IIX fibers compared to Angus. In our study, sire A (high percentage Limousin) had a higher proportion of IIX muscles fibers in the semitendinosus compared to sire B (Angus-Limousin), in alignment with the breed effects reported by Picard and Gagaoua (2020). Calves from Jersey dams had smaller Type IIA fibers than calves from Holstein dams (Table 2.4 and Fig. 2.2).

Carcass Traits - Yield

Overall, the carcasses in this study were leaner than industry average and slightly leaner than those beef-dairy crosses typically marketed by the cooperating producer (Table 2.5). The F94L allele did not affect external fat thickness or KPH fat percentage ($P > 0.05$). The F94L allele did cause an increase in ribeye area of 6.9 square cm., resulting USDA Yield Grade improvement of approximately one-half grade and an increase in percentage of Yield Grade 1's and 2's. Previous studies showed additive effects of the F94L allele in Limousin-Jersey backcross progeny in Australia and New Zealand indicating an increase in ribeye area +10.5% and +4.8%, respectively (Esmailizadeh et al., 2008) and +4.15cm² (Sellick et al., 2007). The F94L allele increased both ribeye length and ribeye width, but perhaps more importantly, increased width:length ratio indicating a more symmetrical, beef-type ribeye shape.

The sire effect on ribeye area and Yield Grade was even greater than the F94L effect, revealing the importance of sire selection regardless of F94L genotype (Table 2.5). Sire A offspring had a ribeye area 11.0 square cm. larger than sire B resulting in 90% Yield Grade 1's and 2's for sire A versus 64% for sire B. Greater muscularity in carcasses of offspring from Sire A is in alignment with what is widely known regarding Limousin cattle (Continental breed) propensity for muscularity and cutability compared to Angus cattle (British breed) proclivity for size and marbling ability (Hammack, 2012). Dam breed did not affect fat thickness, ribeye area,

or Yield Grade, but carcasses from Jersey dams had more KPH fat than carcasses from Holstein dams (Table 2.5).

Carcass Traits - Quality

Among the carcasses with 0 copies of F94L, three carcasses were dark cutters (11.3%), while there were no dark cutters among carcasses with 1 copy of F94L ($P = 0.0547$, Table 2.6). The cause of dark cutting is difficult to definitively determine; however, this F94L reduction in dark cutters could be “real” because the F94L muscles tended to be more glycolytic in fiber type (Table 2.4). If the F94L allele reduces the probability of dark cutting carcasses, it would be particularly beneficial in beef-on-dairy systems because dairy carcasses tend to be more susceptible to dark cutting than native beef carcasses due to more oxidative muscle fiber types causing a higher ultimate pH and darker colored lean (Page et al., 2001).

As previously mentioned, the carcasses in this study were slightly leaner than those beef-dairy crosses typically marketed by the cooperating producer, resulting in lower marbling scores and lower percent Choice than typical beef-dairy crosses (Table 2.6). The F94L allele resulted in a marbling score reduction of approximately 50 points (Table 2.6). The reduced marbling from F94L is important to carcass value, and it is interesting because there was no reduction in adjusted fat thickness or KPH fat (Table 5). The reduced marbling score may be due to a ‘dilution’ effect on muscle collagen content resulting from the increased ribeye area, instead of a reduction in the total quantity of intramuscular fat. While the reduced marbling from F94L is certainly a negative effect, milk fat content in dairy breeds has been genetically linked to greater marbling (Thaller et al., 2003). In fact, Rust and Abney (2005) reported that Holstein steaks have advantages in flavor and tenderness compared to native beef cattle which could potentially mitigate some of the negative marbling effects of F94L in a beef-on-dairy system. There was no

effect of F94L on carcass maturity, muscle color, or fat color ($P > 0.05$, Table 2.6). Sire did not affect marbling score, even though there were large differences in marbling EPD between the four sires (Table 2.1). Carcasses from Jersey dams had higher marbling scores resulting in a greater percentage of Choice or Higher quality grades ($P < 0.05$, Table 2.6) than Holstein dams.

Boxed Beef Yields

Boxed beef weights are shown in Table 2.7 and summarized graphically in Figure 2.3. Carcasses with the F94L allele yielded a greater amount of 85/15 lean trimmings, top round, strip loin, eye of round, tenderloin, boneless foreshank, and tri-tip (greatest increases listed first). Overall, carcasses with one F94L copy yielded 7.82 kg more boxed beef ($P = 0.0732$) and 11.21 kg more total red meat yield (boxed beef + lean trimmings, $P = 0.0440$). On a percentage of hot carcass weight basis, the F94L allele resulted in a one percentage point increase in total boxed beef yield and a 1.65 percentage point increase in total red meat yield (boxed beef + lean trimmings, Table 2.8). The F94L allele decreased the percentage of trimmable fat at the packing plant from 16.95% to 15.55%. Similarly, Sellick et al. (2007) and Esmailizadeh et al. (2008) indicated that Limousin-Jersey backcross calves with one F94L allele produced carcasses with approximately 2.3% and 3.0% more meat, respectively, compared to animals with no F94L allele. Carcasses from dairy cattle have inferior red meat yield, and the F94L gene is an effective tool available to the beef industry to increase red meat yield in beef- dairy crosses.

Carcasses from Sire A had less bone and higher boxed beef and total red meat yields than carcasses from Sire B (Tables 2.7 and 2.8). Carcasses from Jersey dams had significantly less weight of boxed beef, lean trimmings, and bone (Table 2.7), mostly attributed to 34-kg lighter carcasses (Table 2.2) for Jersey dams versus Holstein dams. On a percentage basis, carcasses

from Jersey dams had a lower total boxed beef yield than carcasses from Holstein dams, resulting from lower yields in subprimals from the round (Table 2.8).

Retail Cuts Yields

Retail yields were affected by F94L, sire, and dam breed in a similar manner as boxed beef yields (Tables 2.9 and 2.10). Overall, one F94L allele resulted in 11.57 kg more total retail product per carcass and a 1.78 percentage point increase in total retail product yield (retail cuts plus ground beef). The F94L gene could be used in beef-on-dairy production systems to increase retail product yields, a trait inherently deficient in dairy cattle.

Muscle Proportion

The F94L allele increased muscling uniformly throughout the carcass, as it did not affect the proportional muscle growth across the wholesale cuts (Table 2.11). Carcasses from Sire A had higher proportions of middle meats and sirloin subprimals and a lower proportion of chuck subprimals than carcasses from Sire B. Interestingly, and perhaps of some importance to the beef-on-dairy industry segment, carcasses from Jersey dams had a higher proportion of middle meat muscles and a lower proportion of round muscles compared to carcasses from Holstein dams.

Implications

In a beef-on-dairy system, one copy of the F94L myostatin allele caused increased muscling, resulting in larger, more symmetrical ribeyes, more desirable yield grades, and higher boxed beef and retail yields, all of which address inherent deficiencies in dairy and dairy-cross carcasses. These improvements were realized with no negative effects on calving ease or live performance and minimal effects on steak quality. The F94L did cause a significant and meaningful reduction in marbling score; therefore, marbling ability should be paramount in sire

selection if F94L sire are utilized. Using a beef sire homozygous for F94L myostatin in a beef-on-dairy system would ensure that all resulting progenies have exactly one copy of the F94L allele, meaning that this genetic tool could be rapidly implemented in the beef-on-dairy industry segment.

Table 2.1. Sire characteristics (NALF, 2022).

	Sire A		Sire B		Sire C		Sire D	
Phase(s) utilized	Phases I & II		Phase I		Phase II		Phase II	
Breed Composition								
Limousin, %	78.0		44.0		66.6		65.3	
Angus, %	18.1		53.8		29.5		29.6	
Hereford, %	2.2		1.3		2.8		2.8	
Chianina, %	1.7		0.0		0.0		0.0	
Charolais, %	0.0		0.9		1.1		0.0	
Red Angus, %	0.0		0.0		0.0		2.2	
EPD's	EPD	Acc	EPD	Acc	EPD	Acc	EPD	Acc
Gestation Length, d	-4	0.19	-2	0.13	-2	0.19	-4	0.66
Calving Ease Direct, %	+10	0.61	-1	0.62	+11	0.63	+12	0.70
Birth Wt., kg	+0.1	0.94	+2.9	0.87	+0.2	0.87	+0.5	0.90
Weaning Wt., kg	+30	0.51	+35	0.49	+28	0.52	+29	0.72
Yearling Wt., kg	+44	0.50	+55	0.48	+41	0.50	+39	0.73
Carcass Wt., kg	+10	0.93	+9	0.87	+7	0.87	+12	0.88
Fat Thickness, mm	-2.8	0.90	-1.8	0.83	-2.3	0.84	-1.8	0.85
Ribeye Area, cm ²	+11.9	0.92	+3.4	0.85	+3.0	0.86	+6.5	0.88
Yield Grade	-0.81	0.66	-0.28	0.64	-0.35	0.64	-0.41	0.65
Marbling	+0.08	0.92	+0.74	0.87	+0.44	0.88	+0.13	0.88

Table 2.2. Least-squares means of cattle performance traits for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire				P _{SIRE}	Dam Breed ^a	
	0	1		A	B	C	D		Jersey	P _{BREED}
Animals, <i>n</i> ^b	28/53	30/63		28/66	30/30	0/11	0/9		58/116	
Gestation length, d	283.8	282.1	0.1445	283.7	282.1			0.2404	+5.8	0.0220
Birth weight, kg	40.0	40.0	0.9991	37.1	42.9			0.0001	-6.7	0.0057
Unassisted births, %	92.9	96.8	0.4780	97.4	92.2			0.4093	+36.8	0.0026
ADG 1 ^c , kg	0.39	0.42	0.1105	0.43	0.38			0.0534	-0.03	0.4917
ADG 2 ^d , kg	1.23	1.26	0.4090	1.24	1.25			0.7042	-0.12	0.0856
ADG 3 ^e , kg	1.35	1.34	0.9752	1.34	1.35			0.8904	-0.19	0.0859
ADG 4 ^f , kg	0.87	0.73	0.0908	0.80	0.80			0.9773	-0.36	0.6038
Total feedlot ADG, kg	1.06	1.07	0.8711	1.07	1.06			0.6860	-0.10	0.0266
Harvest weight, kg	595	597	0.8563	595	596			0.9428	-60	0.0172
Hot Carcass weight ^g , kg	382	384	0.7884	381	378	376	396	0.4257	-34	0.0019
Dressing percentage	63.9	64.4	0.1465	64.7	63.7			0.0102	-0.9	0.1769

