IMPACT OF USING REDUCED-FAT DISTILLERS GRAINS IN BEEF FEEDLOT DIETS ON CARCASS AND MEAT QUALITY

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DEDICATION

This work is dedicated to my wife, Brandi, my family, and all the friends, who have supported me over the years. Although it is my name on the front page:

"We did it."

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ABSTRACT

Purebred Jersey steers (n=12) and Limousin X Jersey Crossbred steers (n=24) were blocked by breed. Nineteen purebred Jersey steers (initial BW 455 ± 49 kg) and 29 Jersey-Limousin crossbred steers (initial BW 518±40 kg) were individually fed in Calan gates for 93 d. Four dietary treatments were evaluated in this experiment (Table 1). A dry rolled corn-based diet served as the control treatment (CON). Distillers grains treatments consisted of feeding reduced-fat distillers dried grains dietary inclusion at 20% with corn oil (RF-O), to represent full fat distillers grains, reduced-fat distillers dried grains dietary inclusion at 20% (RF-L), or reduced-fat distillers dried grains dietary inclusion at 47% (RF-H) of dietary DM. The latter was intended to provide similar dietary fat content as the RF-O treatment. A vitamin and mineral premix containing monensin was added to all diets; a similar premix containing urea was added to the CON diet to meet cattle dietary protein requirements. Cattle were harvested at a commercial abattoir and objective carcass measurements as well as USDA Yield and Quality Grades were collected. Strip loins (IMPS #180) and shoulder clods (IMPS #114) were removed from the right side of each carcass 48 h postmortem. Strip loins were evaluated for purge and drip loss and ultimate pH. Strip steaks were used to determine objective (L*, a*, and b*) and subjective color for six consecutive days. Warner-Bratzler shear force (WBSF) was determined from two steaks from each loin. Fresh strip steaks were cooked for consumer sensory evaluation. Ground shoulder clods were used in ground beef objective and subjective color evaluation. Thiobarbituic acid reactive substance assay was evaluated on ground clods day 0 and day 7. The ground beef was then used in further processed production of bologna. Bolonga

was evaluated for objective and subjective color. A consumer sensory panel evaluated bologna samples. Data were analyzed using the Mixed procedure of SAS 9.3; statistical significance was declared at P<0.05 and trends discussed when P \le 0.10. Hot carcass weight (HCW) was greater (P=0.02) in RF-L compared to CON (388.7 vs. 334.7±12.65 kg). Back fat depth was unaffected (P=0.81) by dietary treatment but tended (P=0.06) to be less in Jersey steers (7.36 vs. 9.65±2.79 mm). Ribeye area (REA) was not impacted (P=0.81) by dietary treatment. However, Jersey steers had smaller (P=0.01) REA (83.94 vs 102.26±7.94 cm²). USDA Yield Grade (YG) was not influenced (P=0.73) by dietary treatment, but Jersey steers had lower (P=0.02) YG (2.69 vs. 2.90 ± 0.06). Steers fed CON tended (P=0.09) to have greater WBSF compared to steers fed RF-O (3.00 vs. 2.24±0.20 kg). Steak objective color (L*) was greater (P=0.03) in steers fed RF-Low than steers fed CON (31.23 vs. 27.04±0.95). Consumers rated the liking of the steak flavor higher for samples from the Crossbred cattle (P=0.03) but preferred the texture of Jersey (P<0.001) strip steaks. On the other hand, consumers their overall liking of the bologna samples from Crossbred steers as compared to Jerseys in overall liking, flavor liking, and texture liking (P<0.001, P<0.001, and P=0.02, respectively). In conclusion, Jersey X Limousin Crossbred steers had greater REA and HCW but no differences in the carcass or meat quality attributes evaluated. Feeding reduced-fat distillers grains in replacement of dry-rolled corn did not substantially affect the carcass or meat quality attributes evaluated.

Keywords: Beef, Jersey, distillers grains, tenderness, color evaluation

CHAPTER 1

REVIEW OF LITERATURE

Various beef breeds around the world have been bred to suit their respective

Breed Impact on Meat Quality

environments to ensure survival. Cattle in continental Europe were developed as first a draft animal and second for meat production. This led to an increase in size of cattle and more heavily-muscled, strong cattle. These breeds produce well yielding carcasses but low marbling (NALF, 2014). Resources in continental Europe were readily available; this allowed the size of the cattle to increase without regulation. Across the English Channel on the British Isles the cattle were quite different. British breeds mature at a lower weight and fatten at a lower weight. Since resources were limited, the cattle there needed to be smaller and require less input. The British breeds are made up of predominantly Hereford, Angus, and Shorthorn. Limousin, Charlois, Simmental, and Chiannia are just a few of the continental breeds. Lastly, there are the dairy breeds. Limousin cattle originated in France over 20,000 years ago, but only more recently in the nineteenth century mankind began developing the breed to the state it is in today (NALF, 2014). They are commonly known for their growth performance, finishing out at a much higher weight than British breeds. According to MacNeil et al. (2001) Limousin cattle marble less than Hereford, but a possibly conflicting study shows that Limousin beef provided a juicier steak sample compared to Angus (Chanbaz et al., 2003). In the same

study Limousin recorded the heaviest hot carcass weight (HCW) at 405 kg compared to Angus at 275 kg. This suggests that Limousin cattle are much later maturing and thus are traditionally marketed at a heavier finished weight.

Jersey cattle originated in England around 1771 (US Jersey, 2010). Jersey cows on average have a 360-550kg mature bodyweight, which is less than Limousin. Jerseys are raised in the U.S. for their high proportion of butterfat in milk (Briggs, 1980). Yet in the dairy industry, aside from a few elite sires, the male calves have very little utility. Jersey beef is known for high marbling (Campion et al., 1976), but growth characteristics are not as efficient as traditional beef species (Bond et al., 1972).

Theoretically, by breeding Jersey cows with Limousin bulls, an efficient and highly marbled carcass can be produced. A study conducted in New Zealand found that Jersey-crossbred cattle (Limousin, Simmental, Murray Grey) grew faster than purebred Jersey (Purchaz et al, 1992). National Agricultural Statistics Service (NASS) archives report that there was a decrease in beef production from 2012-2013 of 200 million pounds. The world population is projected to reach approximately 9 billion people by the year 2050 (United Nations, 2009). One way to address the challenge of feeding the world is to boost efficiency.

Dietary Fat in Beef Production

Fat in beef diets provide a high concentration of energy to enhance growth performance. Many studies have evaluated the difference between tallow and yellow grease. Utilizing yellow grease in beef diets does not affect feedlot performance (Plascencia et al., 1999). Nelson et al., 2008 found beef fed yellow grease to be more tender and contain high

concentration of polyunsaturated fatty acids (PUFA) as compared to tallow. PUFA and monounsaturated fatty acids contain double bonds which more readily allows for oxidation. Oxidation leads to the production of free radicals that eventually produce off odors and flavors in the product (Pearson et al., 1977). Studies with monogastrics has found that the type of dietary fat source has varying effects on the processing qualities of meat (Xu et al., 2010 and Lee et al., 2012). Addition of dried distillers grain with solubles (DDGS) to pork diets reduces belly firmness and increases iodine value (IV). This leads to reduced production from the belly (Leick et al., 2010). Conflicting data suggests that feeding DDGS improves belly firmness, but the experiment reduced the level of DDGS in the last phase to half the previous levels (Browne et al., 2013).

Biohydrogenation is a process in which the microbes in the rumen of cattle saturate PUFA and MUFA. This process greatly reduces the effect of diet on fatty acid profile in ruminant animals (Jenkins et al., 2008).

Distillers Grains as a Feedstuff

The beef industry utilizes DDGS as a low cost alternative to traditional feedstuffs (RFA, 2011). Traditional Midwest beef diets consist of corn/corn silage and a protein supplement. Since the early 2000's the demand on traditional feedstuff has increased dramatically (USDA, 2014).

The drive to produce biofuels such as ethanol and biodiesel has driven the price of commodities such as corn and soybeans up (Federal Reserve, 2009). Corn production and prices have been on a steady incline since 1988, the price per bushel in 1988 was approximately \$2.50 compared to \$7 per bushel in 2012 (USDA, 2014). At the same

time production levels have increased in the U.S. from 8 billion bushels to near 15 billion bushels over the same time frame (Childs, 2013). Animal research in recent years has turned to utilizing alternative feedstuffs, including many forms of DDGS (Leupp et al., 2009; Stein et al., 2009), bakery waste (Rojas et al., 2013; Guiroy et al., 2000), and glycerin (Gunn et al., 2010; Hales et al., 2013; Mendoza, 2010).

Distiller's grains have been available for as long as the beverage industry has been distilling grains for alcohol production. Distillers only utilize the starches in the grain for the production of alcohol, therefore the protein, fat, and fiber was traditionally considered waste for a large part of history. Eventually the waste from the distilling process was used as livestock feed, albeit a localized product. Prior to the fuel ethanol boom the only source of distiller's grains were breweries and spirit distilleries. Thus, the amount of distiller's grains available were very limited. The fuel ethanol boom in the U.S. can be credited with producing a readily available supply of various distillers' grains, especially in the corn belt of the U.S. As of January, 2014 there were 216 ethanol plants in operation in the United States (*Ethanol Producer Magazine*, 2014). Early in fuel ethanol production the only product available was wet distiller's grains. Then, distilleries began drying the by-product to produce dried distillers grains and adding back the solubles to create dried distillers grains with solubles.

