FORAGE TYPE EFFECTS ON BEEF CATTLE PERFORMANCE AND CARCASS TRAITS

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FORAGE TYPE EFFECTS ON BEEF CATTLE PERFORMANCE AND CARCASS TRAITS

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FORAGE TYPE EFFECTS ON BEEF CATTLE PERFORMANCE AND CARCASS TRAITS

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VITA

Abby Nicole Frank, the daughter of Edward M. and Patsy Frank, was born September 19, 1984 in Pensacola, Florida. She graduated from Foley High School in Foley, Alabama in 2002. After high school, she attended Auburn University where she received a Bachelor of Science in Animal Science in May 2006. Abby entered graduate school in August 2006 to pursue the degree of Master of Science in Animal Science with an emphasis in Meat Science and Muscle Biology.

THESIS ABSTRACT

FORAGE TYPE EFFECTS ON BEEF CATTLE PERFORMANCE AND CARCASS TRAITS

Abby Nicole Frank

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Non-implanted Angus crossbred steers from Auburn University's Wiregrass Research and Extension Center resident herd in Headland, Alabama were used for a two-year forage-finishing study. In January 2006 (n = 18) and December 2006 (n = 18) steers were randomly assigned a forage treatment. Steers (n = 3) were grazed on duplicate 1. 42 ha paddocks of Wren's Abruzzi Rye (*Secale cereale* L.), Harrison Oats (*Avena sativa* L.), or Marshall Ryegrass (*Lolium multiflorum* Lamarck). Pen served as experimental unit for the project. The steers were humanely harvested after 85 or 95 d on test for year 1 and

145 or 152 d on test for year 2. Following harvest, ADG, liveweight, carcass traits, WBS, sensory traits, fatty acid profile, and lipid oxidative stability were analyzed from the longissimus muscle. The type of forage did not affect ($P > 0.05$) ADG, liveweight, dressing percentage, kidney, heart, and pelvic fat, marbling, yield grades, quality grades, lean maturity scores, bone maturity scores, or pH. Steers finished on ryegrass had heavier ($P < 0.05$) HCW than rye and oats. Year did not affect ($P > 0.05$) liveweight, kidney, heart, and pelvic fat, marbling, preliminary yield grade, actual yield grade, quality grades, and bone maturity scores. Year 1 had higher ($P < 0.05$) ADG, but lower ($P < 0.05$) HCW, dressing percentages, adjusted yield grades, lean maturity scores, and pH. Lipid oxidative stability was not significant ($P > 0.05$) among forage type, year, or forage*year groups. Forage*year did not have a significant affect ($P > 0.05$) on L*, a*, or b* values for fat or lean color. Forage type did not affect ($P > 0.05$) fat L* or b*, lean L*, a*, or b* values. Ryegrass and oats had higher ($P < 0.05$) fat a* values than rye. Year did not affect ($P > 0.05$) fat L* or b*, or lean L* values. Sensory traits, WBS, and cookloss were not affected ($P > 0.05$) by forage type. Off flavor, WBS, and cookloss were not affected ($P > 0.05$) by year. Year 1 had higher ($P < 0.05$) sensory panel rating for juiciness, tenderness, and flavor intensity. Off-flavor descriptors were not affected ($P > 0.05$) by forage or year. Year did not affect ($P > 0.05$) C8:0, C9:0, C11:0, C11:1, C12:0, C12:1, C13:0, C13:1, C14:0, C14:1, C16:1t, C20:3n3, C20:5n3, C22:0, C22:1, C22:4, C22:3, and C24:1 concentrations. Year affected ($P < 0.05$) saturated, mono, poly, omega-6, and omega-3 fatty acid concentrations.

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CHAPTER I

INTRODUCTION

With the high cost of feed grain production and the demand for alternative fuels, the future availability of feed grains for livestock is uncertain. In the long run, it is likely that the U.S. beef cattle production will depend more heavily on forages as feed for livestock (Bowling et al., 1977; Bidner et al., 1981a). Currently in the U.S., 70% of the cattle's harvest body weight is from forage before they are sent to feedlots. The forage-based stocker system is the least expensive step in beef production (Martin, 2004). Because of their lower hot carcass weights and quality grades, cattle sold into a natural, forage-fed, nonimplanted beef market would need to earn a 16% premium to be economically competitive with cattle finished in a feedlot (Berthiaume et al., 2006). From a producer's standpoint, grain- or silage- finished- cattle would be more beneficial if other factors are not considered. McMillin et al., (1981) stated that silage-finished cattle had higher live weight value and mean carcass value than forage-finished cattle because of silage producing higher quality grades and heavier carcass weights. With rising fuel costs, there has been an increase in transportation cost.

Research on forage-finished beef has been published since the 1930s. The early literature focused on many of the same issues of today's meat industry. Fore fathers in meat science focused on factors influencing carcass quality in beef. Wanderstock and Miller (1948) stated that in 1936, fatter animals had higher dressing percentages, carcass

grades, percentage of fat in the carcass, and lower percentage of moisture, protein, lean, and bone. In the 1940s researchers found that cattle produced on grass alone had lower grades and were less palatable than cattle finished on a supplemental concentrate diet. Color difference between forage- and grain-finished meat was published in the 1940s. Forage-finished cattle had yellower fat and darker lean which is still true today. It is believed that lower amounts of lactic acid lead to brighter colored beef. A slight difference in flavor between forage- and grain-finished roasts was noted by researchers as early as 1931 (Wanderstock and Miller, 1948). Prior to World War II most of the beef consumed in the United States was from cattle finished on forage or limited grain. By the 1950s the demand for grain-finished beef had increased. Americans started to demand the flavor of cattle finished on high-grain diets in the 1970s (Schupp et al., 1980).

The problem with comparing grain- versus forage-finished beef is the endpoint. If the comparison groups of cattle are finished to the same age, fat deposit can affect the palatability outcome. If cattle are finished to the same degree of fat, then there is a difference in maturity which affects the color. Bowling et al., (1978) found that steers finished on grass or grain and grass were approximately six mo older when they reached slaughter weight and grade comparable to steers finished on grain in a drylot. Nuernberg et al. (2005) also found that cattle finished on grass-based diets were 4-6 mo older at finishing.

As consumers become more concerned about health, they are looking for alternatives to traditional beef. Many of them are turning towards niche markets. “Organic” and “Naturally-raised” products are becoming more prevalent on supermarket shelves. Forage-finished beef is also becoming more popular. Realini et al. (2004) stated that

dietary recommendations for humans to consume less saturated fats have prompted an increased interest in meats with more unsaturated fatty acids. Antioxidant and redox potential of fresh meat was significantly improved when cattle are finished on forage (Descalzo et al., 2007). The purpose of this research is to compare different forages as a successful means of finishing beef cattle.

CHAPTER II

REVIEW OF LITERATURE

Cattle Production

The southeastern United States has been classified as a cow-calf operation region. Mature cattle are shipped to feedlots in western states for finishing. The southeastern United States is a good candidate for forage-finishing because of the production of quality forage year-round (Sapp et al., 1998). Ryegrass and clover are produced for winter grazing from November to May. Bermudagrass and Bahia grass is used for forage between April and November (Bagley and Feazel, 1987). In Alabama, bahiagrass, bermudagrass, crabgrass, endophyte-infected tall fescue, and annual ryegrass are excellent grazing forages (Ball et al., 1999). Ryegrass is more labor and equipment intensive due to annual plantings. There are many factors that affect the cattle's performance during forage-finishing such as breed and climate of the production area. Breed of cattle has been a known factor of determining the rate of gain. Heat and humidity in the Southeast can also affect cattle performance.

Abdullah et al. (1979) used forty-eight steers consisting of either Angus-Hereford crosses or Angus-Hereford-Brahman crosses and placed in two groups. The grain- finished steers gained over 45kg more than the forage steers. Grain steers had a 7:13 feed to gain ratio. Because of forage quality problems at the experiment station, it took forage steers approximately 280 d to gain 90kg. Bidner et al. (1981c) found that cattle fed on forage had slower rates of gain than cattle in feedlots. Cattle finished on forage can take 6

mo longer to finish than their counterparts in feedlots (Brown et al., 2007). Forage-finished cattle have lower live weights just prior to slaughter than their feedlot counterparts (Muir et al., 1998a).

Bagley et al. (1981) conducted an experiment in Louisiana and found that cattle finished on winter annuals and white clover had the best performance out of the forage treatments while cattle finished on bermudagrass and white clover did the worst of all the treatments. Overall, the amount of gain was lower in the forage-finished cattle than grain-fed cattle. Out of both treatments, cattle slaughtered during July performed the poorest. Nuernberg et al. (2005) had similar findings as Bagley et al. (1981) and Abdullah et al. (1979) in which bulls finished on a grass-based diet that consisted of pasture, wilted silage, hay, and a pelleted concentrate mix of 12% barley and 10% cracked linseed had lower average daily gains than strictly concentrate-fed bulls.

Nutrient content, availability, and abundance can play a major part in the effectiveness of warm- or cool-season forages. Bermudagrass is a common forage grown in the Southeast. Cattle performance can be determined by the variety of bermudagrass planted. Utley et al. (1981) found that Callie bermudagrass increased steers' average daily gain by 13% more than Tifton 44 and 16% more than Coastal bermudagrass which is the most commonly used in the southeastern United States. Callie bermudagrass is high quality and fast growing, but is more susceptible to dying in winter. Redfearn et al. (2002) demonstrated that Crude Protein (CP) of ryegrass decreased during the later growing season which is around April. During the study from 1997-1999, CP decreased from 260 - 120 g CP kg^{-1} as the growing season progressed. Forty percent of total forage production from annual ryegrass cultivars occurred in early season (December - February) and 60% growth occurred during late season (March - May). Approximately 30% of the total forage production occurred in April alone. Marshall Ryegrass is a cold-tolerant annual forage that is excellent for winter grazing (Bransby et al., 1997). Bidner et al. (1986) stated that cattle finished

on high-energy diets have a higher amount of body fat compared to cattle on low-energy diets. Allen et al. (2000) found ways to balance year-round forage in a simple three paddock system that consisted of warm- and cool-season forage that was able to improve beef production per unit land area while reducing inputs of labor and equipment.