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bPhase I / Phases I + II

^cBirth to 96 days of age

^d97 to 285 days of age

^e286 to 457 days of age

^f458 to 524 days of age

^gVariable measured in both Phase I and Phase II

Table 2.3. Least-squares means of slaughter floor weights and liver scores for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire		P _{SIRE}	Dam Breed ^a	
	0	1		A	B		Jersey	P _{BREED}
Animals, <i>n</i> ^b	28	30		28	30		58	
Weights								
Feet, kg	10.42	10.34	0.7141	10.19	10.58	0.1225	-1.10	0.0236
Hide, kg	48.75	49.16	0.7696	48.04	49.16	0.3369	-2.19	0.3205
Head (skinned, tongue out), kg	15.69	15.84	0.6115	15.34	16.19	0.0107	-0.88	0.1490
Tongue, kg	1.41	1.50	0.0050	1.40	1.51	0.0025	-0.10	0.1452
Liver, kg	8.15	8.24	0.7454	7.89	8.49	0.0493	-0.85	0.1415
Spleen, kg	1.35	1.32	0.4737	1.30	1.36	0.1456	+0.02	0.7747
Lungs/trachea, kg	6.62	6.48	0.5398	6.60	6.49	0.6411	-0.25	0.5847
Heart, kg	2.70	2.63	0.3364	2.56	2.77	0.0139	-0.24	0.1246
Oxtail, kg	1.47	1.47	0.9531	1.44	1.50	0.4030	-0.08	0.5140
Percent of live wt								
Feet, %	1.757	1.733	0.4307	1.722	1.768	0.1647	-0.001	0.9893
Hide, %	8.222	8.118	0.3798	8.123	8.216	0.4721	+0.494	0.0470
Head (skinned, tongue out), %	2.650	2.657	0.8728	2.597	2.710	0.0121	+0.139	0.0988
Tongue, %	0.238	0.251	0.0032	0.237	0.252	0.0024	+0.010	0.2831
Liver, %	1.374	1.382	0.8616	1.336	1.421	0.0977	+0.016	0.8665
Spleen, %	0.227	0.221	0.3168	0.220	0.228	0.2382	+0.029	0.0381
Lungs/trachea, %	1.118	1.087	0.4277	1.117	1.087	0.4820	+0.076	0.3468
Heart, %	0.456	0.440	0.0670	0.433	0.463	0.0021	+0.007	0.6897
Oxtail, %	0.248	0.246	0.8559	0.244	0.250	0.4527	+0.012	0.4567
Liver abscess score ^c	2.35	2.47	0.7422	1.86	2.96	0.0078	-0.75	0.3251

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bPhase I / Phases I + II

^cLiver abscess score: 0 = no abscesses, 1 = 1 to 2 abscesses less than 2 cm dia., 2 = 2 to 4 abscesses between 2 and 4 cm dia., 3 = 1 abscess >4 cm dia. or >4 small abscesses, 4 = same criteria as 3 with adhesions to the body cavity.

Table 2.4. Least-squares means of muscle histology traits for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire		P _{SIRE}	Dam Breed ^a	
	0	1		A	B		Jersey	P _{BREED}
Animals, <i>n</i>	27	30		27	30		57	
Longissimus muscle								
Muscle area ^b , cm ²	99.5	105.4	0.0371	107.8	97.2	0.0008	-6.1	0.3157
Type I fiber proportion, %	36.2	36.4	0.8986	36.4	36.2	0.9068	-1.9	0.5921
Type IIA fiber proportion, %	31.2	30.1	0.5516	29.5	31.7	0.2846	+0.2	0.9569
Type IIX fiber proportion, %	32.6	33.5	0.6113	34.1	32.1	0.3108	+1.6	0.6702
Type I fiber quantity, 1,000's ^c	1,017	1,073	0.3601	1,064	1026	0.5747	+150	0.2757
Type IIA fiber quantity, 1,000's ^c	872	889	0.7899	856	905	0.4800	+175	0.2290
Type IIX fiber quantity, 1,000's ^c	926	984	0.4099	994	917	0.3130	+255	0.1061
Type I fiber size, μm ²	2,416	2,357	0.5456	2,346	2,427	0.4347	-235	0.2624
Type IIA fiber size, μm ²	3,855	3,754	0.6594	4,061	3,549	0.0442	-1,571	0.0026
Type IIX fiber size, μm ²	4,129	4,211	0.7009	4,466	3,874	0.0125	-617	0.1833
Semitendinosus Muscle								
Muscle area ^d , cm ²	91.4	101.8	0.0005	96.9	96.3	0.8320	-21.3	0.0009
Type I fiber proportion, %	20.1	17.2	0.1099	17.8	19.5	0.3610	-2.1	0.5840
Type IIA fiber proportion, %	35.2	35.9	0.7612	33.3	37.7	0.0760	+2.2	0.6603
Type IIX fiber proportion, %	44.8	46.9	0.4078	48.9	42.7	0.0326	-0.1	0.9910
Type I fiber quantity, 1,000's ^c	405	401	0.9183	387	420	0.4502	-92	0.3012
Type IIA fiber quantity, 1,000's ^c	710	830	0.0351	726	814	0.1525	-10	0.9372
Type IIX fiber quantity, 1,000's ^c	921	1122	0.0418	1,123	920	0.0579	-106	0.6132
Type I fiber Size, μm ²	3,363	3,202	0.4152	3,217	3,348	0.5392	+322	0.4528
Type IIA fiber Size, μm ²	4,056	3,898	0.5225	3,963	3,990	0.9181	-203	0.7041
Type IIX fiber Size, μm ²	5,013	4,917	0.7798	4,923	5,006	0.8243	-1,061	0.1592

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bLongissimus muscle area is average of steaks 3 through 7 of strip loin.

^cFiber quantity estimate calculated assuming total fiber area comprises 94% of total muscle area.

^dSemitendinosus muscle area is average of two steaks from center of eye round.

Table 2.5. Least-squares means of yield grade and ribeye traits for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire				P _{SIRE}	Dam Breed ^a	
	0	1		A	B	C	D		Jersey	P _{BREED}
Animals, <i>n</i> ^b	28/53	30/63		28/66	30/30	0/11	0/9		58/116	
Adj. Fat Thickness ^c , cm	0.95	0.80	0.1017	0.98	1.04	0.78	0.85	0.1352	+0.20	0.0549
Ribeye Area ^c , cm ²	92.3	99.2	0.0001	102.7	91.7	92.3	96.3	0.0001	-3.8	0.2506
KPH Fat, %	3.00	2.85	0.1709	3.02	2.79	2.94	2.94	0.5210	+0.57	0.0072
USDA Yield Grade ^c	2.66	2.21	0.0001	2.16	2.69	2.43	2.46	0.0028	+0.21	0.2903
Yield Grade 1 & 2 ^c , %	75.1	90.3	0.0220	90.3	64.5	81.1	94.7	0.0479	+0.0	0.9965
Ribeye Shape Traits										
Length, cm	15.0	15.4	0.0292	15.6	14.8			0.0004	-0.3	0.4068
Width 25, cm	8.1	8.2	0.2648	8.4	7.9			0.0023	-0.4	0.2073
Width 50, cm	7.2	7.7	0.0020	7.9	7.0			0.0001	-0.2	0.5880
Width 75, cm	6.1	6.3	0.3374	6.5	5.9			0.0006	-0.1	0.8616
Width 50: Length Ratio	0.479	0.498	0.0372	0.509	0.469			0.0002	-0.004	0.8348

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bPhase I / Phases I + II.

^cVariable measured in both Phase I and Phase II.

Table 2.6. Least-squares means of quality grade and color traits for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire				P _{SIRE}	Dam Breed^a	
	0	1		A	B	C	D		Jersey	P _{BREED}
Animals, <i>n</i> ^b	28/53	30/63		28/66	30/30	0/11	0/9		58/116	
Dark Cutters, %	11.3	0.0	0.0547	3.8	6.8			0.6604	-9.9	0.4340
Skeletal Maturity ^c	169	173	0.1950	178	164			0.0001	+11	0.0720
Lean Maturity ^c	164	172	0.0926	163	172			0.0805	-9	0.3318
Overall Maturity ^c	168	173	0.0633	173	167			0.0337	+2	0.7234
Marbling ^{d,e}	480	438	0.0035	474	451	466	444	0.5316	+61	0.0286
Choice & Higher ^e , %	73.8	66.9	0.3666	84.0	66.1	57.1	74.2	0.1181	+29.5	0.0439
Top Choice & Higher ^e , %	25.9	12.7	0.1196	35.4	19.0	11.6	11.1	0.1766	+35.4	0.0298
Muscle L* ^f	39.04	39.75	0.2043	40.00	38.79			0.0554	+1.34	0.2426
Muscle a* ^g	24.53	25.14	0.1859	25.39	24.27			0.0328	+1.47	0.1246
Muscle b* ^h	10.43	10.94	0.1209	11.09	10.27			0.0267	+1.07	0.1153
Fat L* ^f	74.53	74.86	0.5061	74.54	74.84			0.5844	+1.08	0.2870
Fat a* ^g	11.49	11.33	0.7504	11.72	11.10			0.2635	+0.55	0.5855
Fat b* ^h	15.30	15.62	0.3062	15.47	15.45			0.9610	-0.51	0.4360

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bPhase I / Phases I + II

^c100 = A⁰⁰, 200 = B⁰⁰, 300 = C⁰⁰, etc.

^d300 = Slight⁰⁰, 400 = Small⁰⁰, etc.

^eVariable measured in both Phase I and Phase II.