Alternative Processing of DDGS

The ethanol industry has undergone several swings in the market over the past decade. In late 2008 the price per gallon of ethanol was approximately \$1.40, while corn was hitting historical highs (Obrien et al., 2009 and Wisner, 2009). The volatility of oil demand and

prices has pushed ethanol producers to become more efficient with all products and byproducts. One example of an area of opportunity and expansion for the ethanol industry was in the dairy industry. The high levels of unsaturated fats found in distillers grains has kept the product largely out of dairy diets for years, but with newer ethanol processing methods such as post-fermentation extraction, there may be opportunity. This provides a new product on the market for not only dairy producers but all livestock producers.

Reduced Fat Distillers in Beef

To understand reduced-fat distiller's grains, production of full-fat distiller's grains must be understood. The most common grain used for ethanol production is corn. Upon receiving, the plant grinds the corn one of three ways: wet milling, dry milling, and dry grinding (Kingsly et al., 2009 and Singh et al., 2001). According to the *Renewable Fuels Association*, dry milling is the most common method used in the midwest. Singh et al. (2001) lays out the 5 steps for ethanol production using the dry milling process: grinding, liquefaction, heating, saccharification and fermentation. During the fermentation process ethanol is produced from the sugar/starches with the help of yeast. The product undergoes distillation that removes the ethanol and leaves a slurry. The stillage is made up of two parts: thin stillage and wet distiller grains (WDG). The thin stillage is allowed to dry into a syrup known as condensed distiller's solubles (CDS) and WDG can be sold as-is or dried. That is how dried distillers grains (DDG) is produced. If the CDS is added back to the DDG then it is referred to as dried distiller's grains with solubles (DDGS).

DDGS: centrifugation or use of solvents. Both of these methods are designed to remove oil. The net return on a gallon of ethanol in January of 2013 was less than twenty five cents, which has led ethanol producers to look for alternative values in their processes (Shurson, 2013). The results include a high fiber and high protein feedstuff. This may be advantageous in the attempt to reduce unsaturation in the fatty acid profile of the feedstuff.

DDGS Effect on Beef Quality

Fresh Beef

Color

Meat color is a combination of the expression of heme iron (myoglobin) and marbling. Oxidation of both the lipids and myoglobin affect both subjective and objective color. Myoglobin is a catalyst in lipid oxidation (Johns et al., 1989). At the onset of shelf life evaluation, the myoglobin is in the oxymyoglobin state. Exposure to light and atmospheric oxygen induces oxidation. Heme iron is found in two states in meat: ferrous (2+) and ferric (3+). Ferrous heme iron can be seen in two main states either deoxymyoglobin (purple) or oxymyoglobin (bright red;(Gray et al., 1996). During oxidation an electron is scavenged from the heme iron moving it to ferric state. This is where metmyoglobin is represented with a dull brown color in meat retail display (Greene and Price, 1975).

When evaluating the effect of DDGS on beef color, Leupp et al. (2009) found that the a* value, or redness, was reduced compared to a control diet. There was also a difference in a* values when comparing corn DDGS and sorghum DDGS (Gill et al., 2008). On the other hand, feeding

up to 75% DDGS, Gordon et al. (2002) found no difference in a* and b* (yellowness) values. Several more studies show quite a contradiction of DDGS effect on meat color, thus the effect of DDGS on color is not fully understood (Depenbusch et al., 2009, Roeber et al., 2005, and Aldai et al., 2010). At levels of 20-40% WDGS and DDGS, Koger et al. (2010) found no difference in discoloration and lean color scores on ground beef patties.

Lipids

Current trends in human nutrition have driven consumers away from fat as a dietary energy source due to historical over consumption which leads to obesity (CDC, 2012). Oxidative stability is greatest in saturated fats and lowest in PUFA. Oxidation of fatty acids induces off flavors in all forms of meat products, especially further processed products. Further processing (grinding, mixing and emulsifying) abuses fatty acids through temperature and mechanical action.

Saturated fats are characteristically associated with animal fats (Valsta et al., 2005). The composition of saturated fats allows for the most dense energy rich nutrient in animal diets. Fatty acid profile of meat is primarily affected by the diet of the animal (Wang et al., 2012 and He et al., 2014).

Feeding higher levels of DDGS increases the concentration of PUFA consumed. Although not at the levels found in pork, PUFA concentrations do increase in beef (A.S. Mello et al., 2012). More specifically there is a common trend of finding high levels of 18:2 linoleic acid when feeding DDGS (Gill et al. 2008). In a study evaluating trans 18:1, Aldai et al. (2010) found the level of saturated fats to be unaffected by DDGS supplementation, but PUFA were higher in the DDGS diets and MUFA were higher in the control diets. Another study found that replacing barley silage with WDGS increased total PUFA and omega-3 levels (He et al., 2012).

Tenderness

Tenderness is one of the most important factors affecting meat quality. Factors that determine tenderness are found throughout the animal production cycle and include genetics, age (Weston et al, 2002), and proteolytic degradation of the z-line (Taylor et al., 1995). An important factor in tenderness is the amount of collagen and more importantly the level of collagen cross linking that has taken place within the muscle. Cross linking is affected by several factors. The older the animal, the more cross-linking. Location of the muscle also affects cross-linking. Muscles of locomotion, for instance in the leg, will have a tougher collagen matrix than a muscle of posture (Weston et al, 2002).

Tenderness is evaluated in two ways, the first of which is instrumental testing (Warner-Bratzler Shear Force) which evaluates a cooked core from a steak that runs parallel with the muscle fiber and then measures the force required to shear the core. The second evaluation is sensory testing. But the correlation between the two is not necessarily strong (Wezememael et al, 2014). This is most likely due to levels of perception in the human panel. Shear force gives an absolute value, but various factors may affect the perception to the consumer such as degree of doneness, marbling, and breed (Mckenna et al, 2004). Inclusion of DDGS in beef diets have shown an increase of tenderness and palatability. According to Aldai et al (2010) a control barley based diet recorded the highest number of objectively tough shear values when compared to wheat and corn DDGS.

Sensory Characteristics

Sensory science considers the perception of a human panelist compared to the objective physical characteristics of a food product (Lawless and Heymann, 2010). Due to the variability in human

perception, there are many ways to induce bias into sensory evaluation (Lawless and Heymann, 2010, Resurreccion, 1998, and Meilgaard et al., 1998). Trained and untrained panels vary in their evaluation of a product. When the evaluation of a product is compared to the chemical composition and cooking factors, a link can be made between uncooked products and potential consumption quality. According to Meilgaard et al. (2007) the flavor is the sum of sensations at the beginning of the respiratory and alimentary tracts. Texture is evaluated by other receptors in the mouth other than the taste sensors (Lawless and Heymann, 2010). Texture is another aspect of sensory evaluation, combining reaction to stress and tactile feel. Mechanical properties of reaction to stress is evaluated many different ways including gumminess, springiness, hardness, and many more (Meilgaard et al., 2007). Resurreccion (1998) defined tactility as a measure of geometrical or moisture properties, dry, oily, gritty, flaky grainy, and wet.

Contrary to objective color data, sensory characteristics are generally improved with the addition of DDGS or WDGS to beef diets. Depenbusch et al. (2009) showed DDGS fed at 40-65% recorded the strongest beef flavor intensity. A comprehensive evaluation by Gill et al. (2008) found no difference between corn DDGS, sorghum DGS and a corn control for juiciness and off flavors. The same study found the most tender steaks came from corn DDGS over sorghum DGS. Aldai et al. (2010) reported similar results with wheat DDGS.

Conclusion

Combining the high growth performance of the Limousin breed and the marbling advantages of Jersey cattle may improve beef quality and efficiency. The use of distillers grains in beef feedlot diets is ever increasing, leading to a potential change in overall composition of the subsequent beef from these animals. Currently, beef cow numbers are

at their lowest number in decades, utilizing the resources at hand: DDGS and cooperation with the dairy industry, beef production can continue to fulfill demand.

CHAPTER 2

IMPACT OF USING REDUCED FAT DISTILLERS GRAINS IN BEEF FEEDLOT DIETS ON CARCASS AND MEAT QUALITY

INTRODUCTION

Limousin cattle originated in France over 20,000 years ago, but only more recently in the nineteenth century mankind began developing the breed to the state it is in today (NALF, 2014). They are commonly known for their growth performance; finishing out at a much higher weight than British breeds. According to MacNeil et al. (2001) Limousin cattle marble less than Hereford, but a possibly conflicting study demonstrated that Limousin beef provided a juicier steak sample compared to that of Angus beef (Chanbaz et al., 2003). In the same study Limousin had the heaviest hot carcass weight (HCW) at 405 kg compared to Angus at 275 kg. This suggests that Limousin cattle are much later maturing and thus are traditionally marketed at a heavier finished weight.