The breed of cattle can affect forage-finishing performance. Bidner et al. (1986) studied the results of 44 Angus-Hereford (AH) and 43 Angus-Hereford-Brahman (AHB) steers on either corn- or forage-finishing diets. Angus-Hereford-Brahman steers had higher final weights and gain on forage than AH steers. Angus-Hereford steers gained faster in the feedlot. Angus-Hereford steers had more marbling and higher quality grades. Warner-Bratzler shear was greater for AHB steers. Angus-Hereford steers had larger longissimus area and lower yield grades. No color difference was noticed between breed types. Rib steaks from AH steers were rated more flavorful than AHB steers. Consumers ranked both AH and AHB steers as acceptable. Bidner's findings were similar to Abdullah's findings in 1979. Camfield et al. (1999) found that intermediate-framed, early-maturing cattle, such as current purebred Angus, and small-framed, early-maturing cattle, such as Angus cattle from the 1950s, had higher amounts of marbling and quality grades than large-framed, late-maturing cattle, such as Chianina or Charolais, and intermediate-framed, intermediate-maturing cattle, such as Red Poll or Hereford, when finished on forage. These findings are similar to Brown et al. (2005). Cattle that reach maturity faster have more soluble collagen and likely to be more tender (Muir et al. 1998b).

Benefits of Forage Finishing

As health concerns increase, more consumers are turning to alternatives in the food they eat. Beta-carotene is a fat soluble antioxidant that can be converted to vitamin A in the body. Vitamin A is a fat-soluble vitamin that is critical for epithelial tissues, respiratory and genital-

urinary tract mucous membranes, cornea and conjunctiva of the eye, preventing night blindness by catalyzing metabolism in the retina. It also promotes growth, promotes resistance to infections, maintains the nervous system, and helps successful reproduction (Romans et al., 2001). Forage-finished beef has higher concentrations of β-carotene compared to concentrate- finished beef (Irie, 2001; Yang et al., 2002; Descalzo et al., 2005).

Vitamin E is a fat-soluble vitamin that is also an antioxidant. Vitamin E is beneficial to fertility and sperm development (Romans et al., 2001). Lee et al. (2005) indicated that there is a decrease in cardiovascular deaths with vitamin E in the diet. Vitamin E is found in higher concentration in forage-finished beef than concentrate- finished beef (Yang et al., 2002; Descalzo et al., 2007).

Bidner et al. (1985) showed that consumers' recent concern over high fat and cholesterol along with economic pressures have led to purchasing leaner beef products which can be produced on all-forage or predominately-forage diets. Beef would be healthier if there was an increase in polyunsaturated fatty acids: saturated fatty acid ratio (French et al., 2000). Saturated fatty acids are risk factors for heart disease (Romans et al., 2001). Eicosapentaenoic (EPA; C22:5n3) and docosahexaenoic acid (DHA; C22:6n3) help prevent the formation of blood clots. Docosahexaenoic acid is also critical for brain development in infants along with normal brain maintenance in adults (Horrocks and Yeo, 1999). Duckett et al. (1993) found an increase in EPA and DHA in forage-finished cattle. Omega-3 fatty acids included in the diet can reduce the risk of heart-related deaths (Daviglus et al., 1997). High omega-6: omega-3 ratios can result in cardiovascular disease, cancer, inflammatory disease, and autoimmune disease (Simopoulos, 2002). Simopoulos also stated that a ratio of 4:1 is associated with a 70% decrease in total mortality, a 2.5:1 ratio can reduce rectal cell proliferation in colorectal cancer patients, and a lower

ratio can decrease risk of breast cancer in women. Further a 2 or 3:1 ratio suppresses inflammation of rheumatoid arthritis, and 5:1 ratio is beneficial to asthma patients. Forage-finished beef has lower omega-6: omega-3 ratios than grain-finished beef (French et al., 2000; Rule et al., 2002).

In a review by Whigham et al. (2000) it is hypothesized that the health benefits of conjugated linoleic acid (CLA) are anticarcinogenic, immunomodulation, decrease in fat mass, increase in lean mass, and antiatherosclerosis. Health benefits depend on the type of animal, breed, dosage of CLA, and age. These findings corresponded to those of Kelley and Erickson (2003). There is a significant increase in CLA with cattle finished on grass (French et al., 2000).

Carcass Data

Carcass traits of beef can be altered by the production practices. Forage-finished cattle have lower carcass weights (Baublits et al., 2004; Realini et al., 2004; Cox et al., 2006; Kerth et al., 2007). McMillin et al. (1984) found that silage-finished cattle had higher carcass weights, dressing percentages, quality grades, and backfat thickness while forage-finished cattle had lower yield grades. Kerth et al. (2007) found that steers finished on ryegrass had lower USDA yield grades than steers finished on grain or ryegrass/grain mixture. Lower yield and quality grades were found in a study by Schaake et al. (1993). Bennett et al. (1995) found there is not a significant difference in yield grade between steers finished on rhizoma peanut –tropical grass pastures versus concentrate.

Carcasses with higher yield grades will produce higher levels of fat and less saleable lean. Ribeye area between the 12^{th} and 13^{th} rib is a good representation of the total lean of the body. Ribeye area is not significantly different between feedlot and pasture cattle (Camfield et al., 1999). Forage-finished cattle have less fat over the ribeye and smaller longissimus muscle area than concentrate-finished steers (Bidner et al., 1985; May et al., 1992; Bennett et al., 1995; Realini et

al., 2004; Kerth et al., 2007). As days on feed increase, there is a linear increase in slaughter weight and dressing percentage (May et al., 1992; Brown et al., 2005). Marbling is the intramuscular fat that is between muscle fiber bundles. Marbling score is the only trait found in cattle that has the greatest effect of quality grade in young beef carcasses (May et al., 1992). Forage-finished beef has lower marbling scores compared to concentrate- finished beef (Harrison et al., 1978; Crouse et al., 1984; Brown et al., 2005). Kerth et al. (2007) did not find a significant difference in marbling score between steers finished on ryegrass, ryegrass/grain, or grain when fed to a constant fat thickness. Cattle fed grain for 112 d are the first to achieve a marbling score of small to meet the USDA quality grade of Choice, but marbling scores did not significantly increase after 112 d on feed (Duckett et al., 1993). Cattle finished on forage with soyhull pellets supplement achieved a USDA quality grade of Choice while cattle just finished on fescue had an USDA quality grade of Standard (Baublits et al., 2004). Brown et al. (2005) also found that cattle finished in a feedlot had higher quality grades than forage-finished cattle.

Bowling et al. (1977) stated that grain-finished beef had twice as much subcutaneous fat; longer sarcomeres; higher percentage of kidney, pelvic, and heart fat; and higher carcass confirmation scores than forage-finished beef. Kidney, pelvic, and heart fat is higher in steers finished on grain or ryegrass/grain than those finished solely on ryegrass (Kerth et al., 2007). Schaake et al. (1993) also found less KPH fat in cattle finished on fescue-clover or summer pasture.

Davis et al. (1981) found no significant differences in sarcomere length between cattle finished on grain or forage. Grain-finished steers have higher dressing percentages and quality grades (Bowling et al., 1978; Schroeder et al., 1980). Cattle finished on high-forage diets usually weigh less and have less fat than cattle finished on high-energy diets, making them susceptible to

cold shortening (Bidner et al., 1985; Schroeder et al., 1980; Muir et al., 1998b). Increase of tenderness is associated with increased fat cover opposite the longissimus muscle between the 12th and 13th rib, which suggests that fatter carcasses sustain less shortening of muscle fibers. The correlation between fat and tenderness does not account for all the tenderness variations between grass- and grain-finished beef (Bowling et al., 1977). Cattle finished on Fescue-clover and summer pasture have lower percentage of fat in the 9-10-11th rib section (Schaake et al., 1993). Grain-finished cattle are fatter and have less moisture content than forage-finished cattle (Davis et al., 1981; Schaake et al., 1993; Sapp et al., 1999). The increase in fat is correlated with a decrease in moisture, protein, and ash (Duckett et al., 1993). As time on feed increased the amount of magnesium, potassium, and iron increased (Duckett et al., 1993). Forage-finished heifers have coarser lean texture than grain-finished heifers (Harrison et al., 1978; Davis et al., 1981; Crouse et al., 1984). Schaake et al. (1993) stated that beef from cattle finished on fescue-clover or summer pasture had softer lean and external fat. Bone and lean maturity scores are higher for forage-finished beef than grain-finished beef (Kerth et al., 2007).

Color

Myoglobin is the basic pigment of meat color. When myoglobin interacts with other environmental factors, it changes the color of meat. Myoglobin gives meat a dark red, fresh meat color that occurs in meat that is freshly cut and has not been exposed to oxygen. Oxymyoblobin is formed when oxygen interacts with myoglobin which produces the bright-red color of fresh meat. Oxidation can turn myoglobin into metmyoglobin. Meat can be oxidized by UV lights, certain metals, or oxygen producing a brownish color in meat (Aberle et al., 2001). During retail conditions, muscle color from forage-finished cattle deteriorates quickly (Bowling, 1977). Alpha-

tocopherol inhibits lipid oxidation, thus maintaining the color of oxymyoglobin in meat (Faustman et al., 1998). It is believed that forage is high in α-tocopherol (Descalzo et al., 2005).