^fL*: 0 = black, 100 = white

^ga*: -60 = green, +60 = red

^hb*: -60 = blue, +60 = yellow

Table 2.7. Least-squares means of boxed beef weights (per head) for F94L, sire, and dam breed effects.

	# F94L copies			Sire			Dam Breed ^a	
	0	1	P _{F94L}	A	B	P _{SIRE}	Jersey	P _{BREED}
Animals, <i>n</i>	28	30		28	30		58	
109B Cap and Wedge ^b , kg	3.68	4.00	0.0132	4.00	3.67	0.0210	-0.98	0.0004
112A Ribeye Roll, kg	13.62	14.11	0.1823	14.66	13.06	0.0002	-1.02	0.1813
114A Shoulder Clod ^c , kg	18.01	18.58	0.2806	18.45	18.13	0.5795	-3.34	0.0034
115D Pectoral Meat ^b , kg	1.89	2.03	0.0565	1.94	1.98	0.6676	-0.48	0.0027
116A Chuck Roll ^c , 1x1, kg	18.06	18.38	0.6410	18.06	18.38	0.6817	-2.90	0.0500
116B Chuck Tender ^b , kg	2.88	2.95	0.5460	2.97	2.86	0.3235	-0.58	0.0112
116G Chuck Flap ^b , kg	1.57	1.57	0.9720	1.56	1.59	0.7178	-0.14	0.3861
117 Foreshank, bnls, kg	4.97	5.30	0.0129	5.17	5.10	0.6156	-0.80	0.0039
120 Brisket ^d , kg	9.89	10.21	0.3253	9.62	10.48	0.0193	-1.71	0.0146
121C Outside Skirt ^b , kg	1.67	1.68	0.9435	1.72	1.63	0.1884	-0.19	0.1573
121D Inside Skirt ^b , kg	2.66	2.69	0.7472	2.79	2.56	0.0272	-0.05	0.7765
123A Plate Short Ribs ^c , kg	5.09	4.98	0.5579	4.95	5.12	0.3983	-0.59	0.1240
124 Back Ribs, kg	3.35	3.47	0.0610	3.46	3.36	0.1679	-0.26	0.0629
130 Chuck Short Ribs ^b , kg	3.14	3.12	0.8407	3.11	3.15	0.6948	-0.40	0.0609
140 Hanging Tender, kg	1.23	1.21	0.7439	1.27	1.17	0.0858	+0.03	0.7687
157 Hindshank, bnls, kg	4.97	5.18	0.0791	5.08	5.07	0.9100	-0.67	0.0093
167A Knuckle ^b , kg	9.81	10.19	0.1118	10.11	9.89	0.4062	-1.99	0.0002
168 Top Round ^c , kg	19.25	20.75	0.0043	20.58	19.42	0.0391	-3.30	0.0025
171B Outside Round ^c , kg	13.39	13.81	0.2916	13.84	13.67	0.2834	-3.49	0.0001
171C Eye of Round ^c , kg	5.57	6.09	0.0057	5.90	5.76	0.5077	-1.58	0.0001
171F Heel ^c , kg	4.48	4.68	0.1255	4.62	4.54	0.5832	-1.20	0.0001
180 Strip Loin ^c , 0x1, kg	11.66	12.33	0.0240	12.21	11.78	0.1871	-1.00	0.1059
184 Top Sirloin Butt ^c , kg	10.56	10.09	0.1083	11.17	10.48	0.0588	-1.21	0.0782
185A Bot. Sirloin Flap ^b , kg	3.13	3.16	0.7829	3.23	3.07	0.1316	-0.72	0.0007
185B Ball Tip ^b , kg	1.37	1.43	0.1146	1.42	1.39	0.3972	-0.28	0.0002
185D Tri-Tip ^b , peeled, kg	2.64	2.86	0.0111	2.92	2.57	0.0005	-0.34	0.0602
189A Tenderloin ^b , kg	5.50	5.85	0.0205	5.70	5.66	0.7777	-0.60	0.0503

193 Flank Steak ^b , kg	1.72	1.81	0.1488	1.89	1.64	0.0008	-0.07	0.5757
Lean Trimmings 85/15, kg	57.29	60.69	0.0201	59.14	58.85	0.8530	-8.15	0.0077
Fat, kg	64.45	60.08	0.1725	61.20	63.34	0.5404	+1.72	0.7942
Bone, kg	59.54	59.48	0.9634	57.67	61.35	0.0091	-9.11	0.0008
Kidneys, kg	1.40	1.42	0.6720	1.42	1.40	0.7885	+0.10	0.3997
<u>Summations</u>								
Total Boxed Beef, kg	184.50	192.32	0.0732	191.13	185.70	0.2518	-29.92	0.0014
Total Boxed Beef + Lean Trimmings, kg	241.80	253.01	0.0440	250.26	244.55	0.3412	-38.07	0.0014

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bTrim = 0-mm fat.

^cTrim = 6-mm fat.

^dTrim = 25-mm fat.

Table 2.8. Least-squares means of carcass shrinks and boxed beef yields (% of hot carcass weight) for F94L, sire, and dam breed effects.

	<u># F94L copies</u>			<u>Sire</u>			<u>Dam Breed^a</u>	
	0	1	P _{F94L}	A	B	P _{SIRE}	Jersey	P _{BREED}
Animals, <i>n</i>	28	30		28	30		58	
Carcass Cooler Shrink, %	2.05	2.08	0.8005	2.02	2.11	0.4927	-0.18	0.5010
Carcass Cutting Shrink, %	0.44	0.46	0.8686	0.47	0.43	0.7036	+0.01	0.9706
109B Cap and Wedge ^b , %	0.97	1.03	0.0171	1.04	0.96	0.0038	-0.14	0.0117
112A Ribeye Roll, %	3.60	3.65	0.3561	3.83	3.42	0.0001	+0.12	0.2089
114A Shoulder Clod ^c , %	4.75	4.81	0.4778	4.82	4.73	0.3246	-0.29	0.0944
115D Pectoral Meat ^b , %	0.50	0.52	0.1288	0.51	0.52	0.6861	-0.06	0.0901
116A Chuck Roll ^c , 1x1, %	4.76	4.75	0.9012	4.71	4.80	0.5525	-0.18	0.4958
116B Chuck Tender ^b , %	0.76	0.76	0.9993	0.78	0.75	0.2566	-0.06	0.2721
116G Chuck Flap ^b , %	0.42	0.41	0.6444	0.41	0.42	0.6891	+0.01	0.7153
117 Foreshank, bnls, %	1.31	1.38	0.0284	1.35	1.24	0.6232	-0.06	0.3734
120 Brisket ^d , %	2.61	2.65	0.4640	2.52	2.74	0.0007	-0.12	0.3140
121C Outside Skirt ^b , %	0.44	0.43	0.5066	0.45	0.43	0.1082	+0.00	0.9552
121D Inside Skirt ^b , %	0.70	0.70	0.6838	0.73	0.67	0.0055	+0.07	0.0662
123A Plate Short Ribs ^c , %	1.34	1.29	0.0806	1.30	1.34	0.2464	+0.00	0.9504
124 Back Ribs, %	0.88	0.90	0.2449	0.91	0.88	0.1703	+0.04	0.2291
130 Chuck Short Ribs ^b , %	0.83	0.81	0.3365	0.81	0.82	0.6550	-0.01	0.9188
140 Hanging Tender, %	0.33	0.31	0.2973	0.33	0.31	0.0442	+0.05	0.0548
157 Hindshank, bnls, %	1.31	1.34	0.1386	1.33	1.33	0.9548	-0.01	0.7511
167A Knuckle ^b , %	2.59	2.64	0.2140	2.64	2.59	0.2367	-0.20	0.0202
168 Top Round ^c , %	5.09	5.37	0.0024	5.38	5.08	0.0039	-0.23	0.2283
171B Outside Round ^c , %	3.53	3.57	0.5999	3.61	3.49	0.1152	-0.48	0.0015
171C Eye of Round ^c , %	1.47	1.58	0.0076	1.54	1.51	0.4172	-0.22	0.0081
171F Heel ^c , %	1.18	1.21	0.2484	1.21	1.19	0.4454	-0.17	0.0019
180 Strip Loin ^c , 0x1, %	3.08	3.20	0.0211	3.19	3.08	0.0478	+0.11	0.2709
184 Top Sirloin Butt ^c , %	2.79	2.87	0.1931	2.92	2.74	0.0104	+0.01	0.9060

185A Bot. Sirloin Flap ^b , %	0.83	0.82	0.5846	0.84	0.80	0.0313	-0.09	0.0111
185B Ball Tip ^b , %	0.36	0.37	0.2178	0.37	0.36	0.2389	-0.03	0.0193
185D Tri-Tip ^b , peeled, %	0.70	0.74	0.0145	0.76	0.67	0.0001	-0.01	0.8674
189A Tenderloin ^b , %	1.45	1.52	0.0239	1.49	1.48	0.7515	+0.02	0.6724
193 Flank Steak ^b , %	0.45	0.47	0.2212	0.49	0.43	0.0001	+0.03	0.1566
Lean Trimmings 85/15, %	15.14	15.73	0.0282	15.49	15.39	0.7322	-0.23	0.6712
Fat, %	16.95	15.55	0.0308	15.93	16.57	0.1843	+2.25	0.0906
Bone, %	15.73	15.47	0.3217	15.13	16.06	0.0019	-0.42	0.4395
Kidneys, %	0.37	0.37	0.9455	0.37	0.37	0.7769	+0.08	0.0270
<u>Summations</u>								
Total Boxed Beef, %	48.72	49.78	0.0155	49.94	48.55	0.0044	-1.87	0.0397
Total Boxed Beef + Lean Trimmings, %	63.86	65.51	0.0051	65.43	63.94	0.0197	-2.09	0.0796

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bTrim = 0-mm fat.

^cTrim = 6-mm fat.

^dTrim = 25-mm fat.

Table 2.9. Least-squares means of retail cut weights (per head) for F94L, sire, and dam breed effects.