Jersey cows on average have a 360 to 550kg mature bodyweight,. Jersey are raised in the U.S. for their high proportion of butterfat in milk (Briggs, H, 1980). Yet in the dairy industry, aside from a few elite sires, the male calves have very little utility. Jersey beef is known for high marbling (Campion et al., 1976), but growth characteristics are not as efficient as traditional beef species (Bond et al., 1972). Theoretically, by breeding Jersey cows with Limousin bulls, an efficient and highly marbled carcass can be produced. A study conducted in New Zealand found that Jersey-crossbred cattle (Limousin, Simmental, Murray Grey) grew faster than purebred Jersey (Purchaz et al, 1992).

NASS archives report that there was a decrease in beef production from 2012 to 2013 of 200 million pounds. The world population is projected to reach approximately 9 billion

people by the year 2050 (United Nations, 2009). One way to address the challenge of feeding the world is to boost production efficiency.

Fat in beef diets provide a high concentration of energy to enhance growth performance. Fatty acid profile of meat is primarily affected by the diet of the animal (Wang et al., 2012 and He et al., 2014). Feeding higher levels of DDGS increases the concentration of PUFA consumed. Although not at the levels found in pork, PUFA concentrations do increase in beef (A.S. Mello et al., 2012). More specifically there is a common trend of finding high levels of 18:2 linoleic acid when feeding DDGS (Gill et al. 2008). In a study evaluating trans 18:1, Aldai et al. (2010) found the level of saturated fats to be unaffected by DDGS supplementation, but PUFA were higher in the DDGS diets and MUFA were higher in the control diets. Another study found that replacing barley silage with WDGS increased total PUFA and omega-3 levels (He et al., 2012). Tenderness is one of the most important factors affecting meat quality. Factors that determine tenderness are found throughout the animal production cycle include genetics age (Weston et al, 2002) and proteolytic degradation of the z-line (Taylor et al., 1995). Muscles of locomotion, for instance in the leg, will have a tougher collagen matrix than a muscle of posture (Weston et al, 2002). Contrary to objective color data, sensory characteristics are generally improved with the addition of DDGS or WDGS to beef diets. Depenbusch et al. (2009) demonstrated the DDGS fed at 40-65% yielded the strongest beef flavor intensity. A comprehensive evaluation, Gill et al. (2008) found no difference between corn DDGS, sorghum DGS and a corn control for juiciness and off flavors. In same study the most tender steaks came from corn DDGS over sorghum DGS. Aldai et al. (2010) reported similar results with wheat DDGS. Combining the high growth

performance of the Limousin breed and the marbling advantages of Jersey cattle may improve beef quality and efficiency. The use of distiller's grains in beef feedlot diets is ever increasing, leading to a potential change in overall composition of the subsequent beef from these animals. With beef cow numbers at their lowest number in decades, utilizing DDGS and cooperation with the dairy industry, beef production can continue to fulfill demand.

Material and Methods

Dietary Treatments

Animal management procedures were approved by the University of Minnesota Animal Care and Use Committee.

Nineteen purebred Jersey steers (initial BW 455 ± 49 kg) and 29 Jersey-Limousin crossbred steers (initial BW 518 ± 40 kg) were arranged in a generalized randomized complete block design. Steers were blocked by breed and allotted randomly to 1 of 4 pens.

Steers were individually fed in Calan gates (American Calan, Inc., Northwood, NH) for 93 d. Four dietary treatments were evaluated in this experiment (Table 1). A dry rolled cornbased diet served as the control treatment (CON). The remaining treatments contained various concentrations of reduced-fat distillers dried grains partially replacing dry rolled corn. A single, reduced-fat distillers grains source was used, and where necessary, corn oil was added to the diet at mixing to achieve the lipid content of full-fat distillers grains. Distillers grains treatments consisted of reduced-fat distillers dried grains dietary inclusion at 20% with corn oil (RF-O), reduced-fat distillers dried grains dietary inclusion at 20% (RF-L), or reduced-fat distillers dried grains dietary inclusion at 47% (RF-H) of dietary DM. The latter was intended to provide similar dietary fat content as the RF-O treatment. A vitamin and mineral premix containing monensin was added to all diets; a similar premix containing urea was added to the CON diet to meet cattle dietary protein requirements.

Carcass Data Collection

Upon completion of the finishing phase, steers were transported in one group approximately 440 km to a commercial abattoir (Tyson Foods, Dakota City, NE). Fortyeight h postmortem, hot carcass weight (HCW), 12th rib back fat, percent kidney pelvic heart fat (KPH) and ribeye area (REA) were collected. Marbling score, USDA Yield Grade and USDA Quality Grade were evaluated by a USDA grader and recorded.

Fresh Beef Fabrication and Collection

Fresh beef products were fabricated 96 h post-mortem, according to Institutional Meat Purchasing Specifications (IMPS). Strip loins (IMPS #180) and shoulder clods (IMPS #114) were removed 52 h postmortem from the right side of the carcass, and individually identified using carcass identification tags crossbred-referenced to animal identification tags during harvest. Strip loins and shoulder clods were vacuum packaged and maintained at 2° C during transport to the Andrew Boss Laboratory of Meat Science at the University of Minnesota, (St. Paul, MN). All beef products were inspected for vacuum seal, re-packaged if necessary, and shoulder clods were placed in a blast freezer (-20° C) until further evaluation. The strip loins were processed upon return to the University of Minnesota 72H post-mortem.

Strip Loin Sample Preparation

Strip loins were faced perpendicular to the length of the loin, and steaks were serially cut, 2.54cm thick, from the anterior end of each strip loin. The first steak was designated for drip loss analysis, the second and third were designated for shelf-life analysis. The fourth through seventh steaks were designated for sensory analysis and the eighth and ninth

steaks were designated for Warner Bratzler shear force (WBSF) evaluation. A 10g back fat sample was collected from the posterior end of each strip loin before cutting steaks, vacuum packaged, and stored frozen (-20° C) until processing for fatty acid profile analysis.

Shoulder Clod Preparation

Shoulder clods, (approximately 9.5 kg each) were thawed (vacuum packaged) at 4° C for 3 d. Entire, untrimmed shoulder clods were ground twice (Cabela's Electric Meat Grinder, Model: 32, Kearney, NE) with a 0.375 cm plate

Retail Display

Two steaks were placed on polystyrene trays with polyvinylchloride (PVC) overwrap (oxygen transmission rate 1400 cc/m2) and stored at 4° C under cool white fluorescent lighting (Sylvania H968, 100w, 2, 640 LUX) for seven days. Objective color values (CIE, L^* , a^* , and b^*) were taken at six locations on each steak (Hunter Lab Miniscan EZ model 4500S, Reston, VA).

For ground beefretail display, one 225g (\pm 5g) patty per clod was placed on a polystyrene tray with polyvinylchloride (PVC) overwrap (oxygen transmission rate 1400 cc/m²) and stored at 4° C under cool white fluorescent lighting (Sylvania H968, 100w, 2, 640 LUX) for seven days. Objective color values (CIE, L^* , a^* , and b^*) were taken at six locations on each patty with a Hunter Lab Miniscan EZ (model 4500S, Reston, VA).

Subjective color scores (lean color, surface discoloration, and overall appearance) were evaluated by an eight-person trained panel for seven days for both steaks and ground patties. Lean color was evaluated on a 1 to 8 scale with 1 = extremely brown and 8 = extremely brown

extremely bright, cherry red. Surface discoloration was evaluated on 1 to 11 scale with 1 = 91-100% discoloration and 11 = 0% discoloration. Overall appearance was evaluated on a 1 to 8 scale with 1 = extremely undesirable and 8 = extremely desirable (AMSA, 1991).

Moisture Loss

Drip loss was evaluated for each steak (approximately 158 g) by suspending steak samples for 24 h at 4° C in a sealed Ziploc® bag wrapped loosely. Percent drip loss was calculated as the difference between the initial (product and moisture minus the dried bag) and final weight (unpacked and patted dry) divided by the initial weight multiplied by 100. Vacuum-packaged purge loss of the strip loin and shoulder clod was measured after transport and before further fabrication. Purge loss was calculated as the difference between the initial and final weight divided by the initial weight multiplied by 100.

Warner-Bratzler Shear Force (WBSF)

Duplicate steaks were thawed for 24 h at 4° C, individually wrapped in aluminum foil, and cooked at 180°C, using a George Foreman Indoor/Outdoor Grill (Model: GGR62. Lake Forest, IL) to an internal temperature of 71° C as indicated by a probe placed at the geometric center of the steaks (Type T thermocouple, Omega Engineering, Stanford, OH). Steaks were stored refrigerated (2° C), equilibrated to room temperature (25° C), and six, 1.27-cm diameter cores were removed from each steak parallel to the muscle fiber by a hand corer. Each core was sheared on a texture analyzer fitted with a Warner-

Bratzler shear force attachment (Shimatzu Texture Analyser, Model: EZ-SX, Kyoto, Japan). Six cores were sheared per steak to represent the entire surface of the *longissimus* dorsi muscle.