Color of the lean and (or) fat are important criteria for consumers' decision on what meat to purchase (Gatellier et al., 2005). Color is determined by visual appraisal or a spectrophotometer. When steaks are placed on polystyrene/ ethylvinyalcohol/ polyethylene trays and overwrapped with oxygen-permeable PVC film then placed in retail conditions, O'Sullivan et al. (2004) found that redness (a*) decreased as the days in retail increased regardless of live animal diet.

Fresh grain-finished beef is usually a bright, cherry red, with the exception of older animals and animals that were stressed during the slaughter process having darker red meat (Muir et al., 1998a). Dark firm dry (DFD) meat occurs when glycogen is unable to deplete the pH levels such as when an animal is stressed. Dark firm dry meat also contains less moisture, thus darker color meat due to proteins being able to bind more tightly to water. Craig et al. (1959) stated that retained moisture results in brighter color. They also hypothesized that the more fat in a sample, the more light is reflected. It is stated that there is more moisture loss in forage-finished cattle along with less fat. Faustman and Cassens (1990) stated that pigment oxidation can be accelerated by high relative humidity and low air velocity.

Types of production and stress during slaughter are the main factors that determine beef color. Forage-finished cattle have darker lean color than grain-finished cattle (Bowling, 1977; Baublits et al., 2004; Realini et al., 2004; Nuernberg et al., 2005). The darker lean color makes forage-finished cattle susceptible to higher lean maturity scores (Crouse et al., 1984; May et al., 1992). Cattle finished in a drylot have brighter meat (Craig et al., 1959; Schroeder et al., 1980; Schaake et al., 1993). It has been speculated that forage-finished cattle have darker meat because of exercise required during grazing and the animals are more prone to stress during slaughter from

handling that they have not experienced before and confinement. The backfat thickness is correlated with the meat color. Page et al., (2001) stated that backfat thickness lower than 0.76 cm causes darker meat and higher pH. There is no significant difference in myoglobin content between grain- and forage-finished steers (Bowling et al., 1978). Traditionally darker meat has been associated with higher pH.

Forage-finished beef has greater color stability than grain-finished beef (O'Sullivan et al., 2004). Ascorbic acid delays the formation of metmyoglobin, thus allowing meat to retain oxymyoglobin. Forage-finished cattle have higher concentrations of ascorbic acid (Descalzo et al., 2005).The a* values for forage-finished cattle is lower than grain-finished (Yang et al., 2002). Yang et al. (2002) speculated that color differences between cattle could be due to variations in chilling rates because of carcass size.

The a* value is useful in measuring metmyoglobin formation which is beneficial in determining color stability, because metmyoglobin changes the meat from red to greenish-brown. Based on a* values, forage-finished steaks retained higher red color over a 10 d display period (Sapp et al., 1999). Lean maturity is highly correlated with L* (whiteness) because it is directly related to lightness/darkness. Forage-finished beef has higher L* values than grain-finished beef (Baublits et al., 2004; Gatellier et al., 2005). The pH effect on muscle is determined with a* and b* values, thus it alters the hue of the muscle not the lightness/darkness. High pH causes the meat to appear more purple and low pH causes the meat to appear more orange (Page et al., 2001).

The fat of forage-finished cattle is more yellow than grain-finished cattle (Harrison et al., 1978; Baublits et al., 2004; Realini et al., 2004; Kerth et al., 2007). Irie (2001) stated that yellow fat is less desirable to consumers. Yellow fat is mainly due to β-carotene and hemoglobin

concentrations. Yang et al. (2002) stated a diet high in forage increases the amount of β-carotene in an animal.

Tenderness

Beef tenderness can be affected by numerous factors such as feeding regimen, age, breed, and muscle location. Collagen is a structural connective tissue that is prevalent in muscle that can alter the tenderness of meat (Aberle et al., 2001). As the animal ages, the collagen matrix becomes more complex, thus younger animals tend to be more tender than older animals. The amount of collagen found in the muscle is determined by the location. Muscles used for locomotion have more collagen and are tougher than loin muscle (Cox, 2004). It has been hypothesized that forage-finished beef is tougher due to more exercise to fulfill hunger, increased age at finishing, and the growth rate of the animal. Grain-finished carcasses are more tender than the grass-fed (Bowling et al., 1977; Davis et al., 1981; May et al., 1992; Nuernberg et al., 2005; Brown et al., 2007). Other researchers have found no difference in Warner-Bratzler shear values between forage- and grain-finished beef (Bidner et al., 1981a; Baublits et al., 2006; Cox et al., 2006). Sapp et al. (1998) had similar findings in which the forage-finished steaks were as tender to a sensory panel and shear force as grain-finished steaks. Many other researchers have found that grain-finished beef is more tender than forage-finished beef.

Feeding cattle concentrates more than 84 d does not increase tenderness and that after 196 d there is a decrease in tenderness (May et al., 1992). Crouse (1984) stated that forage-finished heifers had steaks that were similar in tenderness as grain-finished heifers. Forage-finished beef had greater quantities of organoleptically-detectable connective tissue. Cox et al. (2006) did not find a difference in the amount of collagen found in grain- or forage-finished beef. Bowling et al. (1977) found that there is a 28.4 % sarcomere shortening of conventionally chilled (48h at 0±1ºC),

forage-finished carcasses while conventionally chilled, grain-finished carcasses had 17.2% more sarcomere shortening. When subcutaneous fat thickness is increased from 1.27 to 8.9 mm there is an increase in sarcomere length, increase in panel tenderness rating, and decrease in shear force. Subcutaneous fat above 10.2 mm had no benefit to sarcomere length or enhancing tenderness. Thus, differences in tenderness between grain- and forage-finished beef is partially due to differences in fat thickness which is related to cold-shortening (Purchas and Davies, 1974; Bowling, 1977; Harrison et al., 1978; Schroeder et al., 1980). Wu et al. (1981) did not find differences in total collagen content, sarcomere length, Warner-Bratlzer shear force values, or sensory panel ratings for tenderness between forage-finished and grain-finished beef. May et al. (1992) also stated that forage-finished beef had shorter sarcomere lengths which was most likely due to subcutaneous fat and lighter carcass weights, but there is not a correlation between this and taste panel tenderness.

Fatty acid concentration can also affect tenderness. Camfield et al. (1997) found that there is a positive correlation between shear force and longmissimus muscle stearic acid (C18:0) concentration. During the study, they found that cattle finished on grain for 90 d had lower stearic acid concentrations. Tenderness of beef can be improved by postmortem techniques. Bidner et al. (1981b) found that electrical stimulation can decrease WBS values, while vacuum-aging and blade tenderization can increase tenderness when measured by WBS and trained sensory panel.

Flavor

Consumers rank flavor as one of the top requirements for a desirable piece of meat. Melton et al. (1990) stated that numerous feeds such as fish products, raw soybeans, canola oil/meal, and pasture grasses can alter the flavor of red meat. Forage-finished beef has many off-flavors. The

type of forage can alter the flavor of forage-finished beef. Flint hills pasture of Kansas, orchardgrass-clover, rye-oats-ryegrass, forage sorghum, bluegrass-clover, rye-ryegrass-clover, arrowleaf clover, bermuda-clover-sudan and Coastal bermudagrass can affect the flavor of beef cattle (Melton et al., 1990). Low-energy forage diets have a less desirable and intense flavor than high-energy grain diets (Harrison et al., 1978; Melton et al., 1990; Schaake et al., 1993). The flavor of forage-finished beef can be attributed to beef fat (Griebenow et al., 1997). Grain-finished cattle have a "slightly to moderately intense" beef flavor (Davis et al., 1981; Melton et al., 1990). Grain-finished steaks have "moderately desirable" flavor, while forage-finished beef has been described as "slightly undesirable" (Montgomery and Bidner, 1979).

Forage-finished beef has been described as metallic, spicy, oxidized, astringent, chemically, sour, turpentine, cooked vegetable, nutty, pungent, earthy, oily, bread-like, tangy, saltiness, cheesy, blood flavor, plastic, dairy-like, or grassy (Davis et al., 1981; Bruce et al., 2005). In the same study by Bruce et al., some grain-finished beef was also characterized as earthy, musty, acidic, fatty, and liver. The longer cattle are finished on grain, the more palatable the flavors because of the increased amount of marbling (Camfield et al., 1997). Forage-finished cattle have a lower carbohydrate content which can alter the flavor (Melton et al., 1982). Forage-finished beef has a lower quantity of free sugars which can result in lower quantities of desirable beef flavor volatiles (Brown et al., 1979).

The fatty acid composition and fat content can determine the flavor of meat. Reduced desirability of flavor in forage-finished beef can be due to composition of intramuscular lipids and/ or concentration of nitrogen extractives in muscle (Bowling, 1977). Off-flavors can be linked to fatty acid concentrations. Camfield et al. (1997) found that myristic (C14:0), myristoleic (C14:1n5), palmitic (C16:0), margaric (C17:0), and oleic (C18:1n9) acid are negatively related to a

"cowy" flavor while vaccenic (C18:1), linoleic (C18:2n6), 5, 8, 11, 14, 17-eicosapentaenoic (C20:5n3) acids are positively related. Cardboard flavor is positively correlated to linoleic (C18:2n6), cis-11, 14-eicosadienoic (C20:2n6), and homo-gamma-linolenic (C20:3n6) acids and negatively correlated with myristoleic (C14:1n5) and stearic (C18:0) acid concentrations. Beef fat, liver, and sour flavor along with metallic feel are negatively correlated with vaccenic (C18:11) acid concentrations. Sourbit flavor is positively correlated with vaccenic (C18:11) acid concentrations. Myristoleic (C14:1n5), margaric (C17:0), stearic (C18:0), vaccenic (C18:1I), linoleic (C18:2n6), cis-11, 14-eicosadienoic (C20:2n6), homo-gamma-linolenic (C20:3n6), 5, 8, 11, 14, 17-eicosapentaenoic (C20:5n3) acid concentrations decreased with the days on feed. Myristic (C14:0), palmitic (C16:0), and oleic (C18:1n9) increase with days on feed.