	# F94L copies			Sire			Dam Breed ^a	
	0	1	P _{F94L}	A	B	P _{SIRE}	Jersey	P _{BREED}
Animals, <i>n</i>	28	30		28	30		58	
Ribeye Steaks ^b , kg	12.04	12.58	0.0975	12.93	11.68	0.0007	-0.54	0.1098
Arm Roasts ^c , kg	10.15	10.65	0.1342	10.40	10.40	0.9929	-1.22	0.0007
Flat Iron Steaks ^c , kg	2.32	2.29	0.7375	2.39	2.22	0.0519	-0.17	0.0447
Shoulder Tender ^c , kg	0.76	0.80	0.1139	0.80	0.75	0.0674	-0.07	0.0171
Under Blade Roasts ^c , kg	17.70	17.96	0.6879	17.74	17.92	0.8069	-1.33	0.0557
Chuck Tender Roast ^c , kg	2.88	2.95	0.5460	2.96	2.86	0.3235	-0.58	0.0112
Chuck Flap ^c , kg	1.57	1.57	0.9720	1.56	1.59	0.7178	-0.14	0.3861
Brisket, Point-Half ^d , kg	5.22	5.30	0.6653	5.06	5.45	0.0753	-0.45	0.0310
Brisket, Flat-Half ^d , kg	3.95	4.13	0.1458	3.91	4.18	0.0526	-0.29	0.0292
Outside Skirt ^c , Peeled, kg	1.36	1.40	0.4015	1.41	1.35	0.3055	-0.12	0.0324
Inside Skirt ^c , kg	2.65	2.68	0.7472	2.79	2.56	0.0272	-0.05	0.7765
Plate Short Ribs ^c , kg	4.56	4.50	0.6897	4.45	4.60	0.4004	-0.27	0.1088
Back Ribs, kg	3.35	3.47	0.0610	3.46	3.36	0.1679	-0.26	0.0629
Chuck Short Ribs ^c , kg	3.14	3.12	0.8407	3.11	3.15	0.6948	-0.40	0.0609
Round Tip Steaks ^c , kg	8.84	8.85	0.9906	8.88	8.82	0.8601	-1.08	0.0009
Top Round Roasts ^c , kg	15.30	16.72	0.0133	16.32	15.71	0.3215	-1.50	0.0121
Bottom Round Roasts ^c , kg	11.04	11.48	0.1924	11.49	11.04	0.2211	-1.36	0.0002
Eye of Round Roasts ^c , kg	4.93	5.49	0.0037	5.27	5.15	0.5374	-0.60	0.0028
Strip Steaks ^b , kg	11.06	11.73	0.0217	11.57	11.21	0.2515	-0.53	0.0775
Top Sirloin Steaks ^c , kg	5.79	6.14	0.0592	6.19	5.74	0.0265	-0.42	0.0310
Coulotte Steaks ^c , kg	2.10	2.25	0.0970	2.28	2.07	0.0429	-0.06	0.5314
Flap Steaks ^c , kg	3.13	3.16	0.7829	3.23	3.07	0.1316	-0.72	0.0007
Ball Tip Steaks ^c , kg	0.90	0.94	0.1164	0.93	0.91	0.4250	-0.10	0.0002
Tri-Tip Roast ^c , kg	2.64	2.86	0.0111	2.92	2.57	0.0005	-0.34	0.0602
Tenderloin Steaks ^c , kg	4.56	4.82	0.0242	4.70	4.68	0.8874	-0.32	0.0078
Tenderloin Tips ^c , kg	0.54	0.52	0.5007	0.53	0.52	0.8839	-0.02	0.4761
Flank Steak ^c , kg	1.72	1.81	0.1488	1.89	1.64	0.0008	-0.07	0.5757

Ground Beef 85/15, kg	71.64	76.00	0.0077	74.37	73.28	0.4734	-10.24	0.0046
Fat, kg	69.83	64.95	0.1515	66.31	68.47	0.3616	+1.39	0.9200
Bone, kg	59.54	59.48	0.9634	57.67	61.35	0.0091	-9.11	0.0008
<u>Summations</u>								
Bnls. Trim. Retail Cuts, kg	133.13	139.08	0.0794	138.16	134.06	0.2665	-22.21	0.0022
Total Retail Cuts + Ground Beef 85/15, kg	235.75	247.32	0.0330	244.30	238.76	0.3422	-37.80	0.0011

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bTrim = 3-mm fat.

^cTrim = 0-mm fat.

^dTrim = 6-mm fat.

Table 2.10. Least-squares means of retail cut yields (% of hot carcass weight) for F94L, sire, and dam breed effects.

	# F94L copies			Sire			Dam Breed ^a	
	0	1	P _{F94L}	A	B	P _{SIRE}	Jersey	P _{BREED}
Animals, <i>n</i>	28	30		28	30		58	
Ribeye Steaks ^b , %	3.19	3.26	0.1704	3.39	3.06	0.0001	+0.10	0.3817
Arm Roasts ^c , %	2.69	2.76	0.2357	2.73	2.72	0.9214	-0.29	0.0284
Flat Iron Steaks ^c , %	0.61	0.60	0.3107	0.63	0.58	0.0127	-0.02	0.5859
Shoulder Tender ^c , %	0.20	0.21	0.2256	0.21	0.20	0.0441	-0.01	0.4225
Under Blade Roasts ^c , %	4.68	4.65	0.7998	4.64	4.69	0.7237	-0.12	0.6163
Chuck Tender Roast ^c , %	0.76	0.76	0.9993	0.78	0.75	0.2566	-0.06	0.2721
Chuck Flap ^c , %	0.42	0.41	0.6444	0.41	0.42	0.6891	+0.01	0.7153
Brisket, Point-Half ^d , %	1.38	1.38	0.9937	1.33	1.43	0.0189	-0.06	0.4389
Brisket, Flat-Half ^d , %	1.04	1.08	0.1645	1.02	1.09	0.0061	-0.02	0.6959
Outside Skirt ^c , Peeled, %	0.36	0.36	0.7881	0.37	0.35	0.1997	-0.02	0.3927
Inside Skirt ^c , %	0.70	0.70	0.6838	0.73	0.67	0.0055	+0.07	0.0662
Plate Short Ribs ^c , %	1.21	1.17	0.1526	1.17	1.20	0.2474	+0.00	0.9696
Back Ribs, %	0.88	0.90	0.2449	0.91	0.88	0.1703	+0.04	0.2291
Chuck Short Ribs ^c , %	0.83	0.81	0.3365	0.81	0.82	0.6550	-0.01	0.9188
Round Tip Steaks ^c , %	2.34	2.30	0.4543	2.33	2.31	0.7977	-0.28	0.0310
Top Round Roasts ^c , %	4.05	4.33	0.0150	4.27	4.12	0.2203	-0.27	0.2455
Bottom Round Roasts ^c , %	2.92	2.98	0.3915	3.01	2.89	0.0857	-0.36	0.0097
Eye of Round Roasts ^c , %	1.30	1.42	0.0048	1.38	1.35	0.4535	-0.14	0.0961
Strip Steaks ^b , %	2.93	3.05	0.0162	3.03	2.94	0.0750	+0.08	0.4175
Top Sirloin Steaks ^c , %	1.53	1.59	0.0932	1.62	1.50	0.0031	-0.03	0.6393
Coulotte Steaks ^c , %	0.56	0.59	0.2247	0.60	0.54	0.0341	+0.03	0.4734
Flap Steaks ^c , %	0.83	0.82	0.5846	0.84	0.80	0.0313	-0.09	0.0111
Ball Tip Steaks ^c , %	0.24	0.24	0.1967	0.24	0.24	0.2446	-0.02	0.0113
Tri-Tip Roast ^c , %	0.70	0.74	0.0145	0.76	0.67	0.0001	-0.01	0.8674
Tenderloin Steaks ^c , %	1.21	1.25	0.0558	1.23	1.23	0.9135	-0.02	0.7220
Tenderloin Tips ^c , %	0.14	0.13	0.3682	0.14	0.14	0.8977	+0.01	0.7131
Flank Steak ^c , %	0.45	0.47	0.2212	0.49	0.43	0.0001	+0.03	0.1566

Ground Beef 85/15, %	18.89	19.69	0.0061	19.46	19.12	0.0061	-0.22	0.7067
Fat, %	14.76	13.42	0.0309	13.67	14.51	0.2132	+1.68	0.1861
Bone, %	15.72	15.47	0.3277	15.13	16.06	0.0020	-0.40	0.4643
<u>Summations</u>								
Bnls. Trim. Retail Cuts, %	35.23	36.06	0.0527	36.18	35.11	0.0254	-1.49	0.0949
Total Retail Cuts + Ground Beef 85/15, %	62.40	64.18	0.0051	64.01	62.56	0.0344	-2.19	0.0888

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bTrim = 3-mm fat.

^cTrim = 0-mm fat.

^dTrim = 6-mm fat.

Table 2.11. Least-squares means of major subprimal proportion (% of major subprimals) for F94L, sire, and dam breed effects.

	<u># F94L copies</u>		P_{F94L}	<u>Sire</u>		P_{SIRE}	<u>Dam Breed^a</u>	
	0	1		A	B		Jersey	P_{BREED}
Animals, <i>n</i>	28	30		28	30		58	
Chuck Muscles ^b , %	29.6	29.0	0.1682	28.7	29.8	0.0344	-0.3	0.7443
Middle Meat Muscles ^c , %	22.5	22.6	0.6727	22.9	22.2	0.0424	+1.7	0.0038
Sirloin Muscles ^d , %	11.9	11.9	0.8652	12.1	11.7	0.0358	+0.3	0.4479
Round Muscles ^e , %	36.0	36.5	0.1506	36.3	36.2	0.8803	-1.7	0.0183

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bChuck = Shoulder Clod + Chuck Roll + Chuck Tender + Chuck Flap.

^cMiddle Meat = Ribeye Roll + Strip Loin + Tenderloin.

^dSirloin = Top Sirloin Butt + Bottom Sirloin Flap + Tri-Tip.

^eRound = Knuckle + Ball Tip + Top Round + Bottom Round + Eye of Round.