Fresh Beef Sensory Evaluation

Procedures utilizing human subjects for consumer panel evaluation of sensory attributes were approved by the University of Minnesota Institutional Review Board. The University of Minnesota Food Science and Nutrition Sensory Center recruited eightynine untrained consumer panelists for sensory evaluation of fresh strip steaks. All panelists were 18 years of age or older, had no food allergies, and consumed steak at least twice per month. Panelists were paid \$5 for their time. Sensory evaluation was conducted by the University of Minnesota Food Science and Nutrition Sensory Center following the research guidelines for sensory evaluation (AMSA, 1995).

Steaks were thawed for 36 h at 4° C, individually wrapped in aluminum foil, cooked at 180° C (General Electric® Range, JASO2; Fairfield, CT), to an internal temperature of 71° C as indicated by a probe place at the geometric center of the steak (Pyrex Professional Acu rite Thermometer; Racine, WI). Steaks were cut into 1-cm x 1-cm x 2.54 cm cubes and placed in the top portion of double boilers containing water in the bottom portion heated to ~82° C (replaced every h) to keep samples warm. Each panelist received two pieces of steak per sample (approximately 38° C) in lidded 60 ml plastic soufflé cups coded with random 3-digit numbers. To maintain sample serving temperature, the cups were nested in heated sand (~60° C) contained in round, aluminum pans. Samples were served to subjects balanced for order and carryover effects. Subjects

were asked to taste one piece of the sample and rate it for overall liking, liking of flavor, liking of texture, and off flavor intensity. Subjects were then instructed to taste the second piece and rate the intensity of toughness and juiciness. Liking ratings were made on 120-point labeled affective magnitude scales, with the left most end labeled *strongest dislike imaginable* and the right most end labeled *strongest like imaginable*. Intensity ratings were made on 20-point line scales with the left most ends labeled *none* and the right most ends labeled *extremely intense* for off flavor, *extremely tough* for toughness, and *extremely juicy* for juiciness.

TBARS

Samples (10 g) of each ground beef batch were collected on days 0 and 7 for analysis, vacuum packaged, and stored frozen (-20°C) immediately for thiobarbituric acid reactive substances (TBARS) analysis (AOCS, 1998). Secondary lipid oxidation products were measured using the thiobarbituric acid assay (Tarladgis et al. 1960). All samples were transported to Agricultural Utilization Research Institute (AURI, Marshall, MN) for analysis. Samples were evaluated in duplicate and measured with a spectrophotometer (Spectronic 20+, Spectronic Instruments, Inc.) at 532 nm.

Bologna Preparation

Meat blocks were created by combining clods from four animals per dietary treatment.

11.34 kg of ground beef from the combined meat blocks were then combined with a commercial bologna seasoning blend (Bologna SCTP, Newly Wed Food, Chicago, IL),

1.13 kg (2.5 lbs) of ice, sodium tripolyphosphate (30 g per batch), and sodium nitrite cure

30 g per batch (Heller's Modern Cure #47688, Newly Wed Food, Chicago, IL). Ground beef and ingredients were emulsified (Alipina, PB 80-890-II Gossau S G, Switzerland, Speed setting 2, 3-knife head with Alipina tangential form blades) to 10° C and then stuffed (Handtmann VF-608, Albert Handtmann Maschimen Fabrik GmbH & Co., Biberach, Germany) into inedible collagen casings (Bologna 10.8 cm Walsrober Casings, Mar/Co Sales, Burnsville, MN). Bologna was cooked to an internal temperature of 65.5° C, (Enviro-Pak, Model CVU 500E-IT, Portland, OR), cooled (12 hours) to 4° C and then sliced. Slices were 12-cm in diameter and 4-mm thick (Globe Slicer, Model 400, Globe Slicing Machine Co, Inc., Stamford, CT).

Processed Beef Retail Display

One slice of bologna from each batch was placed on a polystyrene tray, placed in a vacuum bag (3mil standard barrier, Bunzl PD, North Kansas City, MO) sealed and stored at 4° C under cool white fluorescent lighting (Sylvania H968, 100w, 2, 640 LUX) for ten days. Objective color values (CIE, L^* , a^* , and b^*) were taken at six locations on each slice with Hunter Lab Miniscan EZ (model 4500S, Reston, VA). Subjective color scores (lean color, surface discoloration, and overall appearance) were evaluated by an eight person trained panel for ten days, every other day. Lean color was evaluated on a 1 to 8 scale with 1 = extremely brown and 8 = extremely bright, cured pink. Surface discoloration was evaluated on 1 to 11 scale with 1 = 91-100% discoloration and 11 = 0% discoloration. Overall appearance was evaluated on a 1 to 8 scale with 1 = extremely undesirable and 8 = extremely desirable (AMSA, 1991).

Processed Beef Sensory Evaluation

The University of Minnesota Sensory Center conducted sensory evaluation for bologna. Panelists were untrained consumers that were over 18 years old, had no food allergies, and had consumed beef at least twice per month. Panelists were paid for their participation. The University of Minnesota's Institutional Review Board approved all recruiting and experimental procedures. Bologna slices were cut into eight sections and each untrained consumer panelist (n = 87) received two pieces for each replication with three replications per treatment, stored refrigerated, and then served at room temperature. Samples were served to subjects balanced for order and carryover effects. Subjects were asked to taste one piece of the sample and rate it for overall liking, liking of flavor, and liking of texture. Liking ratings were made on 120-point labeled affective magnitude scales, with the left most end labeled strongest dislike imaginable and the right most end labeled strongest like imaginable. Panelists were then instructed to taste the second piece and rate the toughness and off flavor; these ratings were made on 20-point line scales with the left most ends labeled *none* and the right most ends labeled *extremely tough*, and extremely intense, respectively

Statistical Analyses

Statistical analysis for USDA Quality and Yield Grade categorical data was performed using the GENMOD procedure of SAS (SAS Inst., Inc, Cary, NC; Appendix G). Steer was the experimental unit and the model included dietary treatment as the fixed effect. Type 3 fixed effects were used to determine significance (P < 0.05) or trends (P < 0.10) among treatments. The PDIFF option was used to separate least squares means when a

significant F-test statistic was present. Treatment means are presented as least squared means, and weighted standard errors were calculated as: $\Sigma(\text{error*degrees of freedom})/\Sigma(\text{total degrees of freedom})$.

Mixed model analysis of variance (PROC MIXED procedure of SAS; Appendix H) was used to analyze hot and cold carcass weight, 12th rib back fat, KPH, ribeye area, drip loss, purge loss, fabrication loss, and WBSF. The Restricted Maximum Likelihood (REML) procedure was used to estimate the variance components and the Kenward-Rogers procedure was used to determine degrees of freedom approximation. Steer was the experimental unit and the statistical model included dietary treatment as the fixed effect. For those variables with significant (P < 0.05) ANOVA, mean separations were performed using the LSMEANS and PDIFF functions of SAS. Weighted standard errors were calculated as: $\Sigma(\text{error*degrees of freedom})/\Sigma(\text{total degrees of freedom})$. For sensory analysis, the PROC MIXED procedure of SAS was used to determine if samples differed in any of the attributes. Overall liking, flavor liking, texture liking, off flavor, toughness, juiciness (steaks), were dependent variables and product, replicate and product*replicate were predictors. Subject and subject*product were random predictors in the models. Bonferonni correction was used to determine if specific differences among samples were significant.

RESULTS AND DISCUSSION

Carcass Data

Hot carcass weight varied significantly by dietary treatment with CON recording the lowest value (334 kg) and RF-O and RF-L having the highest (379 and 388kg, respectively, P<0.05). There were no differences among dietary treatments for *longissimus* muscle area (LMA), kidney, pelvic, and heart fat (KPH), 12th rib back-fat (BF), and USDA Yield Grade (YG). All dietary treatments were more likely to grade USDA Choice when compared to the control (P = 0.04). RF-O and RF-L achieved higher marbling scores compared to CON (P = 0.05). HCW (P = 0.0001), LMA (P = 0.0001), BF (P = 0.05), marbling score (P = 0.05), and YG (P = 0.05) (Table 2) were all affected by breed. Jersey carcasses recorded LMA of 83.25cm² and 101.4cm² for the crossbred steers. Crossbred steers had 80 kg heavier for HCW (P < 0.01). Of note, no difference was found in USDA Quality Grade (P=0.87) among breeds. The crossbred steers marbled higher than Jersey steers (P=0.05). (Table 3)

Moisture Loss

Difference in moisture loss across dietary treatment and breed were not statistically significant. There was a trend in differences in drip loss between breeds; Jersey averaged 1.13% loss while Crossbred measured 0.85% loss (P = 0.09). (Table 4 & 5)

Warner Bratzler Shear Force

Warner Bratzler Shear Force (WBSF) was not affected by breed (P=0.09). CON steaks required more force to shear the cores when compared to all other dietary treatments (P = 0.09). The WBSF values range from 2.24 kg for the RF-H group and 2.71 kg for the RF-O. (Table 4 & 5) Similar results were seen in studies by Aldai et al. (2010) and Koger et al. (2010) when evaluating corn and wheat WDGS and DDGS at 20-40% in the diet. Shackelford et al, (1991) reported consumer tenderness perception between 3.9 and 4.6 kg of force for slightly tender beef. With that understanding all the values in this study fall into slightly tender or more tender. Rober et al (2005) and Gill et al (2008) found shear force below consumer threshold when cattle were fed distillers grains.