Flavor can also be linked to diterpenoids which are derived from the break down of chlorophyll by ruminal microorganisms (Griebenow et al., 1997). The main indicator that cattle have been finished on forage is phyt-2-ene which produces a strong off-flavor (Griebenow et al., 1997).

Juiciness

"Juiciness" is an important organoleptic parameter in beef acceptability (Killinger et al., 2004). There is contradicting findings about juiciness of forage-finished beef. Some have found no difference in initial or sustained juiciness for forage- or grain-finished beef (Purchas and Davies, 1974; Bowling et al., 1977; Harrison et al., 1978). Sapp et al., (1998) found that grain-finished steaks are more juicy which Schroeder et al. (1980) also noted. Steers finished on ryegrass, grain, and ryegrass/grain mix had similar initial and sustained juiciness scores (Kerth et al., 2007). Differences among the studies can be directly linked to the amount of marbling in the study. Sensory panel ratings for juiciness are negatively correlated with myristic (C14:0), palmitic

(C16:0), and margaric (C17:0) acid, but is positively correlated with linoleic (C18:2n6), homo-gamma-linolenic (C20:3n6), and arachidonic (C20:4n6) acid concentrations (Camfield et al., 1997). In that study myristic and palmitic acid concentrations increased with days on feed. Margaric, linoleic, homo-gamma-linolenic, and arachidonic acid concentrations decreased with days on feed.

Consumer Acceptability

In a study by Kerth et al. (2007), 43.3% of consumers ranked flavor as leading factor in their steak decision followed by 40.3% ranked tenderness as most important and 16.4% ranked juiciness as key factor. Overall steak quality is more important to the consumer than feeding system, more humane production, no preference, perceived value, steak appearance, taste, and texture. Bowling et al. (1977) confirmed that grain-finished beef was more palatable which Kerth et al., (2007) also noted. Cattle finished on ryegrass have a less flavor intensity and beef flavor ratings during sensory panel than cattle finished on grain or grain/ryegrass mix (Kerth et al., 2007). There is no difference in cooking loss, cooking time, or degree of doneness between forage- and grain-finished beef. In a test market, Melton et al. (1990) found that 52% of 87 consumers would buy forage-finished beef again after the first use. Montgomery and Bidner (1979) stated that there were no differences in juiciness, tenderness, or connective tissue between forage- or grain-finished beef. Bidner et al., (1981c) stated that consumers did not find the fat color of forage-finished beef objectionable. In the same study, consumer panel members did not find a significant difference between cattle finished on forage or feedlot. The darker lean color of forage-finished beef is not as acceptable as the brighter lean of grain-finished beef (Schroeder et al., 1980). Brown et al., (1979) found that forage-finished ground beef had lower taste panel scores than concentrate- finished ground beef. Cox et al., (2006) stated that grain-finished steaks had higher retail consumer

acceptance for flavor, overall palatability, and price comparison. Of the retail consumers, 34% preferred the forage-finished steaks. In the same study there was difference in the take-home steaks between flavor, palatability, or price. A total of 54% of the take-home consumers preferred the forage-finished steaks. Forage-finished steaks can charge a premium of $2.38/kg to $5.61/kg (Cox et al., 2006). Sitz et al., (2005) found that consumers prefer domestic beef that is traditionally produced over Australian grass-fed or Canadian beef. They found that when domestic, Australian, and Canadian beef is matched in tenderness and marbling, consumers still believe domestic beef is more flavorful, juicy, tender, and overall more acceptable. In the same study, consumers stated that Australian grass-fed beef has off-flavors and off-odors. Consumer's perception of juiciness, flavor, and tenderness of forage-finished beef can be altered by enhancement (Robbins et al., 2003). Producing value-added products, such as heat-and-eat convenience meals, is a great way to increase consumer acceptance of forage-finished beef (Martin, 2004).

Fatty Acids

The fatty acid composition of ruminant fat can be altered by the animal's nutritional background (Realini et al., 2004). Rye/ryegrass pasture increases the amount of C18:2, C18:3, C20:3, C20:4, and C22:5 concentrations in the muscle (Faustman et al, 1998). Forage-finished beef is high in polyunsaturated fatty acids such as arachidonic (C20:4), eicosapentaenoic (C20:5), and docosapentaenoic (C22:5) acids (Montgomery and Bidner, 1979; French et al., 2000; Yang et al., 2002). Saturated fatty acids are less likely to experience lipid oxidation than unsaturated fatty acid (Yang et al., 2002). Forage-finished beef has more polyunsaturated fatty acids which can lead to oxidation (Gatellier et al., 2005). Oxidation can alter the color and flavor of meat. Antioxidants reduce the rate of oxidation in meat. Polyunsaturated fatty acids are more prone to attack of free

radicals at the allylic carbon between double bonds when the steak is heated causing oxidation to form short chain volatile compounds (Montgomery and Bidner, 1979).

Beef from forage-finished cattle have higher levels of α-tocopherol and other anti-oxidants than grain-finished cattle because of the compound occurs naturally in grasses (Yang et al., 2002; Nuernberg et al., 2005; Descalzo et al., 2007). Linolenic (C18:2n6) and stearic acid concentrations are higher in forage-finished beef while oleic (C18:1n9) and linoleic acid concentration is lower (Montgomery and Bidner, 1979; Yang et al., 2002). Duckett et al. (1993) found that polyunsaturated fatty acid concentrations decreased while monounsaturated fatty acid concentrations increased when time on feed increased. Forage-finished beef had lower hypercholesterolemic (C14:0+C16:0): hypocholesterolemic (MUFA+PUFA) ratios. As time on feed increased, myristic (C14:0) acid, myristoleic (C14:1) acid, pentadecyclic (C15:0) acid, palmitic (C16:0) acid, palmitoleic (C16:1) acid, 10-heptadecenoic (C17:1), oleic (C18:1) acid, nonadeyclic (C19:0) acid, gadoleic (C20:1) acid, and mead (C20:3) acid increased. Stearic (C18:0) acid, linoleic (C18:2), linolenic (C18:3) acid, arachidic (C20:0) acid, 11,14-eicosadienoic (C20:2) acid, arachidonic (C20:4) acid, timnodonic (C20:5) acid, adrenic (C22:4) acid, clupanodonic (C22:5) acid, and cervonic (C22:6) acid decreased as time on feed increased (Duckett et al., 1993; Realini et al., 2004).

Increasing forage intake will result in a linear decrease in saturated fatty acids and n-6:n-3 polyunsaturated fatty acid ratio along with a linear increase in polyunsaturated fatty acid: saturated fatty acid ratio and conjugated linoleic acid concentration (French et al., 2000; Rule et al., 2002, Duckett et al.; 2003; Descalzo et al., 2005; Nuernberg et al., 2005). Forage-finished cattle have a significant increase in omega-3 fatty acids (Realini et al., 2004; Noci et al., 2005; Nuernberg et al., 2005). Nuernberg et al., (2005) also stated that eicosapentaenoic acid (EPA) and docosahexaenoic

acid (DHA) are higher in cattle finished on grass-based diets. Nonruminants have higher polyunsaturated fatty acid: saturated fatty acid ratio and are lower in saturated fatty acids because the rumen nurtures the hydrogenation of dietary unsaturated fatty acids. The ruminal bacteria *Butyrivibrio fibrisolvens* is critical in the hydrogenation of linoleic acid into conjugated linoleic acids (CLA). Grass produces a more favorable rumen environment for *Butyrivibrio fibrisolvens* growth (French et al., 2000; Noci et al., 2005). Conjugated linoleic acid is found in higher levels in cattle finished on grass-based diets (Realini et al., 2004; Nuernberg et al., 2005).

Research Objectives

With growing fuel and grain prices along with consumers' increased concern over health benefits, the forage-finished cattle industry is becoming a growing enterprise. The purpose of this two year study is to show that consumer acceptable beef can be produced on winter annual forage. During the review of literature, it was noted that very few studies finished cattle on oats, rye, or ryegrass. It is hypothesized that there will be a difference among steers finished on the forage types. The steers used in the current research were finished on one of three forage treatments on a 1.42 ha paddock. Average daily gains, liveweights, carcass traits, and sensory trait will be analyzed for all three forage treatments along with years. Fatty acid profiles are evaluated for the beef from the different forage types and years.