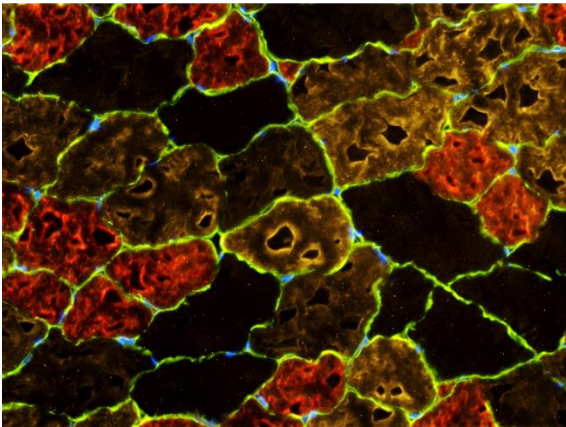
Figure 2.1. Representative micrographs of longissimus cross-sections from an offspring of sire A versus an offspring of sire B (red = Type I, yellow = type IIA, black = type IIX). ^{a,b}Least-squares means within a fiber type differ (P < 0.05).

Sire A

I: 2,346

IIA: 4,061^a

IIX: 4,466^a



Sire B

I: 2,427

IIA: 3,549^b

IIX: 3,874^b

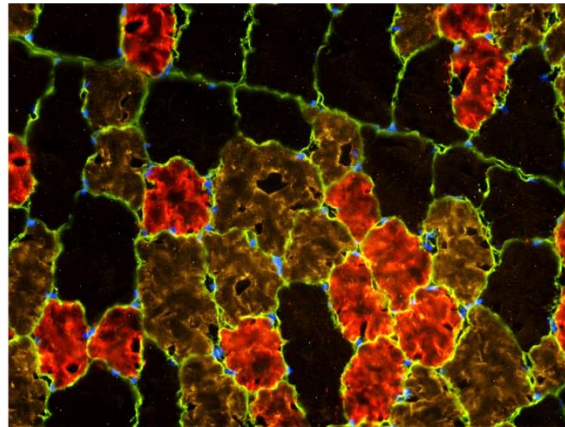


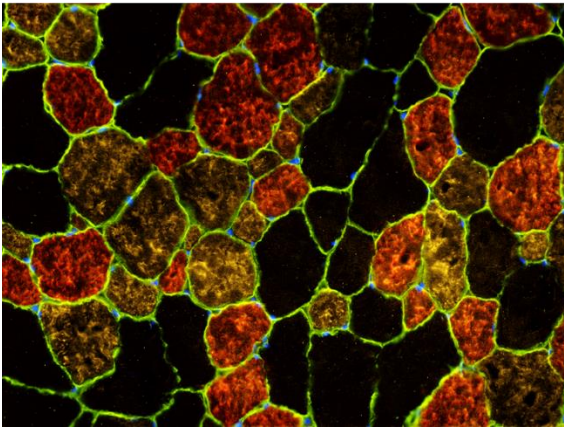
Figure 2.2. Representative micrographs of longissimus cross-sections from an offspring of a Jersey dam versus an offspring of a Holstein dam (red = Type I, yellow = type IIA, black = type IIX). ^{a,b}Least-squares means within a fiber type differ ($P < 0.05$).

Jersey dam

I: 2,323

IIA: 3,378^a

IIX: 4,002



Holstein dam

I: 2,558

IIA: 4,949^b

IIX: 4,619

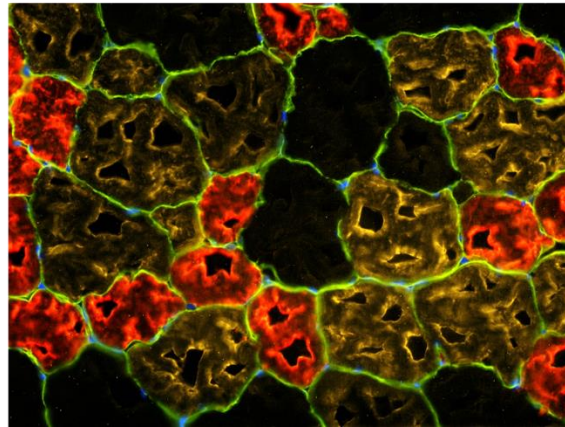
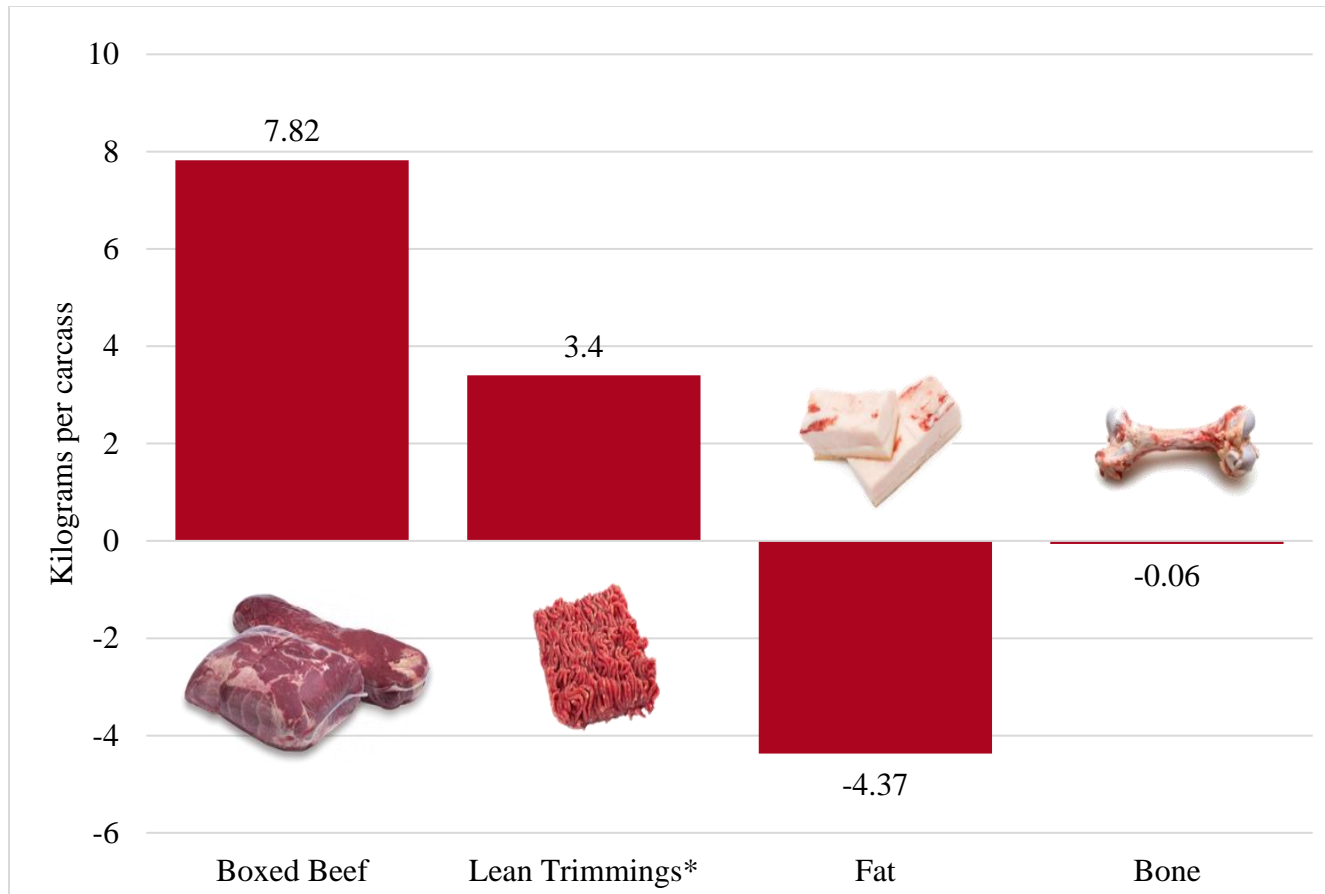


Figure 2.3. Summary of the effect of one copy of the F94L allele on boxed beef yield weights (lbs./carcass). *P < 0.05



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CHAPTER 3: THE EFFECTS OF THE F94L MYOSTATIN GENE MUTATION
IN BEEF-X-DAIRY CROSSED CATTLE ON LOIN STRIP STEAK
DIMENSIONALITY, SHEAR FORCE, AND SENSORY ATTRIBUTES.

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Abstract

Carcasses from steers (n=57) resulting from the mating of two Limousin/Angus sires heterozygous for the F94L myostatin mutation to Jersey/Holstein dams were utilized to evaluate the effects of one copy of the F94L allele on striploin dimensionality, Warner-Bratzler shear force (WBS) and slice shear force (SSF), and sensory panel ratings. Following carcass fabrication, samples of longissimus (LD), psoas major (PM), gluteus medius (GM), semitendinosus (ST), serratus ventralis (SV), triceps brachii (TB), and biceps femoris (BF) muscles from each carcass were vacuum packaged, aged until 10 days postmortem, and frozen until processed into 2.54-cm-thick steaks for sensory and shear force analysis. Carcasses from steers (n=60) from two additional heterozygous F94L sires were identified at a commercial packing plant; strip loins were excised for sensory panel analysis and shear force measurements. Individual strip steaks were imaged at a fixed height on a gridded background and processed through image analysis software capable of measuring individual pixel size (Fiji Image J) for strip loin dimensionality. Frozen steaks allocated to both sensory and shear force analyses were thawed for 24 hours at 2°C to 0-4°C prior to cooking. Steaks were cooked at 204°C with 0% relative humidity until peak internal temperature of 69°C was reached then removed from the oven and rested until the internal temperature reached 71°C. Steak sensory attributes were identified and quantified by trained panelists using the lexicon developed by Adhikari et al. (2011). In strip steaks from carcasses with one F94L allele, longissimus muscle area was increased in steaks 4, 5, 7, 8, and 9 which resulted in less angular steaks from the center and posterior end of the strip loin (steaks 6-10) ($P < 0.05$). Of the seven muscles observed, there were no differences among the WBS or SSF regardless of carcasses F94L genotype. LD and GM cooked steaks from carcasses with one copy of the F94L had no differences in sensory panel

ratings compared to steaks from carcasses with no copies of the F94L allele ($P > 0.05$). ST cooked steaks from carcasses with one F94L rated lower in fat-like flavor compared to carcasses with no F94L allele ($P = 0.0348$). PM cooked steaks from carcasses with one F94L allele rated lower in juiciness, fat-like flavor, buttery flavor, and umami flavor compared to no copies of the F94L ($P < 0.05$). One copy of the F94L allele utilized in beef \times dairy cross steers decreased strip loin angularity and fat-like flavor attributes in the PM and ST while WBS and SSF measurements were not impacted. Use of F94L homozygous terminal beef sires would be an easily implemented strategy for dairy producers to improve steak portion size and shape in carcasses from non-replacement calves.