Sensory Evaluation of Fresh Strip Steaks

Overall liking did not differ between the two breed treatments (P = 0.92). However, panelists preferred the flavor of steaks from the Crossbred cattle (P = 0.001) and the texture of steaks from the Jersey cattle (P = 0.03). Jersey steaks were tougher (P = 0.001) yet juicier (P = 0.001).

Steaks from animals fed the RF-H diet were rated higher as compared to those of the animals fed the RF-L diet for texture liking (P=0.04) and flavor liking (P=0.01).

Panelists preferred the flavor liking of steaks from cattle fed the RF-H and CON diet compared to RF-L. (Table 6) Panelists preferred the texture of the steaks from the steers fed the RF-H to the texture of those from steers fed the RF-L diet. The steaks from the steers fed the CON and RF-O diet were tougher than the steaks from the cattle fed the RF-H diet. Luepp et al. (2009) reported no differences in tenderness, juiciness and flavor

in steaks from steers fed 30% DDG in the finishing diet. Haack et al (2011) fed cattle wet distillers grains with solubles at varying concentrations of fat. No differences were found in beef flavor intensity and juiciness. Haack et al (2011) also found control and 4.72% fat WDG beef less tender, with lower levels recorded for off-flavors than 6.91% fat wet distillers grain diet. In the current study there were no differences in off-flavor (P = 0.28) of steaks across breed and dietary treatments.

Sensory Evaluation of Bologna

Overall liking was greater for bologna from Jersey cattle (P = 0.001). Panelists rated higher the flavor (P = 0.001) and the texture (P = 0.03) of bologna from Crossbred as compared to Jersey cattle. Jersey bologna was tougher (P = 0.01). There was no difference in the overall liking and flavor liking of bologna by dietary treatment

Thiobarbituric Acid Reactive Substances (TBARS)

Dietary treatment had no effect on TBARS for ground beef for day 0 or Day 7 (P = 0.96, 0.96). Ground beef from Crossbred steers had higher TBARS levels on day 0 and day 7 (P = 0.001, 0.001, respectively; figures 37 & 38).

Objective and Subjective Color Evaluation of Fresh Strip Steaks

Subjective color evaluation, by dietary and breed treatment, was different for lean color (P=0.01) and a trend was shown for surface discoloration (P=0.09) for dietary treatment.

L* values for RF-L were higher than CON and RF-O on day two (P = 0.001, 0.03, respectively) and three (P = 0.004, 0.01, respectively). Breed had no effect on L* values (P = 0.24). The a* values for strip steaks were unaffected by diet (P = 0.63) but Jersey steaks were more red (a*) on days two (P = 0.003) and three (P = 0.014). On days three (P = 0.05) and four (P = 0.01), steaks from Jersey cattle(11.6, 12.7) had higher b* values than the steaks from crossbred cattle (10.3, 11.0) (Figures 1-12).

Objective and Subjective Color Evaluation of Ground Patties

Starting on day three (P = 0.02) and day four (P = 0.007) lean color was affected by breed (means). CON recorded the highest values from day 2 until the conclusion of the study, for all three subjective parameters (Figures 13-24).

Objective and Subjective Color Evaluation of Bologna

There was no effect of dietary treatment on subjective surface discoloration (P = 0.21) and overall appearance (P = 0.57), however a trend was found for lean color (P = 0.07). Breed affected surface discoloration (P = 0.0002) and overall appearance (P = 0.006). There was a breed effect on objective L* values (P = 0.01; Figures 25-36).

4. CONCLUSION

Feeding reduced fat distillers grains does not negatively impact fresh or processed beef quality, and potentially improves carcass characteristics including, HCW, marbling score, and USDA Quality Grade. Crossbreeding Limousin bulls with Jersey cows improves HCW and LMA without a decline in USDA Quality Grade.

TABLES

Table 1. Dietary treatments and inclusion by replacement of dry rolled corn						
(DRC)-silage-based control (CON) diets						
Treatment Dietary Inclusion Crude						
	-	Fat%				
Control (CON)	Dry rolled corn (DRC)/silage	3.3				
Reduced fat low (RF-L)	20% Reduced fat distillers grains	4.1				
Corn oil (RF-CO)	20% Reduced fat distillers grains plus 1%	5.1				
	corn oil to simulate full-fat DG					
Reduced fat (RF-H)	46% Reduced fat distillers grains at same	5.0				
	dietary fat as RF-CO					

Table 2. Least squared means for carcass traits displayed by dietary treatment							
	Treatmen	nt					
	С	RF-O	SE	P value			
Hot carcass weight, kg	334.74	379.29	388.70	357.07	12.65	0.02	
Longissimus muscle area, cm2	90.44	95.18	91.59	92.10	2.86	0.67	
Kidney, pelvic and heart fat, %	2.34	2.40	2.65	2.38	0.12	0.31	
12th rib backfat, cm	0.74	0.83	0.92	0.79	0.10	0.66	
USDA Yield Grade	2.73	2.82	2.90	2.78	0.10	0.66	
USDA Quality Grade	2.50	2.08	2.09	2.13	0.13	0.04	
Marbling score	416.25	508.50	538.33	468.75	31.27	0.05	

Table 3. Least squared means for carcass traits by breed							
	Trea	atment					
Carcass Characteristic	Jersey	Crossbred	SE	P value			
Hot carcass weight, kg	324.99	404.91	8.88	< 0.0001			
Longissimus muscle area, cm2	83.25	101.40	2.01	< 0.0001			
Kidney, pelvic and heart fat, %	2.36	2.53	0.09	0.17			
12th rib backfat, cm	0.72	0.92	0.07	0.05			
USDA Yield Grade	2.70	2.91	0.07	0.05			
USDA Quality Grade	2.21	2.19	0.10	0.87			
Marbling score	469.67	496.67	21.98	0.05			

Table 4. Least squared means for vacuum purge, Ultimate pH, drip loss, and Warner-Bratzler shear force by dietary treatment Treatment RF-L C RF-O RF-H SE P value Purge, % 0.01 0.01 0.69 0.01 0.26 0.20 0.20 pН 5.51 5.49 5.07 5.48 0.16 Drip, % 1.334 0.173 0.066 0.874 0.168 1.062 Shear Force 2.71 0.20 3.01 2.69 2.24 0.09

Table 5. Least squared means for vacuum purge, Ultimate pH, drip								
loss, and Warner-Bratzler shear force by breed treatment								
Treatment								
	Jersey Crossbred SE P value							
Purge, %	0.35	0.02	0.18	0.21				
pН	5.28 5.48 0.11 0.2							
Drip, %	1.13	0.85	0.12	0.09				
Shear Force	2.79	2.96	0.14	0.21				

Table 6. Least squared means for sensory characteristics of fresh strip steaks by dietary treatment								
		Treat						
	C RF-O RF-L RF-H F I							
Overall Liking	68.0 ^{ab}	67.0ab	65.0b	70.0^{a}	2.90	0.03		
Flavor Liking	70.0a	68.0ab	65.0b	72.0a	5.30	0.00		
Texture Liking	67.0ab	66.0ab	64.0b	70.0a	2.50	0.06		
Off flavor	7.9a	7.5a	7.2a	6.5a	1.30	0.28		
Toughness	5.5a	5.5a	5.5 ^{ab}	5.2b	4.10	0.01		
Juiciness	4.6a	4.4a	5.1a	4.8a	0.40	0.75		

Table 7. Least squared means for sensory characteristics of fresh strip								
steaks by breed treatment								
	Treatment							
	Jersey Crossbred SE P value							
Overall Liking	67.00	0.92						
Flavor Liking	70.00	0.03						
Texture Liking	64.00 70.00 13.00 <0.0							
Off flavor	7.80 6.70 2.90 0.09							
Toughness	4.50 6.30 12.20 <0.001							
Juiciness	4.50	5.00	40.90	< 0.001				

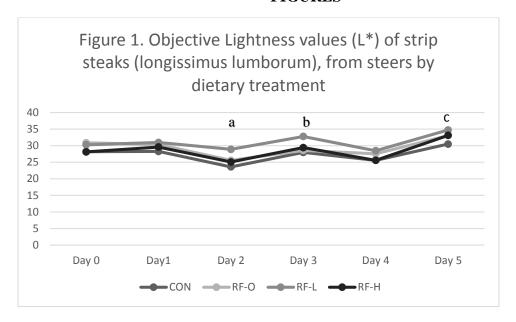
Table 8. Least squared means for thiobarbituric acid reactive substances by dietary								
treatment								
	Treatment							
	С	RF-O RF-L RF-H				SE	P values	
TBARS Day 0		0.83	0.60	0.66	0.72	0.19	0.96	
TBARS Day 7		2.81	2.96	2.83	2.97	0.19	0.96	

Table 9. Least squared means for thiobarbituric acid reactive							
substances by breed treatment							
	Treatment						
	Jersey Crossbred SE P value						
TBARS Day 0	0.48	0.87	0.14	< 0.001			
TBARS Day 7	2.56	3.27	0.14	< 0.001			