CHAPTER III

FORAGE TYPE EFFECTS ON BEEF CATTLE PERFORMANCE AND CARCASS TRAITS

Abstract

Non-implanted Angus crossbred steers from Auburn University's Wiregrass Research and Extension Center resident herd in Headland, Alabama were used for a two-year forage-finishing study. In January 2006 (n = 18) and December 2006 (n = 18) steers were randomly assigned a forage treatment. Steers (n = 3) were grazed on duplicate 1. 42 ha paddocks of Wren's Abruzzi Rye (*Secale cereale* L.), Harrison Oats (*Avena sativa* L.), or Marshall Ryegrass (*Lolium multiflorum* Lamarck). Pen served as experimental unit for the project. The steers were humanely harvested after 85 or 95 d on test for year 1 and 145 or 152 d on test for year 2. Following harvest, ADG, liveweight, carcass traits, WBS, sensory traits, fatty acid profile, and lipid oxidative stability were analyzed from the longissimus muscle. The type of forage did not affect ($P > 0.05$) ADG, liveweight, dressing percentage, kidney, heart, and pelvic fat, marbling, yield grades, quality grades, lean maturity scores, bone maturity scores, or pH. Steers finished on ryegrass had heavier ($P < 0.05$) HCW than rye and oats. Year did not affect ($P > 0.05$) liveweight, kidney, heart, and pelvic fat, marbling, preliminary yield grade, actual yield grade, quality grades, and bone maturity scores. Year 1 had higher ($P < 0.05$) ADG, but lower ($P < 0.05$) HCW, dressing percentages, adjusted yield grades, lean maturity scores, and pH. Lipid oxidative

stability were not significant ($P > 0.05$) among forage type, year, or forage*year groups. Forage*year did not have a significant affect ($P > 0.05$) on L*, a*, or b* values for fat or lean color. Forage type did not affect ($P > 0.05$) fat L* or b*, lean L*, a*, or b* values. Ryegrass and oats had higher ($P < 0.05$) fat a* values than rye. Year did not affect ($P > 0.05$) fat L* or b*, or lean L* values. Sensory traits, WBS, and cookloss were not affected ($P > 0.05$) by forage type. Off flavor, WBS, and cookloss were not affected ($P > 0.05$) by year. Year 1 had higher ($P < 0.05$) sensory panel rating for juiciness, tenderness, and flavor intensity. Off-flavor descriptors were not affected ($P > 0.05$) by forage type or year. Year did not affect ($P > 0.05$) C8:0, C9:0, C11:0, C11:1, C12:0, C12:1, C13:0, C13:1, C14:0, C14:1, C16:1t, C20:3n3, C20:5n3, C22:0, C22:1, C22:4, C22:3, and C24:1 concentrations. Year affected ($P < 0.05$) saturated, mono, poly, omega-6, and omega-3 fatty acid concentrations.

Introduction

With the increasing cost of feed grain and fuel, the southeastern United States will need to find other alternatives for finishing cattle. The Southeast is predominately a cow-calf region with the ability to produce quality year-round forage (Sapp et al., 1998). Mature cattle are shipped to feedlots out west for finishing. Currently forage-finished beef is mostly marketed in organic or natural niche markets. Forage-finished beef is growing in popularity due to associated health benefits such as higher levels of beta-carotene (Irie, 2001; Yang et al., 2002; Descalzo et al., 2005), Vitamin E (Yang et., 2002; Descalzo et al., 2007), omega-3 polyunsaturated fatty acids, CLA, polyunsaturated:saturated fatty acid ratio, and a decreased omega-6:omega-3 ratio (French et al., 2000).

Cattle finished in a feedlot setting have higher dressing percentages, kidney, heart, and pelvic fat, ribeye areas, quality grades, HCW, external fat thickness, marbling score, and yield

grade than cattle finished on forage (Brown et al., 2005). Forage-finished beef has darker lean and yellower fat than beef concentrate-finished beef (Realini et al., 2004). Sapp et al. (1998) found that forage-finished steaks were as tender to a sensory panel and shear force as grain-finished steaks. The distinctive flavor of forage-finished beef can be attributed to beef fat (Griebenow et al. 1997). There is contradicting findings on juiciness of forage-finished beef. The amount of marbling in a study can affect the juiciness rating. Marbling assists with the ease of mastication and gives the perception of juiciness.

Materials and Methods

Experimental Design

The resident herd at Auburn University's Wiregrass Research and Extension Center in Headland, Alabama provided the Angus crossbred steers for the two-yr forage-finishing research. In fall 2004, the steers (n = 18) for the 2006 research were born. The steers (n = 18) for the 2007 research were born in fall 2005. In both years, after the steers were weaned, they were placed on bermudagrass pasture until the beginning of the study. Steers (n = 3) were grazed on duplicate 1.42 ha paddocks of Wren's Abruzzi Rye (*Secale cereale* L.), Harrison Oats (*Avena sativa* L.*)*, or Marshall Ryegrass (*Lolium multiflorum* Lamarck). Rye and Oats pastures were seeded at a rate of 68.0 kg ha^{-1} for both years. Ryegrass was seeded at a rate of 14.5 kg ha^{-1} in 2005 and 13.6 kg ha^{-1} in 2006. The steers were allowed free choice of water, salt, and a trace minerals premix.

In November 2005, 15.9 kg ha^{-1} N, 18.1 kg ha^{-1} P, and 18.1 kg ha K was applied. In December 2005 and February 2006, 31.8 kg ha^{-1} N, 22.7 kg ha^{-1} N, and 4.5 kg ha^{-1} S was applied. Phosphorus was not applied in the 2007 crop. Nitrogen was added in October 2006, November 2006, February 2007, and March 2007 at a rate of 15.9 kg ha^{-1}, 31.8 kg ha^{-1}, 15.9 kg ha^{-1}, and 9.1 kg ha^{-1}, respectively. Sulfur was applied in November 2006, February 2007, and March 2007 at a

rate of 4.5 kg ha^{-1}, 3.6 kg ha^{-1}, and 1.8 kg ha^{-1} respectively. Due to problems with the fertilizer spreader, the required amount of Nitrogen and Sulfur was not applied in February 2007 and had to be reapplied in March 2007. In each of January 2006 and December 2006, non-implanted steers were randomly assigned to different forage treatments. The average weight was 374 kg and 387 kg, respectively, for 2006 and 2007. To monitor animal growth, the steers were weighed every 28 d. The cattle were transported 24 h prior to slaughter to Lambert Powell Meat Laboratory in Auburn, Alabama, to be humanely slaughtered and USDA inspected in late April 2006 and early May 2007 over a two-wk period. In 2007 steers from the Rye group and half of the Harrison Oats group were slaughtered first due to drought causing the pasture to stop producing. The other half of the Oats group and Ryegrass group were slaughtered the following week.

Carcass Evaluation

Just after slaughter, the hot carcass weight (HCW) of each steer was taken. The carcasses were placed in a 2 ± 1º C cooler. The pH was taken one h after each steer was exsanguinated using a Thermo Orion pH meter (Orion Research, Boston, MA) with the probe placed in the Longissimus muscle on the right side of the carcass between the twelfth and thirteenth ribs. Each individual carcass was graded and pH measured 24 h after slaughter. The carcasses were ribbed between the twelfth and thirteenth ribs on the right side and trained graders evaluated each carcass (USDA, 1997). The graders adjusted the PYG as appropriate resulting in the adjusted preliminary yield grade (APYG). The quality grade was then determined using USDA Marbling charts that helped give the marbling scores.

A Hunter Miniscan XE Plus (Hunter Laboratories, Model MSXP-4500C, Reston, VA) with illuminant D65 at 10º observation angle and a 3.5-cm aperture was used to measure L* (lightness), a* (redness), and b* (yellowness) values of both fat and muscle from the longissimus between the

twelfth and thirteenth rib. Two readings were taken for both the muscle and lean color. The two readings for averaged together to give the score for muscle and lean individually. The left side of the carcass was ribbed between the twelfth and thirteenth rib and the strip loin (IMPS #180) was removed from the left side each of carcass. The first steak from the anterior end was saved for fatty acid analysis. The strip loins were then vacuum packaged and placed in a 3 ± 1º C cooler. After 11 d, the left strip loins were removed from the cooler. The first anterior steak was assigned to sensory, the second steak shelf-life and lipid analysis, and the third was for shear force. All steaks were cut 2.54 cm thick. Color of individual steaks was taken at this time and then individually vacuum packaged and placed in -25 ± 1ºC freezer until further use.

Shear

The steaks were analyzed for Warner-Bratzler shear force using the AMSA (1995) guidelines. Frozen steaks were removed from the freezer and allowed to thaw 24 h at 4ºC. A clam-shell style grill (George Foreman Grilling Machine, Lake Forest IL) was pre-heated. To measure cooking loss, a raw steak weight was taken just before the steaks were placed on the grill. Each steak was individually cooked for about six min to a medium degree of doneness (71ºC). The steaks were cooled to room temperature and re-weighed. The steaks were covered with polyvinyl chloride (PVC) film then placed at 4ºC for 24 h. A Warner-Bratzler shear machine (model 1955; GRE Manufacturing, Manhattan, KS) was used to shear the steaks. Six cores (1.3 cm in diameter) were taken from each Longmissimus muscle parallel to the muscle fibers and sheared in the middle, perpendicular across the fibers. The peak value of six cores was averaged together for further analysis.

Sensory

Six people were trained to evaluate the Longmissimus muscle of strip steaks (Cross et. al, 1978). Steaks were randomly selected from the freezer and thawed 24 h at 3 ± 1ºC. The six to eight steaks were cooked on a clam shell grill (George Foreman Grilling Machine, Lake Forest, IL) to a medium degree of doneness (71ºC). Samples were cut in 1.27 cm^2 portions and placed in warming pans until the panelist were served. The panelist evaluated the steak samples for initial and sustained juiciness, initial and sustained tenderness, flavor intensity, beef flavor, and off flavor. The samples were rated on a 1 to 8 scale with 1 being extremely dry, tough, bland and uncharacteristic and 8 being extremely juicy, tender, intense, and characteristic. The panelists were secluded in booths with red incandescent light. They were given two pieces of each sample. Before each sample, panelists cleansed their palates with saltless saltine crackers and water.