Introduction

Beef from dairy herds accounts for over 20% of U.S. beef production (Delcurto et al., 2017) and research that has been conducted on the carcass and meat characteristics from beef-on-dairy systems is limited and outdated. One of the greatest disadvantages of producing beef from dairy cattle is the lack of muscularity in dairy cattle breeds resulting in poor carcass conformation, ill-shaped ribeyes, and poor carcass cutout yields. In fact, Berry (2021) reported negative genetic correlations of milk traits (milk yield, protein yield, fat yield) with carcass conformation (-0.43 to -0.22). It is therefore likely that as selection continues for increased milk production, carcass conformation of straight dairy steer carcasses will continue to decrease. Using beef sires on dairy cows should be an efficient and effective method of reducing these carcass conformation deficiencies; however, it will be critical for the dairy industry to identify the correct sires and market them accordingly.

Myostatin is a protein released by muscle cells responsible for the inhibition of skeletal muscle growth. There are several known mutations of the myostatin gene that result in varying degrees of increased muscle growth through hyperplasia, an increase in number of muscle fibers (Cieploch et al., 2017). Some of these mutations, such as those in Belgian Blue and Piedmontese cattle, result in extreme muscle growth termed “double muscling,” but have been shown to increase dystocia which is a major consideration in dairy production systems (Halfman and Sterry, 2019). The F94L myostatin mutation is most frequently found in Limousin cattle and has an increase in muscularity (Esmailizadeh et al., 2008) without the increased rates of dystocia of more extreme myostatin mutations. The F94L substitution has been associated with decreased marbling scores but has been shown to increase tenderness in both longissimus (Bennet et al., 2019) and semitendinosus (Lines et al., 2008) muscles. While the reduced marbling from F94L is certainly a negative effect, milk fat content in dairy breeds has been genetically linked to greater marbling (Thaller et al., 2003). In fact, Rust and Abney (2005) reported that Holstein steaks have advantages in flavor and tenderness compared to native beef cattle which could potentially mitigate some of the negative marbling effects of F94L in a beef-on-dairy system. The objective of this study was to determine the effect of the F94L substitution in the myostatin gene when utilized in a beef-on-dairy mating system on strip loin dimensionality, Warner-Bratzler shear force (WBS) and slice shear force (SSF), and sensory panel ratings.

Materials and Methods

Palatability Samples

Following carcass fabrication, samples of longissimus (strip loin), psoas major (tenderloin), gluteus medius (top sirloin), semitendinosus (eye of round), serratus ventralis (chuck flap), triceps brachii (boneless arm roast), and biceps femoris (bottom round) muscles from each carcass were vacuum packaged, aged until 10 days postmortem, and frozen for sensory and shear force analysis. These seven muscles were chosen not only because of their popularity as beef cuts but because they represent a wide range of muscle fiber types and tenderness. After freezing, 2.54-cm thick frozen steaks were cut from each muscle on a band saw, re-vacuum packaged, and stored until sensory analysis.

Steak Imaging and Dimensionality

Strip loins, tenderloins, and eye of rounds were saved from fabrication, frozen at 10 days postmortem, and held until processed into 2.54-cm-thick steaks. Individual steaks from the entire strip loin and two steaks from the center of the tenderloin and eye of round were imaged at a fixed height above the steak on a gridded background. Each digital image was processed using an image analysis software capable of measuring individual pixel size (Fiji Image J). Each image was scaled by measuring a 7.6 cm line (distance known from the gridded background in each image) and the corresponding length in pixels was entered into the software. From each steak, total steak area, steak length, and steak widths at 25/50/75/87.5% of length locations were measured.

Cooking Procedure

Frozen steaks allocated to both sensory and shear force analyses were thawed for 24 hours at 2°C to 0-4°C prior to cooking. Steaks were cooked at 204°C with 0% relative humidity and default fan speed in a combi-oven (Model SCC WE 61 E; Rational, Landberg am Lech, Germany) on a grill grate (Model SCC WE 61 E; Rational, Landberg am Lech, Germany). During the cooking process, the temperature of one steak most representative of the entire group from a weight and width standpoint was monitored using an oven core temperature probe (Model SCC WE 61 E; Rational, Landberg am Lech, Germany) placed in the geometric center of the steak until a peak internal temperature of 69°C was reached. The steaks were then removed from the oven and rested for 2 minutes to allow the internal temperature to rise to 71°C. The final temperature of each steak was measured using a calibrated, type K thermocouple thermometer (AccuTuff 340, model 34040, Cooper-Atkins Corporation, Middlefield, CT, USA).

Shear Force Measurements

Pre- and post-cooking weights were obtained for each steak in order to calculate cook loss. Warner-Bratzler shear force (WBS) and slice shear force (SSF) measurements were obtained at the University of Arizona from every steak using procedures described by Lorenzen et al. (2010). Within 3 minutes of recording peak internal temperature, the lateral end of the steak was blocked, and a 1 x 5 cm slice was removed parallel to the muscle fibers. This slice was sheared perpendicular to muscle fibers, using a slice shear force machine (Tallgrass Solutions, Inc., Manhattan, KS, USA) equipped with a flat, blunt-end blade (crosshead speed: 500 mm/min, load capacity: 50 kg), resulting in a single peak SSF measurement for each steak. The remaining steak portions were chilled overnight at 2°C and then 4 to 6 cores (1.2 cm diameter) were removed parallel to muscle fibers. Each core was sheared perpendicular to muscle fibers using a

Warner-Bratzler shear force machine (Tallgrass Solutions, Inc., Manhattan, KS, USA) fitted with a Warner-Bratzler shear head (crosshead speed: 225 mm/min, load cell capacity: 50 kg). Peak shear force of each core was recorded and resulting values were averaged to obtain a single WBS measurement for each steak.

Trained Sensory Analysis

Prior approval from the Institutional Review Board was not necessary as human subjects involvement in this research were exempt per CFR 45 Part 46.104 paragraph d section 6(i). Trained sensory analysis was conducted at Texas Tech University using consenting trained personnel. Attributes of steaks from the longissimus, semitendinosus, psoas major, and gluteus medius were evaluated using the lexicon developed by Adhikari et al. (2011). Panelists were trained to identify and quantify the following attributes: overall tenderness, overall juiciness, beef flavor identity, browned, roasted, umami, metallic, fat-like flavor, buttery flavor, sour, oxidized, and liver-like flavors. Each sample was evaluated by six or seven panelists (mean = 6.6 panelists/sample) that were seated in individual cubicles in a dark room under red incandescent lighting. Distilled water, apple juice and unsalted saltine crackers were used by panelists between samples to cleanse their palate. Cooked steaks were trimmed free of all external fat and connective tissue and then cut into 1 x 3 cm pieces on a cutting block (Model VB-150A, Tallgrass Solutions, Inc., Manhattan, KS, USA) immediately before serving to panelists. Each panelist received 2 to 3 pieces of each sample for analysis of all descriptive sensory attributes. Responses were recorded on a 100 mm continuous, unstructured line scale on an electronic ballot generated by an online survey software (Qualtrics, Provo, UT, USA). Sample ratings for each attribute were averaged across all panelists.

Additional Carcass Data and Shear Force Testing

Carcasses from 58 other steers, also sired by F94L heterozygous bulls were identified in a commercial packing plant for additional carcass data and shear force testing. Longissimus muscle samples from the 13th rib location were excised and transported to Texas Tech University for Warner-Bratzler and Slice shear force testing. Muscle tissue samples from these 58 steers were DNA tested for F94L genotype and breed composition. All aging, freezing, thawing, cooking, and shear force determination procedures were followed as outlined previously.

Statistical analysis

Data were analyzed to determine the effect of one copy of the F94L myostatin gene on strip steak dimensionality, shear force and sensory ratings. The PROC Mixed procedure in SAS Studio (SAS Institute, Cary, NC) was used to analyze data as a mixed model with F94L genotype and sire as fixed effects, slaughter group as a random effect, and percent jersey of dam as a linear covariate, with animal serving as the experimental unit. For shear force data, percent cooking loss was also included in the model as a linear covariate. Treatment comparisons were tested for significance at $\alpha = 0.05$.

Results and Discussion

Steak Dimensionality

Strip steaks from dairy-type carcasses are often discriminated against by chefs and retailers because of their narrow and angular shape. Our research measured strip steak angularity as steaks progressed anatomically through the strip loin from anterior to posterior end (Figure 3.1). Across both genotypes, strip steaks became increasingly angular from anterior to posterior end. Strip steaks from steers with one F94L allele had a less angular shape through the center and posterior portion of the strip loin (steaks 6 to 10), and this effect is important because this region of the strip loin is the most problematic in dairy-type carcasses. Based on data from 2,590 Holstein sires, Berry (2021) reported negative genetic correlations of milk traits (milk yield, protein yield, fat yield) with carcass conformation (-0.43 to -0.22). It is therefore likely that as selection continues within the U.S. dairy herd for increased milk production, carcass conformation straight dairy steer carcasses will continue to decrease. The F94L allele can reduce the strip steak shape deficiencies in dairy and dairy-cross carcasses.