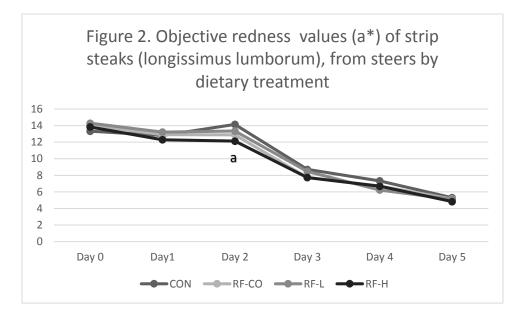
Table 10. Least squared means for sensory characteristics of Bologna by dietary treatment							
		Tre					
	C	RF-CO	RF-L	RF-H	F	P values	
Overall Liking	71.0	70.0	69.0	68.0	2.30	0.07	
Flavor Liking	71.0	70.0	69.0	68.0	2.20	0.09	
Texture Liking	72.0 ^a	70.0^{a}	70.0^{ab}	67.0^{b}	4.10	0.01	
Off flavor	3.7	3.8	4.0	4.0	0.90	0.45	
Toughness	4.7 ^b	3.8°	4.7 ^b	6.6ª	36.60	< 0.001	

Table 11. Least squared means for sensory characteristics of Bologna									
by breed treatment									
	Treatment								
	Jersey Crossbred SE P value								
Overall Liking	71.0a	68.0b	15.00	< 0.001					
Flavor Liking	71.0a	68.0b	11.90	< 0.001					
Texture Liking	71.0a 68.0b 5.80 0								
Off flavor	4.0	3.8	1.60	0.09					
Toughness	5.0	4.4	28.90	< 0.001					

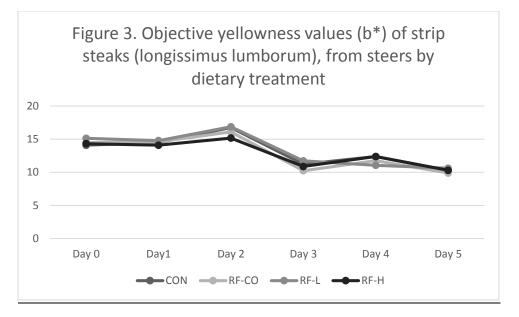
FIGURES

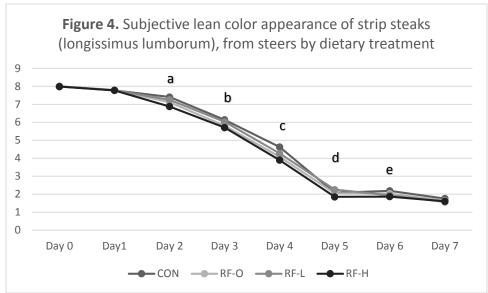


^a CON is different than RF-O, RF-O different then RF-L, RF-L different than RF-H ^bCON different from RF-O, RF-O different from RF-L ^cCON different from RF-L



^aCON different from RF-H





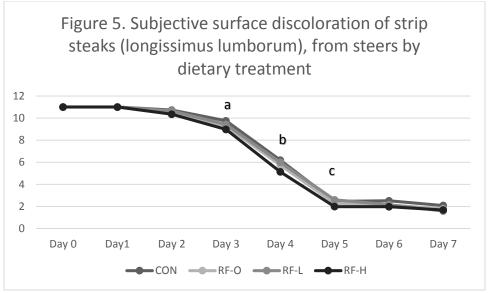
^aCON different from RF-H, RF-L different from RF-H

^bCON different from RF-H, RF-L different from RF-H

[°]CON different from RF-O, RF-L different from RF-H

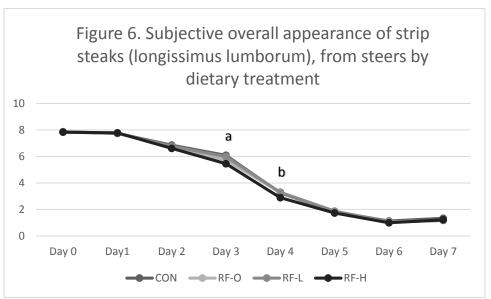
^dRF-L different from RF-H

^eCON different from RF-H



^aCON different from RF-H (P=0.013)

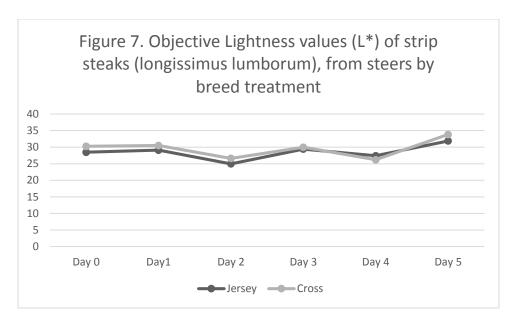
^cRF-L different from RF-H (P=0.031)

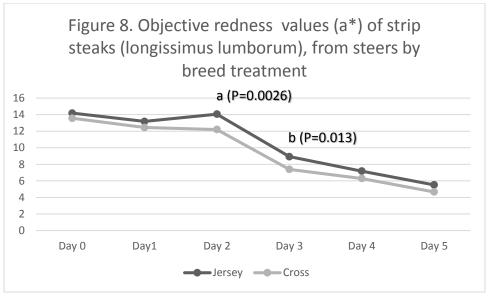


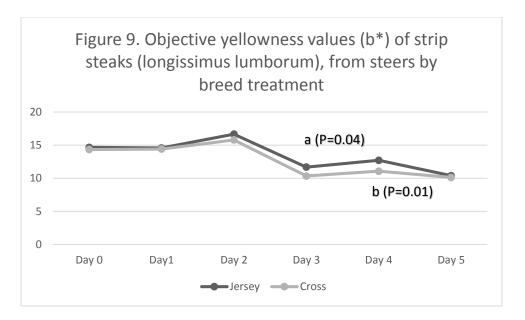
 $^{\rm a}CON$ different from RF-O (P=0.39) and RF-H (P=0.001), RF-L different from RF-H (P=0.001)

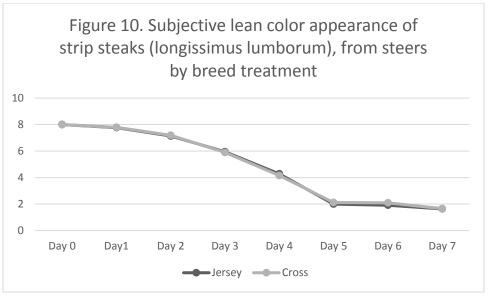
^b RF-H different from CON (P=0.001), RF-O (P=0.037), and RF-L (P=0.004)

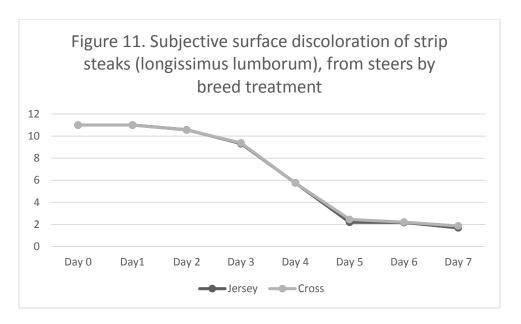
^b RF-H different from CON (P=0.023), RF-O (P=0.007), and RF-L (P=0.009)