Fatty Acid Profile

After the steaks were thawed, external fat was trimmed off the steak and a two-gram sample was removed from the Longmissmus muscle. Trinonadecanoin (C19; Nu-Chek Prep, Inc. Lot #T-165-A23-R) was used as the internal standard. A chloroform-methanol procedure was used for total lipid content extraction. The samples were run on an Agilent Technologies 6890N Network GC System using a 60m long, 0.25 mm inner diameter, 0.1 μm film thickness DB-23 High Resolution Gas Chromatography column (J&W Scientific, Serial #US6489846H). Programming for the column oven temperature was 150-190ºC at 10ºC/ min, 190-230ºC at 4ºC/min and maintained at a constant 230ºC for 20 min with a 25:1 split ratio. The desired temperature for the injector and detector was 250ºC. The injector was set at 1 μL of sample. The carrier gas was helium with a flow rate of 1mL/min. FAME standards (Nu-Chek Prep, Inc.) were ran with samples each day to identify retention times of FAMES. The FAME peaks were then

integrated so that peaks could be labeled for analysis. For detailed procedure, see appendix A and B.

Lipid Oxidative Stability

A modified version of Buege and Aust (1978) thiobarbituric acid (TBA) reactive substance assay was used to evaluate lipid oxidation. Samples from Longmissimus muscle were thawed for 1 d before the assay was run. A 5 g sample was homogenized with 15mL of distilled water. A 2mL homogenate was combined with 4mL TCA/TBA reagent and 100µL BHA. Samples were heated in boiling water for 15 min and then chilled in ice water for 10 min. The samples were then filtered through Whatman #4 paper. The peak absorbance of 532 nm of the supernatant was run on a DU 730 Life Science UV/ vis Spectrophotometer (Beckman Coulter, Fullerton, Ca). Standards were run with the samples. For detailed procedure, see appendix C.

Statistical Analysis

In this experiment, steers were grazed on three different forage types over two yrs. A 2yr by 3 forage treatment factorial arrangement of a completely randomized design was used. At the end of the trial, the steers were harvested and data was collected for carcass traits, sensory, shear force, fatty acid profiles, and lipid oxidation. Data was analyzed using the GLM procedure of SAS. Least square means was found and reported in tables. Type I error set was at 5 %.

Results

Carcass Traits

Least square means for carcass data is in Table 1. Forage type by year interaction did not a have a significant ($P > 0.05$) effect on average daily gain, liveweight, hot carcass weight, dressing percentage, kidney, heart, and pelvic fat, marbling, yield grades, quality grades, lean maturity scores, bone maturity scores, or pH. The type of forage did not affect ($P > 0.05$) average daily gain,

liveweight, dressing percentage, kidney, heart, pelvic fat, marbling, preliminary yield grades, yield grades, quality grades, maturity scores, and pH. Steers finished on ryegrass had larger ($P < 0.05$) hot carcass weights (HCW) than rye and oats was not significantly ($P > 0.05$) different to either rye or ryegrass. Liveweight, kidney, heart, and pelvic fat, marbling, preliminary yield grade, actual yield grade, quality grades, and bone maturity scores were not significantly ($P > 0.05$) affected by year. Year 1 had higher ($P < 0.05$) average daily gains, but lower ($P < 0.05$) hot carcass weights, dressing percentages, adjusted yield grades, lean maturity scores, and ph. TBARS were not significant among forage*year, forage type, or year.

Color

Forage type by year interaction did not have a significant ($P > 0.05$) affect on L*, a*, or b* values for fat or lean color (Table 2). Forage type did not influence ($P > 0.05$) fat L* or b*, lean L*, a*, or b* values. Ryegrass and oats had higher fat a* values than rye. Year did not affect ($P > 0.05$) fat L*, or b*, or lean L* values. Year 2 had higher fat and lean a* and lean b* values.

Sensory

Least square means for sensory data of loin steaks are in Table 3. Forage type by year interaction and forage type did not have a significant ($P > 0.05$) affect on any sensory trait, Warner-Bratzler shear force, or cook loss. Year did not affect ($P > 0.05$) Warner-Bratzler shear force or cook loss. Year 1 had higher sensory panel rating for initial juiciness, sustained juiciness, initial tenderness, sustained tenderness, and flavor intensity. Off flavor descriptors were not affected by forage type, year, or their interaction (data not shown).

Fatty Acid Profile

Oats had higher ($P < 0.05$) C10:0 and C20:1n15 concentrations than rye or ryegrass in year 1, but forage type had no effect on either fatty acid in year 2. Forage type had an affect again ($P < 0.05$) on C16:1t and C18:2n6 concentrations. Ryegrass had higher concentrations of C16:1t than rye and oats. Ryegrass and oats has higher C18:2n6 concentrations than rye. Year did not affected ($P > 0.05$) C8:0, C9:0, C11:0, C11:1, C12:0, C12:1, C13:0, C13:1, C14:0, C14:1, C16:1t, C20:3n3, C20:5n3, C22:0, C22:1, C22:4, C22:3, and C24:1 concentrations. Year affected again ($P < 0.05$) saturated, mono, poly, omega-6, and omega-3 fatty acid concentrations. Year 1 was higher for all significantly different fatty acids except C18:1n9t and C18:1n11 concentrations.

Discussion

Carcass Traits

Average daily gain (ADG) of forage-fed cattle is lower than cattle finished on grain (Abdullah et al., 1979; Bagley et al., 1981; Nuernberg et al., 2005). Cattle in the current study had ADG that ranged from 0.69-1.91kg*d^{-1} which is similar to the findings above. There was a significant year effect. Muir et al. (1998a) stated that forage-finished cattle had lower liveweights just prior to slaughter. The liveweights of the cattle in the current study were similar to those stated by Muir et al. (1998a). The current liveweights ranged from 504.32- 557.54 kg. Researchers have stated that forage-finished beef has lower hot carcass weights than grain-finished cattle (Baublits et al., 2004; Realini et al., 2004; Cox et al., 2006; Kerth et al., 2007). However, HCW for the current study are between 298.46 and 348.36kg which is similar to Brown et al (2005) and Camfield et al. (1997) findings for cattle finished on grain. Dressing percentages for the cattle finished on oats, rye, and ryegrass are comparable to those for pasture and grain-finished cattle in the Camfield et al. (1997) study.

Ribeye area in the current study is similar to feedlot cattle in the Camfield et al. (1999) study. Bowling at al. (1977) stated that grain-finished cattle had twice as much kidney, heart, and pelvic fat than forage-finished cattle. In the current study, %KPH was similar to the findings of the forage-finished cattle from Camfield et al. (1999). It has been stated that forage-finished beef has lower marbling scores (Harrison et al., 1978; Crouse et al., 1984; Brown et al., 2005). Marbling scores of 403.33-541.67 were corresponded with Brown et al. (2005) and Camfield et al. (1999) findings. Yield grades between the current study and those of Kerth et al. (2007) are similar for ryegrass. Schaake et al. (1993) stated that forage-finished beef has lower yield and quality grades. Cattle finished in feedlots have higher quality grades (Brown et al., 2005). In the current study the quality grade was lower than the feedlot and pasture cattle in the Brown et al study. Lean and bone maturity of the cattle were comparable to Kerth et al. (2007). Among the two years of the current study, the pH differed significantly. May et al. (1992) found that 24hr pH decreased as time on feed increased. Year 2 was similar to pasture cattle in the May et al. (1992) study. Year 1 was similar to the cattle fed at least 140 days.

TBARS values in the current study were not significant. TBARS in the current study were similar to those found by Gatellier et al. (2005), Nuernberg et al. (2005), and Realini et al. (2004) for day 0.

Color

Forage-finished beef has higher L*, a*, b* values for fat color and a* and b* values for lean color and lower L* values for lean color (Realini et al., 2004). In the current study, results are similar to both the pasture group and concentrate group of the Realini et al. (2004) study. L* values for year 1 fat is similar to pasture cattle while year 2 is closer to concentrate cattle. L* values of lean for the current study corresponded with the pasture cattle from Realini et al. (2004)

except year 1 oats which was closer to the concentrate group. Findings for a* and b* values of both fat and lean are comparable to the findings of Realini et al. (2004).

Sensory

Initial and sustained juiciness and initial and sustained tenderness in the current study are higher than the grain and pasture values noted by Sapp et al. (1998). Flavor intensity and Warner Bratzler shear force values in the current study was higher than the findings by Camfield et al. (1997). Cook loss in the current study is less than those in the Camfield et al. (1997) study.

Fatty Acid Profile

Forage-finished beef have higher CLA, polyunsaturated:saturated fatty acid ratio, and a decreased omega-6:omega-3 ratio (French et al., 2000; Rule et. al, 2002; Duckett et al. 2003; Descalzo et al. 2005; Nuernberg et al. 2005). A significant increase of omega-3 fatty acids has been noted for cattle finished on forage (Realini et al., 2004; Noci et al., 2005; Nuernberg et al., 2005). Nuernberg also stated that Eicosapentaenoic Acid (EPA) and Docosahexaenoic Acid (DHA) are higher in cattle finished on grass-based diets.

Weather

Planting of the forage for Year 1 occurred during a drought. The steers were placed on the study in January instead of the intended December due to the lack of forage growth. The lack of rain could have affected the forage quality, thus affecting HCW and Dressing percentage. Average high and low temperatures were similar for both years, thus not affecting year differences for carcass traits, fat and lean color, sensory traits, and fatty acid profiles. Rainfall and temperature data can be seen in figure 1 and 2 respectively.

Implications

This two year study evaluated the differences of steers finished on oats, rye, and ryegrass. Carcass traits, fat and lean color, sensory traits, and fatty acid profiles were analyzed. Very few significant differences were found between the forage treatments. Year seemed to have more of an affect than forage treatment. This indicates that any of the three forages will produce acceptable beef.

Table 1. Least square means of carcass traits for steers finished on oats, rye, or ryegrass over a two year study.