Sire A had larger longissimus muscle area throughout the loin strip steaks compared to sire B; however, steak angularity trended similarly with the exception of steak 1 (most anterior) and steak 14 (most posterior) being less angular in sire A than sire B. Carcasses from Jersey dams had decreased longissimus muscle area in steak 9 to 13 leading to increased angularity of loin strip steaks 9, 11, and 12 compared to Holstein dams.

Steak Shear Force

The F94L allele did not affect Warner-Braztler (WBS) or Slice Shear Force (SSF) in any of the seven muscles examined ($P > 0.05$, Table 3.3). In beef cattle, the F94L allele has been shown to increase tenderness in both longissimus (Bennet et al., 2019) and semitendinosus

(Lines et al., 2208) muscles, so we hypothesized that tenderness would be increased in beef-dairy crosses with the F94L allele, but we did not see any shear force effect in the beef-dairy steers in this study.

Sire A had lower cooked steak shear force than Sire B in four muscles (Table 3.3). Jersey dams resulted in lower WBS of cooked bottom round (biceps femoris) steaks and SSF of cooked strip steaks (longissimus) than Holstein dams.

Steak Sensory Panel Ratings

A trained sensory panel evaluated cooked steaks from four different muscles (Tables 3.4, 3.5, 3.6, and 3.7). For three of the muscles (longissimus, semitendinosus, gluteus medius), the F94L allele had minimal to no effect on steak sensory panel ratings. In tenderloin steaks (psoas major), the F94L allele resulted in lower juiciness, fat-like flavor, buttery flavor, and umami flavor ratings, indicating that the F94L psoas major likely had a lower intramuscular fat percentage but we never measured intramuscular fat content of the psoas major. Deep postural muscles like that of the psoas major that runs along the lumbar vertebrae of beef consist of a greater proportion of type I fibers and reduced proportions of type II fibers (Klont et al., 1998; Song et al, 2020; Zou et al., 2023). Komiya et al. (2020) reported positive correlations between cooked meat samples with high levels of type I muscle fibers and umami and richness sensory attributes. However, previous studies have reported that double-muscling in cattle results in increased proportions of type II muscle fibers in animals (Cullen et al., 2009; Girgenrath et al. 2004; Martyn et al. 2004). The F94L allele could be affecting fiber type distribution in the psoas major leading to the lower sensory values observed, but fiber type analysis was not conducted on the psoas major in this study; further research is needed to corroborate this potential effect.

The sire effect on sensory panel ratings of cooked steaks was minimal (Table 3.4, 3.5, 3.6, and 3.7). Jersey dams resulted in higher tenderness, beef, browned, and fat-like flavor ratings in loin strip steaks (*longissimus*), higher liver-like flavor ratings in eye of round steaks (*semitendinosus*), and higher tenderness ratings in top sirloin steaks (*gluteus medius*) compared to Holstein dams.

Implications

In beef-dairy steers, one copy of the F94L allele decreased strip loin angularity and fat-like flavor attributes in the PM while WBS and SSF measurements were not impacted. Reduced strip steak angularity addresses inherent deficiencies in dairy and dairy-cross carcasses; thus, the F94L allele could improve the sustainability of beef-dairy strip in the retail case relative to dairy-type cattle. Using a beef sire homozygous for F94L myostatin in a beef-on-dairy system would ensure that all resulting progenies have exactly one copy of the F94L allele, meaning that this genetic tool could be rapidly implemented in the beef-on-dairy industry segment.

Table 3.1. Least-squares means of longissimus muscle area (sq.cm.) in loin strip steaks by anatomical location^a for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire		P _{SIRE}	Dam Breed ^b	
	0	1		A	B		Jersey	P _{BREED}
Animals, <i>n</i>	27	29		26	30		56	
Steak 1 (most anterior)	99.9	103.4	0.1623	106.7	96.7	0.0004	-8.2	0.1304
Steak 2	98.5	102.3	0.1110	106.3	94.5	0.0001	-4.0	0.4466
Steak 3	101.1	106.1	0.0650	109.7	97.5	0.0001	-4.9	0.4035
Steak 4	102.3	108.2	0.0418	111.2	99.4	0.0003	-6.9	0.2694
Steak 5	99.6	106.4	0.0143	107.8	98.2	0.0018	-7.8	0.1956
Steak 6	98.9	103.2	0.3474	105.2	96.9	0.0984	-4.7	0.6400
Steak 7	96.0	103.3	0.0064	105.2	94.2	0.0002	-6.2	0.2758
Steak 8	94.6	100.9	0.0256	102.3	93.1	0.0032	-9.4	0.1290
Steak 9	94.1	100.1	0.0295	101.9	92.3	0.0017	-13.5	0.0269
Steak 10	96.3	100.7	0.1363	103.8	93.3	0.0015	-14.1	0.0327
Steak 11	98.1	102.1	0.1483	103.9	96.4	0.0141	-17.0	0.0064
Steak 12	97.1	100.9	0.1943	101.4	96.6	0.1319	-17.9	0.0078
Steak 13	94.7	99.0	0.1268	97.2	96.5	0.8182	-14.3	0.0247
Steak 14 (most posterior)	88.7	93.9	0.0952	90.8	91.8	0.7499	-9.2	0.1751

^aSteaks (2.5-cm thickness) numbered starting from rib end (anterior end) of strip loin.

^bDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam

Table 3.2. Least-squares means of loin strip steak angularity (degrees) by anatomical location^a for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire		P _{SIRE}	Dam Breed ^b	
	0	1		A	B		Jersey	P _{BREED}
Animals, <i>n</i>	27	29		26	30		56	
Steak 1 (most anterior)	-2.4	+2.2	0.0169	+3.2	-3.4	0.0020	+4.5	0.2732
Steak 2	-0.2	+0.4	0.7267	+1.8	-1.5	0.1100	-3.5	0.4041
Steak 3	-1.8	-1.1	0.7134	-0.6	-2.4	0.3929	-2.8	0.5220
Steak 4	-1.1	-0.6	0.7816	-0.5	-1.2	0.6704	-4.1	0.2597
Steak 5	-2.3	-2.9	0.7135	-1.9	-3.4	0.3396	-4.7	0.1450
Steak 6	-6.1	-2.9	0.0185	-3.5	-5.5	0.1735	-4.3	0.1508
Steak 7	-5.8	-2.9	0.0430	-4.6	-4.1	0.7610	-2.9	0.3629
Steak 8	-6.0	-4.1	0.1017	-4.3	-5.9	0.2349	-2.8	0.2854
Steak 9	-8.6	-5.3	0.0046	-7.0	-7.0	0.9723	-6.3	0.0046
Steak 10	-7.9	-5.7	0.0764	-6.4	-7.2	0.5620	-3.0	0.2734
Steak 11	-7.1	-5.1	0.2574	-6.5	-5.8	0.7346	-8.5	0.0296
Steak 12	-9.4	-7.4	0.0871	-8.7	-8.1	0.6539	-6.4	0.0187
Steak 13	-8.9	-7.1	0.1432	-7.7	-8.3	0.6602	-3.4	0.2128
Steak 14 (most posterior)	-7.3	-7.7	0.8296	-5.7	-9.3	0.0289	-0.1	0.9736

^aSteaks (2.5-cm thickness) numbered starting from rib end (anterior end) of strip loin.

^bDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam

Table 3.3. Least-squares means of cooked steak shear force for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire				P _{SIRE}	Dam Breed ^a	
	0	1		A	B	C	D		Jersey	P _{BREED}
Animals, <i>n</i> ^b	27/52	30/63		27/65	30/30	0/11	0/9		57/115	
Longissimus WBS ^{c,d} , kg	3.40	3.49	0.5579	3.25	3.90	3.38	3.26	0.0469	-0.44	0.2571
Longissimus Slice Shear ^c , kg	14.9	15.2	0.7995	13.6	17.3	16.0	13.4	0.0188	-6.2	0.0016
Semitendinosus WBS ^d , kg	4.90	5.03	0.4717	4.97	4.96			0.9551	-0.14	0.7171
Semitendinosus Slice Sh., kg	23.6	22.5	0.2150	22.0	24.2			0.0248	-3.0	0.1186
Psoas Major WBS ^d , kg	2.85	2.85	0.9846	2.92	2.78			0.2197	+0.12	0.5651
Gluteus Medius WBS ^d , kg	4.81	4.49	0.2089	4.42	4.88			0.1134	-0.51	0.3579
Triceps Brachii WBS ^d , kg	3.90	3.95	0.6652	3.80	4.05			0.0334	-0.22	0.3508
Serratus Ventralis WBS ^d , kg	3.08	3.18	0.4605	3.13	3.13			0.9899	-0.02	0.9361
Biceps Femoris WBS ^d , kg	4.45	4.35	0.5395	4.14	4.64			0.0145	-1.28	0.0003

^aDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

^bPhase I / Phases I + II.

^cVariable measured in both Phase I and Phase II.

^dWBS = Warner-Bratzler Shear Force.

Table 3.4. Least-squares means of cooked **longissimus** steak sensory panel ratings^a for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire				P _{SIRE}	Dam Breed ^b	
	0	1		A	B	C	D		Jersey	P _{BREED}
Animals, <i>n</i>	52	63		65	30	11	9		115	
Tenderness	55.8	56.0	0.8994	57.8	54.5	55.5	55.9	0.0754	+5.6	0.0075
Juiciness	55.8	55.4	0.5857	55.8	53.0	57.2	56.4	0.0015	+2.7	0.0535
Beef Flavor	51.3	51.2	0.7563	51.7	51.5	50.3	51.6	0.4763	+2.2	0.0323
Browned Flavor	49.4	49.2	0.7901	50.8	49.7	47.9	48.9	0.0029	+2.9	0.0019
Roasted Flavor	52.2	52.8	0.1457	52.8	51.2	52.2	53.8	0.0052	+1.3	0.1096
Fat-Like Flavor	11.1	10.6	0.2392	10.9	10.8	10.8	10.9	0.9969	+1.6	0.0393
Buttery Flavor	1.7	1.5	0.5142	1.5	1.0	2.2	1.7	0.0967	+0.3	0.4473
Umami Flavor	15.7	15.5	0.6508	16.2	15.9	14.3	15.9	0.0703	+1.5	0.0563
Metallic Flavor	1.8	2.1	0.3535	2.1	1.8	2.8	1.0	0.1310	-0.1	0.9135
Oxidized Flavor	0.5	0.6	0.4447	0.6	0.7	0.2	0.8	0.3391	-0.2	0.4744
Liver-Like Flavor	0.6	0.7	0.3628	0.7	1.4	0.1	0.4	0.0001	+0.0	0.9753
Sour Flavor	0.2	0.1	0.3996	0.3	0.2	0.1	0.1	0.5903	+0.0	0.9842

^aAll sensory ratings are on a 0-mm to 100-mm continuous, unstructured line scale.

^bDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam.

Table 3.5. Least-squares means of cooked **semitendinosus** steak sensory panel ratings^a for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire		P _{SIRE}	Dam Breed ^b	
	0	1		A	B		Jersey	P _{BREED}
Animals, <i>n</i>	27	30		27	30		57	
Tenderness	47.0	46.3	0.4765	45.9	47.4	0.1899	+0.0	0.9929
Juiciness	52.8	51.4	0.1781	52.0	52.2	0.8826	+1.0	0.6721
Beef Flavor	50.1	50.3	0.7101	50.4	50.0	0.5649	+1.3	0.4023
Browned Flavor	46.6	47.0	0.5864	47.1	46.5	0.4780	+1.0	0.5061
Roasted Flavor	49.6	50.0	0.4877	50.2	49.4	0.2305	+1.6	0.2580
Fat-Like Flavor	10.1	9.4	0.0348	9.7	9.8	0.7424	+0.6	0.4379
Buttery Flavor	0.1	0.1	0.6519	0.1	0.1	0.9182	+0.1	0.4320
Umami Flavor	13.6	13.0	0.2630	13.3	13.4	0.7795	+0.6	0.4309
Metallic Flavor	3.6	4.0	0.3556	4.3	3.3	0.0393	+1.1	0.2369
Oxidized Flavor	1.2	1.1	0.6703	1.0	1.3	0.3163	-0.4	0.5526
Liver-Like Flavor	1.1	1.6	0.1151	1.8	0.9	0.0062	+1.5	0.0311
Sour Flavor	0.9	0.5	0.2267	1.0	0.5	0.1902	-0.2	0.7549

^aAll sensory ratings are on a 0-mm to 100-mm continuous, unstructured line scale.

^bDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam

Table 3.6. Least-squares means of cooked **psaos major** steak sensory panel ratings^a for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire		P _{SIRE}	Dam Breed ^b	
	0	1		A	B		Jersey	P _{BREED}
Animals, <i>n</i>	27	30		27	30		57	
Tenderness	78.4	77.5	0.4614	77.6	78.4	0.5286	+1.8	0.5072
Juiciness	55.5	53.5	0.0348	53.6	55.4	0.0840	-0.5	0.8118
Beef Flavor	53.3	52.5	0.1039	53.1	52.8	0.5268	-0.2	0.8305
Browned Flavor	50.0	49.8	0.7400	49.9	49.9	0.9458	-0.6	0.6219
Roasted Flavor	53.6	53.3	0.4675	53.9	52.9	0.0666	+0.7	0.5262
Fat-Like Flavor	12.3	11.0	0.0004	11.5	11.8	0.5047	+1.2	0.1370
Buttery Flavor	1.0	0.3	0.0160	0.6	0.7	0.9554	+0.4	0.4303
Umami Flavor	16.4	15.3	0.0107	15.9	15.8	0.9571	+0.4	0.6404
Metallic Flavor	1.7	2.3	0.0758	2.3	1.7	0.1500	-1.0	0.1879
Oxidized Flavor	1.1	1.3	0.5460	1.1	1.3	0.5211	-0.5	0.4762
Liver-Like Flavor	2.3	2.0	0.6105	2.2	2.0	0.7843	-0.4	0.7521
Sour Flavor	0.6	0.4	0.6550	0.5	0.5	0.8821	+0.3	0.6860

^aAll sensory ratings are on a 0-mm to 100-mm continuous, unstructured line scale.

^bDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam

Table 3.7. Least-squares means of cooked **gluteus medius** steak sensory panel ratings^a for F94L, sire, and dam breed effects.

	# F94L copies		P _{F94L}	Sire		P _{SIRE}	Dam Breed ^b	
	0	1		A	B		Jersey	P _{BREED}
Animals, <i>n</i>	27	30		27	30		57	
Tenderness	51.6	52.3	0.5506	53.3	50.5	0.0510	+7.2	0.0124
Juiciness	54.1	52.1	0.0829	52.4	53.8	0.2998	+0.4	0.8647
Beef Flavor	48.7	48.6	0.6480	48.9	48.3	0.3024	+1.6	0.2039
Browned Flavor	47.4	46.8	0.2872	47.4	46.8	0.3363	+1.7	0.1924
Roasted Flavor	48.9	49.3	0.5063	49.1	49.1	0.9761	+1.3	0.3265
Fat-Like Flavor	10.3	10.1	0.4344	10.2	10.2	0.8799	+0.6	0.4395
Buttery Flavor	0.3	0.9	0.3364	0.8	0.3	0.4345	+0.4	0.7833
Umami Flavor	13.4	13.1	0.4033	13.3	13.2	0.8974	+0.9	0.2091
Metallic Flavor	3.5	3.2	0.6050	3.6	3.1	0.3501	+1.1	0.2958
Oxidized Flavor	2.1	2.2	0.7225	2.3	2.0	0.6750	+0.5	0.5926
Liver-Like Flavor	5.4	4.8	0.5015	5.4	4.8	0.5307	-0.6	0.7186
Sour Flavor	1.1	0.6	0.0714	0.9	0.7	0.5271	-0.4	0.5314

^aAll sensory ratings are on a 0-mm to 100-mm continuous, unstructured line scale.

^bDam breed effect is expressed as effect of 100% Jersey dam compared to 100% Holstein dam

Figure 3.1. Example comparison of steak angularity showing the method of angle determination.

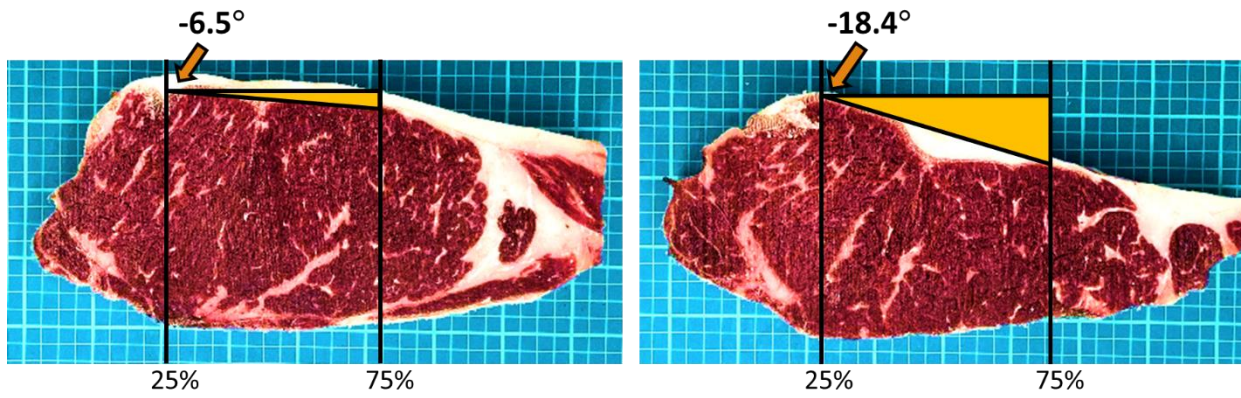
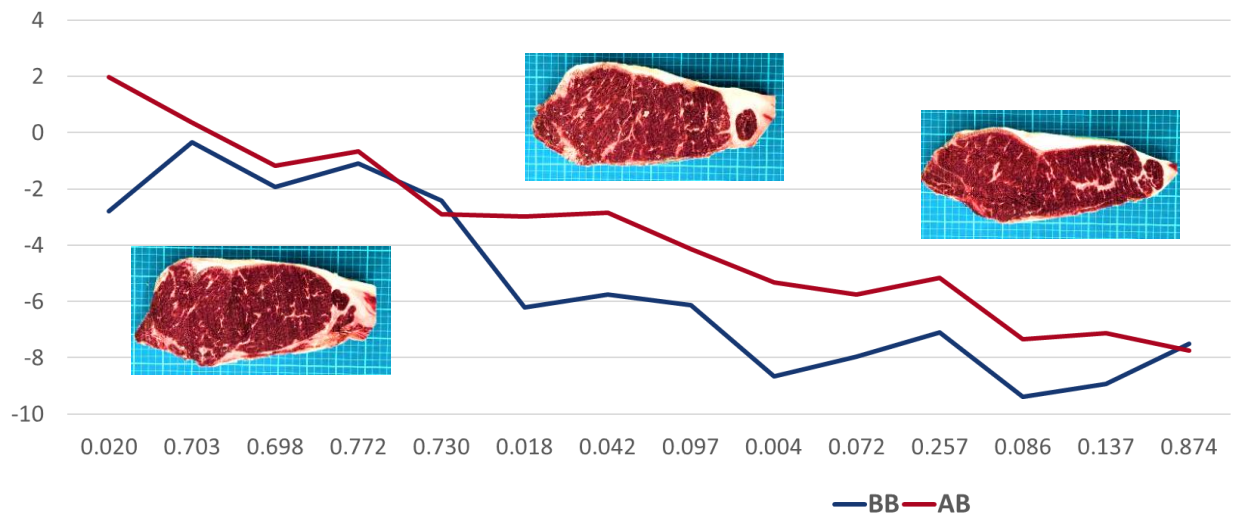


Figure 3.2. Statistical differences between 1-inch steaks cut from anterior to posterior orientation of the entire strip loin among cattle with 0 (BB) or 1 (AB) copies of the F94L mutation.



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