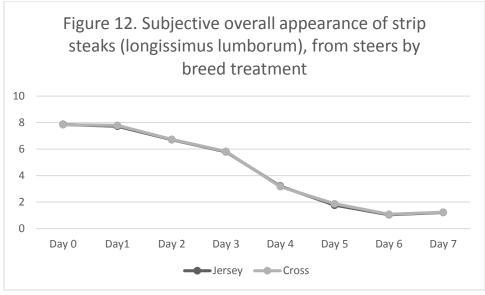


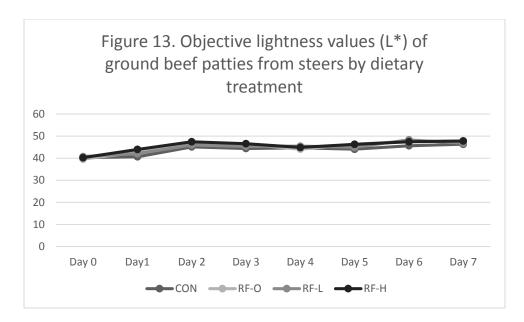


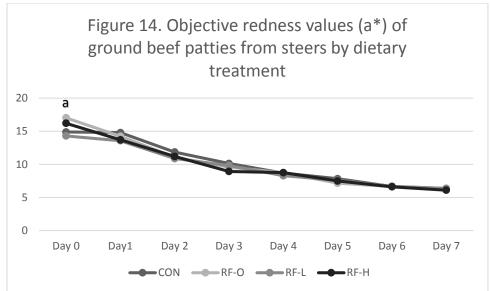




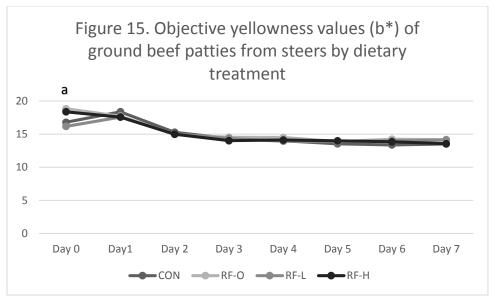




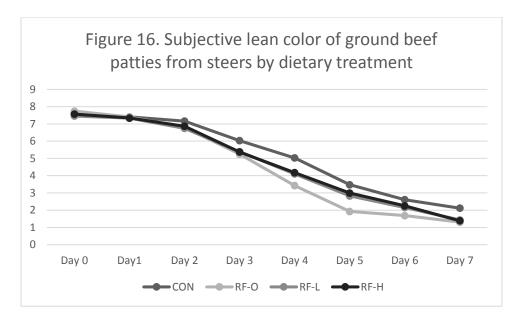


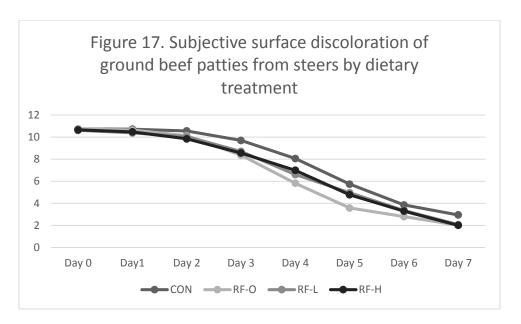


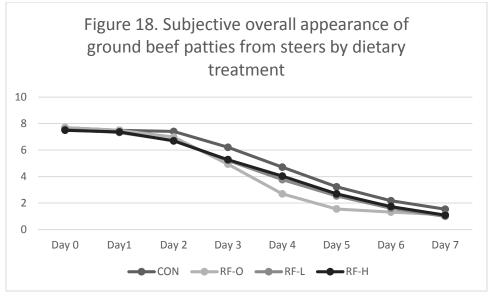
 a CON different from RF-O (P=0.002) and RF-H (P=0.024), RF-O different from RF-L (P=0.001), RF-L different from RF-H (P=0.002)

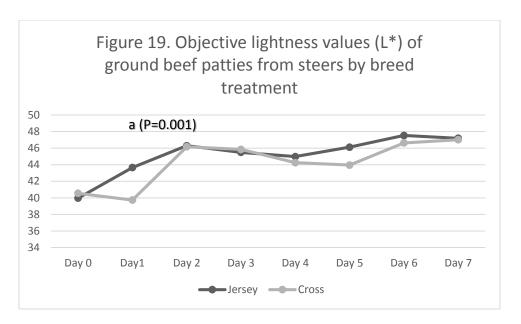


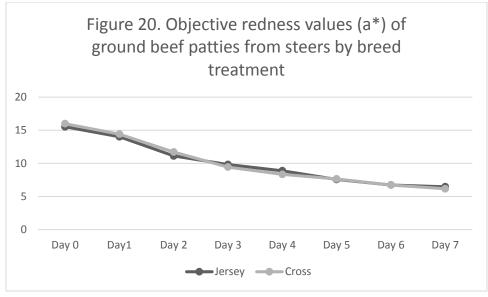
^aCON different from RF-O (P=0.002) and RF-H (P=0.012), RF-O different from RF-L (P=0.001), RF-L different from RF-H (P=0.001)