	Year 1			Year 2			SEM	P>F		
	Oats	Rye	Ryegrass	Oats	Rye	Ryegrass		Forage	Year	Forage*Year
ADG(kg/d)	1.85	1.84	1.91	0.79	0.69	0.96	0.11	0.34	0.001	0.68
Liveweight(kg)	539.40	545.44	547.71	529.04	504.32	557.54	10.03	0.08	0.14	0.11
HCW(kg)	300.58^{bc}	298.46^{c}	309.12^{b}	323.34^{bc}	305.19^{c}	348.36^{b}	6.94	0.02	0.01	0.14
Dressing %	55.73	54.73	56.49	61.14	60.50	62.46	1.05	0.28	0.001	0.96
Ribeye Area (cm^2)	74.70^{y}	72.47^{y}	73.23^{y}	70.43^{y}	68.06^{y}	87.20^{x}	2.70	0.02	0.45	0.02
% KPH	1.58	1.83	2.00	2.25	1.58	2.83	0.26	0.08	0.10	0.16
Marbling	403.33	430.00	470.00	533.33	426.67	541.67	43.15	0.28	0.11	0.36
Preliminary Yield Grade	2.55	2.58	2.63	2.77	2.70	2.80	0.13	0.83	0.15	0.92
Lean Maturity	146.67	158.33	146.67	191.67	233.33	183.33	11.90	0.08	0.002	0.31
Bone Maturity	144.17	152.50	150.00	138.33	151.67	141.67	30.04	0.94	0.85	0.99
pH	5.49^{a}	5.49^{a}	5.47^{a}	5.75	5.87	5.67	0.06	0.39	0.02	0.55

[a] superscript denotes difference in SEM Year 1=0.091

[bc]superscripts denotes significant differences in Forage ($P < 0.05$)

[xy]superscript denotes significant differences in Forage*Year ($P < 0.05$)

Table 2. Least square means of 24 h fat and lean color of steers finished on different forage types.

	Oats	Rye	Ryegrass	SEM	P>F	Year 1	Year 2	SEM	P>F
Fat L*	72.59	72.07	71.77	3.192	2.371	74.84[a]	69.44	2.128	0.24
Fat a*	11.05[d]	9.44[e]	11.73[d]	0.380	0.042	9.94[b]	11.54	0.253	0.036
Fat b*	29.65	28.18	32.05	0.810	0.073	28.63[c]	31.29	0.540	0.066
Lean L*	34.77	33.93	33.39	0.935	0.602	34.87	33.18	0.764	0.18
Lean a*	21.34	21.04	22.72	2.126	0.841	18.32	25.09	1.736	0.033
Lean b*	21.45	20.19	23.17	1.831	0.547	19.01	24.19	1.495	0.050

[abc] denotes different SEM values for Year 1 compared to Year 2 of 3.010, 0.358, and 0.764 respectively

[de] denotes significant ($P < 0.05$) difference among forage treatments

Table 3. Least Square Means of Sensory for Loin Steaks from three different forage types.

	Oats	Rye	Ryegrass	P>F	Year 1	Year 2	P>F
Initial Juiciness	5.65 ± 0.19	5.78 ± 0.18	5.88 ± 0.178	0.70	6.02 ± 0.15	5.52 ±0.15	0.05
Sustained Juiciness	5.35 ± 0.17	5.47 ± 0.16	5.60 ± 0.155	0.58	5.77 ± 0.13	5.18 ± 0.13	0.02
Initial Tenderness	5.81 ± 0.26	5.85 ± 0.25	5.72 ± 0.244	0.93	6.29 ± 0.20	5.30 ± 0.21	0.01
Sustained Tenderness	5.35 ± 0.29	5.36 ± 0.28	5.40 ± 0.269	0.99	5.94 ± 0.22	4.81 ± 0.23	0.01
Flavor Intensity	6.15 ± 0.10	6.31 ± 0.09	6.17 ± 0.089	0.47	6.34 ± 0.07	6.08 ± 0.08	0.04
WBS	6.01 ± 0.51	6.36 ± 0.51	6.35 ± 0.512	0.87	5.76 ± 0.42	6.72 ± 0.42	0.16
Cook Loss	15.05 ± 0.94	15.88 ± 0.94	14.86 ± 0.941	0.73	15.48 ± 0.77	15.05 ± 0.77	0.70

Table 4: Least Square Means of fatty acid profiles of Longissimus Dorsi from steers finished on three different forage types during a two year study. Data presented as mg/100g.

	Year 1			Year 2				P > F		
	Oats	Rye	Ryegrass	Oats	Rye	Ryegrass	SEM	Forage	Year	Forage*Year
C8:0	0.001	0.01[a]	0.08[a]	0.01	0.03	0.001	0.02	0.36	0.38	0.14
C9:0	0.073	0.09	0.04	0.09	0.05	0.013	0.05	0.53	0.68	0.85
C10:0	0.11[y]	0.12[y]	0.24[x]	0.15[bxy]	0.07[y]	0.073[by]	0.03	0.27	0.07	0.05
C11:0	0.05	0.03	0.05	0.001	0.001	0.162	0.07	0.42	0.89	0.52
C11:1	0.001	0.001	0.08	0.001	0.001	0.001	0.03	0.42	0.36	0.42
C12:0	0.28	0.30	0.36[c]	0.86[d]	0.44	0.232[c]	0.21	0.46	0.30	0.33
C12:1	0.001	0.15	0.08	0.03	0.02	0.034	0.06	0.43	0.29	0.39
C13:0	12.82	11.29	13.39	11.68	12.33	6.369	2.36	0.59	0.26	0.28
C13:1	0.70	0.37	1.57[e]	0.15	0.08	0.087	0.48	0.50	0.10	0.49
C14:0	13.06	14.80	17.21	12.77	9.06	11.445	2.09	0.55	0.06	0.38
C14:1	3.05	3.39	3.52	2.91	2.09	3.042	0.43	0.49	0.12	0.43
C15:0	2.23	2.58	2.75	2.06	1.52	1.539	0.33	0.95	0.02	0.31
C15:1	1.57[f]	2.33[g]	2.52[h]	0.17	0.15	0.058	0.58	0.80	0.01	0.73
C16:0	180.64	161.89	188.86	151.24	116.16	132.56	19.30	0.39	0.03	0.79
C16:1t	2.03[q]	1.90[q]	4.57[p]	2.47[q]	1.76[q]	3.73[p]	0.33	0.001	0.52	0.23
C16:1	25.47	22.85	26.97	19.17	15.92	15.35	2.60	0.56	0.008	0.57
C17:0	6.45	7.26	7.23	4.55	4.62	3.75	0.92	0.86	0.01	0.70
C17:1	6.88	6.64	7.08	2.64	2.24	1.36	1.04	0.88	0.001	0.75
C18:0	103.07	90.35	102.26	76.65	65.34	68.56	7.35	0.32	0.003	0.82
C18:1n9t	0.18	0.001	0.001	0.36[i]	0.15	0.74[j]	0.15	0.24	0.04	0.20
C18:1n11	2.48	2.10	0.43	8.46	7.07	9.55	1.69	0.87	0.003	0.48
C18:1n7t	8.74	13.50	21.38	5.90	0.71	6.02	3.22	0.15	0.008	0.20
C18:1n9	285.65	223.41	268.07[k]	206.39	167.27	187.45	22.17	0.15	0.008	0.83
C18:1n7	10.88	12.51	13.83	8.40	6.36	5.99[l]	1.70	0.96	0.008	0.34
C18:2n6t	1.44	1.46	2.25	0.67	0.19	0.15	0.36	0.60	0.003	0.25
C18:2n6	34.44[q]	39.57[p]	36.56[q]	23.80[q]	32.94[p]	26.01[q]	1.65	0.01	0.001	0.43
C18:3n6	0.11	0.12	0.15	0.001	0.001	0.001[m]	0.07	0.95	0.054	0.95
C19:1	0.28	0.40	0.61	0.001	0.001	0.001	0.18	0.66	0.024	0.66

C18:3n3	11.98	12.22	13.73	9.35	10.81	11.32	0.74	0.12	0.012	0.70
C20:0	0.50	0.44	0.54	0.07	0.001	0.03	0.16	0.89	0.013	0.97
C20:1n15	0.77^{y}	1.31^{y}	3.60^{nx}	0.32^{y}	0.001^{y}	0.20^{y}	0.42	0.04	0.003	0.03
C20:1n12	1.11	0.76	0.59	0.05	0.13	0.001	0.27	0.59	0.013	0.65
C20:1n9	1.00	0.68	1.03	0.22	0.001	0.16	0.10	0.06	0.001	0.62
C20:2	3.41	2.41	3.74	1.63	1.75	0.48	0.51	0.65	0.004	0.11
C20:3	4.78	4.68	3.94	2.30	2.91	2.88	0.26	0.39	0.001	0.09
C20:4n6	19.66	18.41	18.43	12.93	16.79	12.35	1.09	0.20	0.002	0.11
C20:3n3	0.41	0.10	0.21	0.001	0.001	0.001	0.15	0.61	0.11	0.61
C20:5n3	10.66	9.76	10.61	7.03	8.86	9.69	1.23	0.59	0.12	0.49
C22:0	0.001	0.02	0.001	0.001	0.001	0.001	0.01	0.42	0.36	0.42
C22:1	0.001	0.001	0.001	0.001	0.001	0.09	0.04	0.42	0.36	0.42
C22:2	0.04	0.10	0.15	0.001	0.001	0.001	0.04	0.50	0.03	0.50
C22:4	1.15	1.18	1.36	1.46	1.09	0.75	0.54	0.90	0.78	0.72
C22:3	0.38	0.19	0.27	0.08	0.13	0.18	0.11	0.80	0.15	0.56
C24:0	0.38	0.46	0.61	0.001	0.001	0.001	0.18	0.82	0.02	0.82
C22:5	15.11	13.81	15.23	10.18	12.07	11.19	0.54	0.61	0.001	0.06
C22:6n3	2.26	1.52	2.12	1.16	1.12	1.09	0.33	0.51	0.02	0.54
C24:1	0.001	0.01	0.09	0.001	0.001	0.001	0.04	0.47	0.30	0.47
Sat	319.67	289.64	333.60	260.06	209.61	224.68	29.55	0.42	0.01	0.72
Mono	350.10	291.40	333.91	257.62	203.94	233.29	29.86	0.24	0.01	0.98
Poly	105.84	105.52	108.74	70.59	88.67	76.07	4.29	0.20	0.001	0.15
Omega-6	83.16	84.30	88.94	59.03	67.60	57.58	4.22	0.55	0.001	0.29
Omega-3	25.31	23.59	26.67	17.54	20.79	22.09	2.06	0.39	0.02	0.51
n-6:n-3	3.35	3.60	3.48	3.62	3.38	2.78	0.40	0.61	0.54	0.52

^{abcdefghijklmn} superscripts for SEM equals 0.02, 0.07, 0.22, 0.23, 0.51, 0.77, 0.87, 0.73, 0.19, 0.17, 23.25, 1.78, 0.07, and 0.44 respectively.