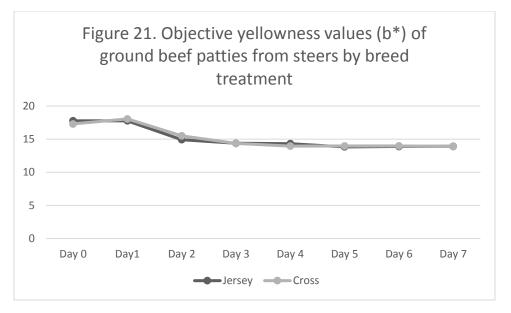


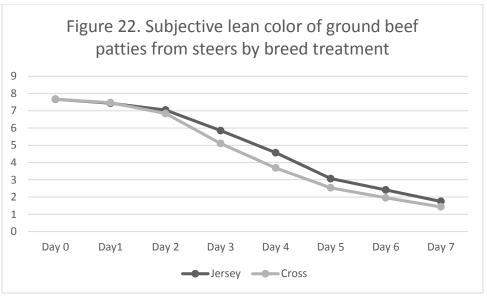


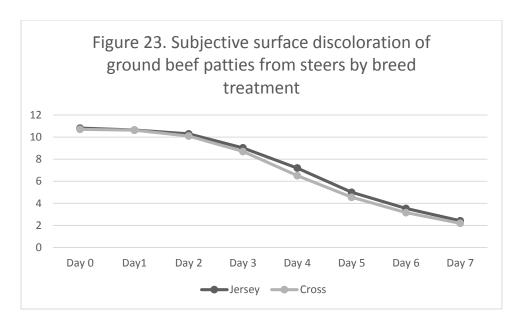


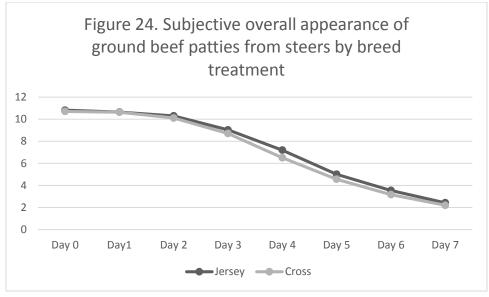


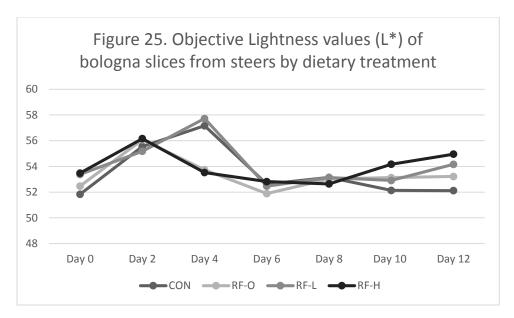


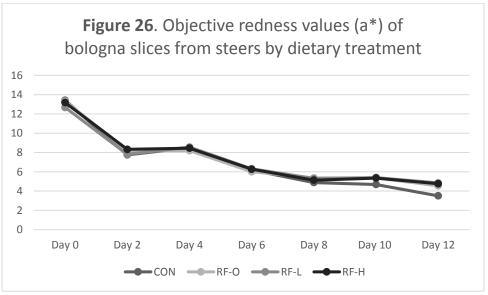


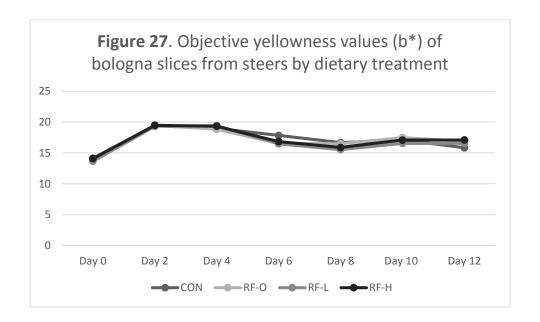


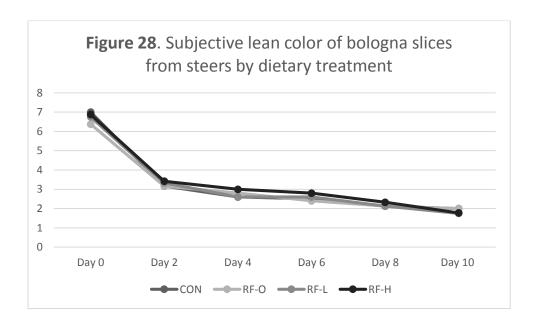


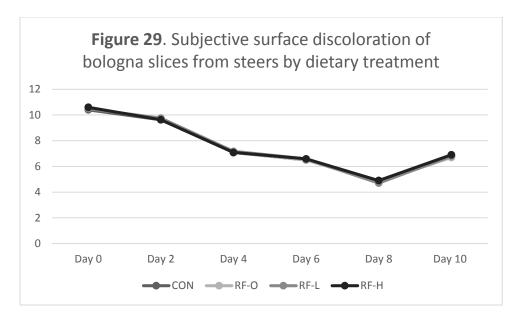


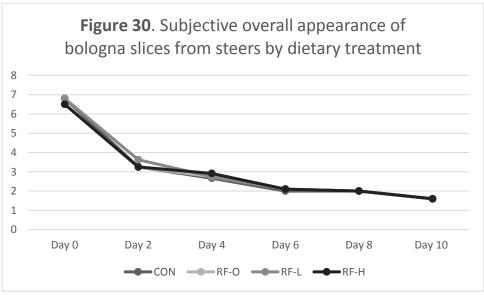


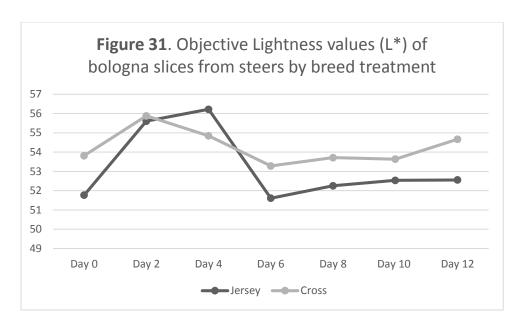


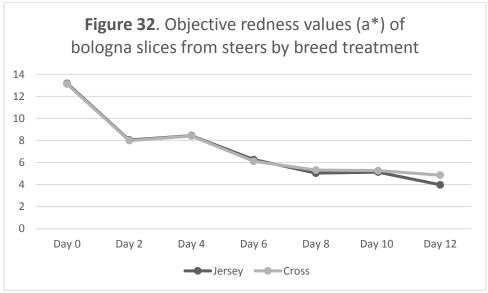


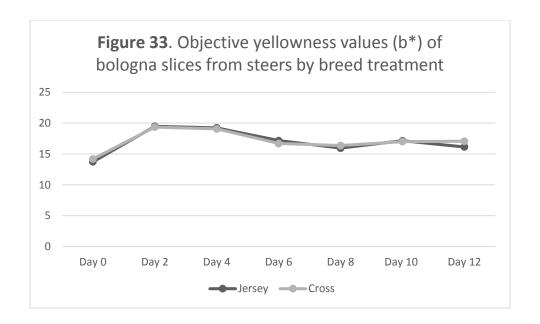


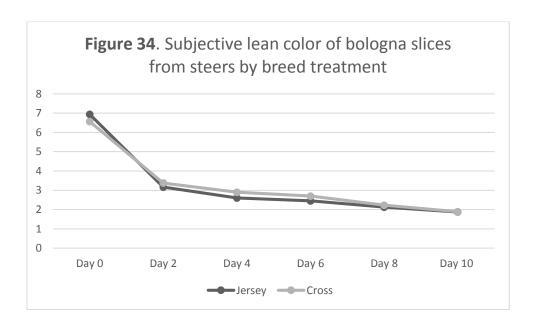


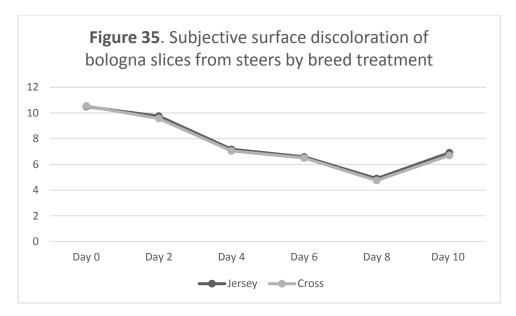


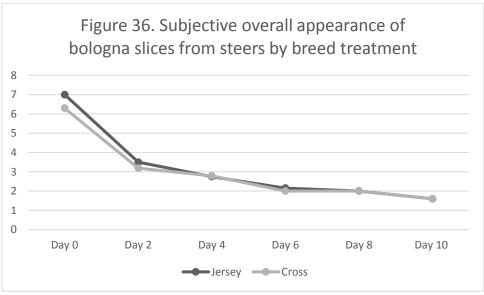


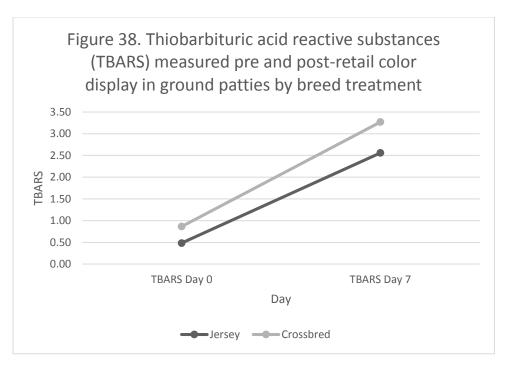


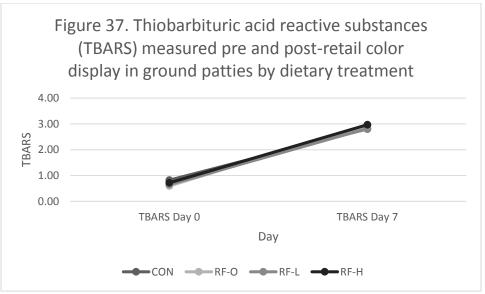












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Appendix A Standard Operating Procedure for TBAR Analysis

Agricultural Utilization Research Institute

SAM # 04

Marshall Fats and Oils Laboratory Standard Analytical Method **Page** 1 of 3

Title: TBARS Distillation Method

I. Reagents/Materials

- A. Hydrochloric Acid: ACS grade, VWR Catalog No. VW3110-3
- B. 2-Thiobarbituric Acid: CAS 504-17-6, Sigma Catalog No. T-5500
- C. <u>Dow Antifoam:</u> Dow Corning Antifoam Catalog No. 1520-US
- D. <u>1:2 Hydrochloric Acid:</u> Carefully combine 1 part concentrated HCl and 2 parts distilled water. Mix well.
- E. <u>0.02M Thiobarbituric Acid (TBA):</u> Dissolve 0.2882 g thiobarbituric acid in 100 mL distilled water. Mix well.
- F. Water: AURI distilled water

II. Instrumentation

- A. Spectrophotometer: Spectronic 20⁺, Spectronic Instruments, Inc.
- B. Water Bath: VWR

III. Procedure

- A. Obtain a 250 mL flat-bottomed round flask and place two glass beads inside.
- B. Weigh out 10 g of sample, record the weight, and blend with 50 mL of distilled water in a laboratory blender.
- C. Carefully transfer the blended sample into the 250 mL flask.
- D. Wash the blender with 47.5 mL of distilled water and add to the flask
- E. Add 2.5 ml of 1:2 hydrochloric acid and 3-4 drops of Dow Antifoam.
- F. Distill the contents of the flask at the highest temperature possible on a hot plate and collect the distillate into a 100 mL beaker.

- G. Continue to distill the contents of the flask until you have collected 50 mL of distillate in the beaker.
- H. Mix the distillate.
- I. Pipette 5 mL of distillate and 5 mL of 0.02M TBA into a scintillation vial and mix well.
- J. Prepare a blank by pipetting 5 mL of distilled water and 5 mL of 0.02M TBA into a scintillation vial.
- K. Immerse the vials in a 95° C water bath for 35 minutes.
- L. Allow the vials to reach room temperature by immersing them in cold water for 3-5 minutes.
- M. Turn on the spectrophotometer by turning the Power Switch/Zero Control (knob on the left side of the instrument) clockwise.
- N. Wait 15 minutes for the spectrophotometer to warm up.
- O. Set the Wavelength Control (knob on top) to 532 nm.
- P. Set the filter lever (small lever at the bottom) to the 340-599 nm wavelength position.
- Q. Adjust the meter to 0% T with the Power Switch/Zero Control knob. Make sure the sample compartment is empty and the sample cover is closed when you do this.
- R. Wash a spectrophotometer test tube twice with a small amount of the blank solution.
- S. Fill the test tube with the rest of the blank solution.
- T. Wipe of the outside of the test tube to remove dust and fingerprints and place it in the sample compartment.
- U. Align the guide mark on the test tube with the guide mark at the front of the sample compartment. Close the lid and adjust the meter to 100% T with the Transmittance/Absorbance Control (knob on the front right side of the instrument).
- V. Remove the test tube from the sample compartment and discard the solution.

- W. Press the Mode button to switch the instrument from percent transmittance to absorbance.
- X. Using the same test tube used for the blank solution, rinse the test tube with small amounts of sample and read each sample solution recording the absorbance value.

IV. <u>Calculations</u>

Sample Absorbance x 7.8 = TBARS(ppm)

V. Reference

- A. Tarladgis et al. (1960)
- B. Spectronic 20⁺ Series manual

Appendix B Smoke House Procedure for Bologna

Thermal Processing

The internal should be held at 140°F for at least 12 min to meet 6.5 log reduction stipulated in the USDA/FSIS Appendix A.

The thermal processing schedule is as follows:

Step	Process	Time	Dry Bulb (°F)	Wet Bulb (°F)	Relative Humidity
1	Smoke	01:00	75	-	-
2	Cook	03:30	150	130	30%

Cook to an internal temperature of 150°F.

Cooling Process

It is very important that cooling be continuous through the given time/temperature control points.

- 1. During cooling, the product's maximum internal temperature should not remain between 130°F and 80°F for more than 1.5 hours nor between 80°F and 40°F for more than 5 hours. This cooling rate can be applied universally to cooked products (e.g., partially cooked or fully cooked, intact or non-intact, meat or poultry) and is preferable to (2) below.
- 2. Over the past several years, FSIS has allowed product to be cooled according to the following procedures, which are based upon older, less precise data: chilling should begin within 90 minutes after the cooking cycle is completed. All product should be chilled from $120^{\circ}F$ (48°C) to $55^{\circ}F$ (12.7°C) in no more than 6 hours. Chilling should then continue until the product reaches $40^{\circ}F$ (4.4°C); the product should not be shipped until it reaches $40^{\circ}F$ (4.4°C).
- 3. The following process may be used for the slow cooling of ready-to-eat meat and poultry cured with nitrite. Products cured with a minimum of 100 ppm ingoing sodium nitrite may be cooled so that the maximum internal temperature is reduced from 130 to 80 °F in 5 hours and from 80 to 45 °F in 10 hours (15 hours total cooling time).

Appendix C Labeled Affective Magnitude Scale