^{qp} superscripts denotes significant difference among forage type means ($P < 0.05$)

^{xy} superscripts denotes significant difference among forage* year interaction means ($P < 0.05$)

Figure 1: Monthly averages of rainfall for Headland, Alabama during a two year forage finishing study

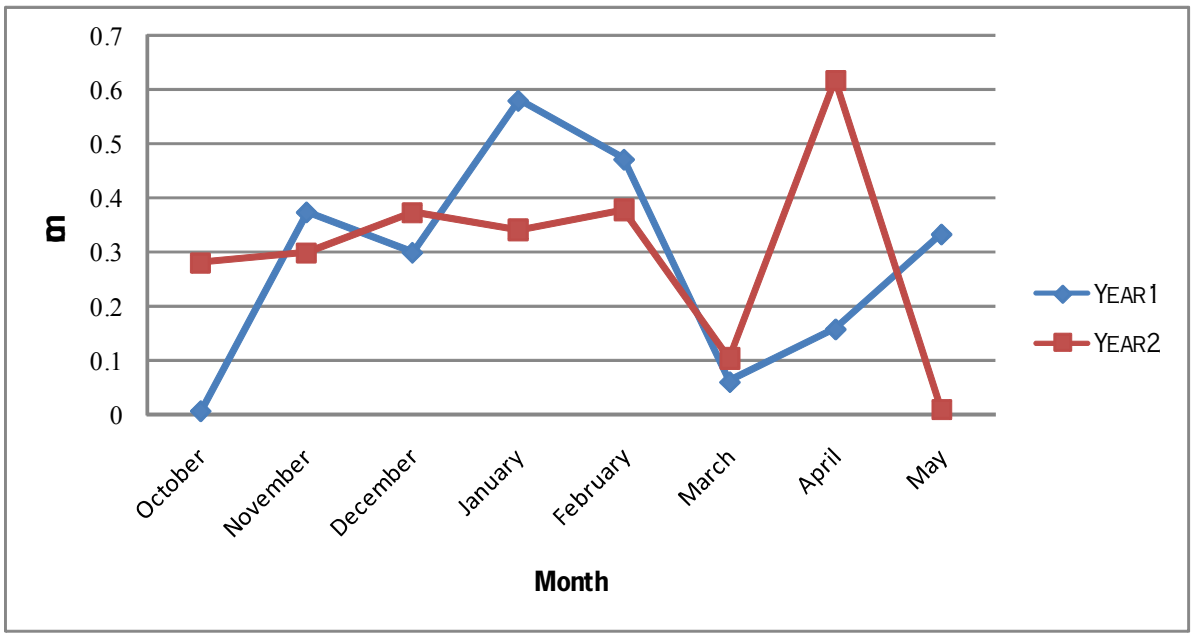
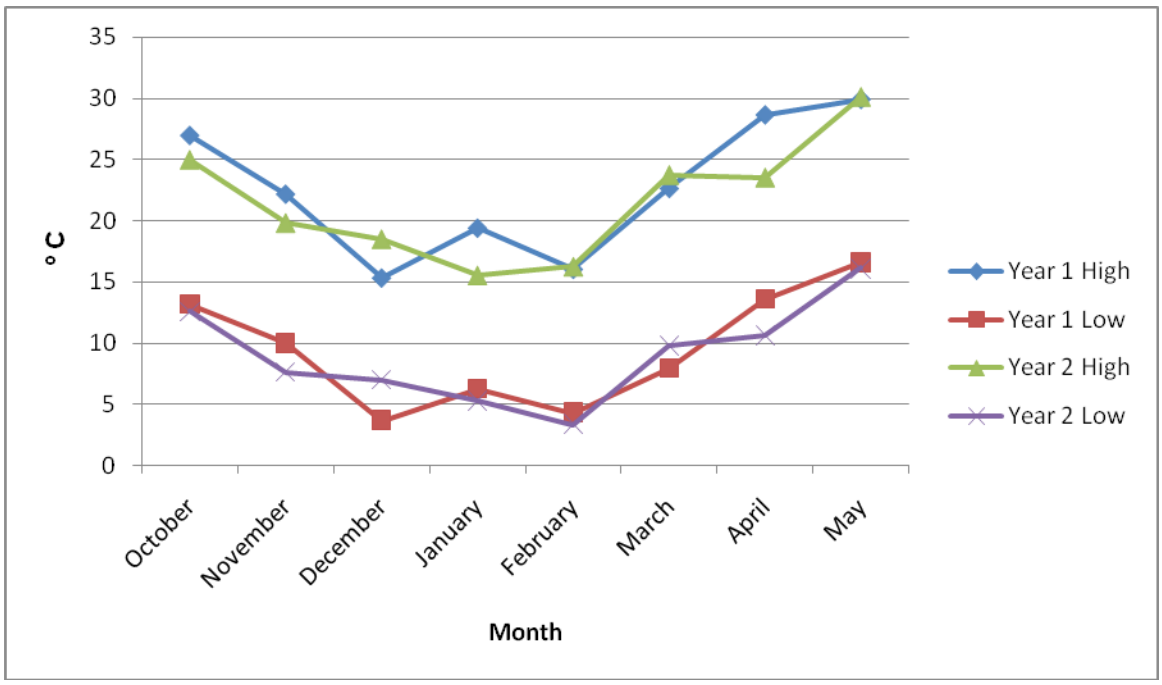


Figure 2: Average monthly temperatures for Headland, Alabama during a two year forage finishing study.



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APPENDICES

APPENDIX I:

Carcass Data Sheet

Data Collector:_____ Date:_____

Animal ID: _____ Pen #:_____

Treatment: _____

Live Wt.	
HCW	
Dressing %	
%KPH	
REA	
Marbling	
PYG	
Adjusted PYG	
Lean Maturity	
Bone Maturity	
Quality Grade	
Lean Color	
Fat Color	
pH	

APPENDIX II:

Sensory Evaluation

1) Thaw steaks 24h at 2-5ºC
2) Cook samples on clam-shell grill to a medium degree of doneness of 71º C (about 7 min)
3) Take temperature of sample in the geometric center
4) Trim fat and connective tissue from the steak. The sample should be in a square or rectangular shape
5) Cut 1.27cm x 1.27cm pieces of the sample
6) Place samples in preheated covered pans that contain sand and kept in a warming oven at about 72ºC
7) Take weight and temperature of each sample immediately before and after it is on the clam-shell grill

Procedure adapted from:

AMSA. 1995. Research guidelines for cookery, sensory evaluation and instrumental tenderness measurement of fresh meat. National Livestock and Meat Board, Chicago, IL.

Kerth, C.R, L.K. Blair-Kerth, and W.R. Jones. 2003. Warner-Bratzler Shear force repeatability in beef Longissimus steaks cooked with a convection oven, broiler, or clam-shell grill. J. Food Science. 68:668-670.

APPENDIX III:

TRAINED SENSORY EVALUATION FORM

Name_____ Date_____ Time: _____ Project_____

Sample No.	Initial Juiciness	Sustained Juiciness	Initial Tenderness	Sustained Tenderness	Flavor Intensity	Off Flavor	Off Descriptor

Juiciness	Tenderness	Flavor intensity	Off Flavor	Off flavor Descriptors
8=Extremely juicy	8=Extremely tender	8=Extremely intense beef	8=Extreme off flavor	8=Metallic
7=Very juicy	7=Very tender	7=Very intense beef	7=Intense off flavor	7=Salty
6=Moderately juicy	6=Moderately tender	6=Moderately intense beef	6=Very off flavor	6=Livery
5=Slightly juicy	5=Slightly tender	5=Slightly intense beef	5=Moderate off flavor	5=Grassy
4=Slightly dry	4=Slightly tough	4=Slightly bland	4=Modest off flavor	4=Bitter
3=Moderately dry	3=Moderately tough	3=Moderately bland	3=Small off flavor	3=Bloody
2=Very dry	2=Very tough	2=Very bland	2=Slight off flavor	2=Rancid
1=Extremely dry	1=Extremely tough	1=Extremely bland	1=No off flavor	1=Other – Explain

APPENDIX IV:

Warner-Bratzler Shear Evaluation

1) Cook samples as described in Appendix II (sensory) steps 1 thru 4
2) Cool samples 24h at 2-5ºC. Cover samples with poly-vinyl chloride (PVC) film
3) Take 6 random 1.27cm cores from the sample going parallel to muscle fibers and insuring no fat or connective tissue in sample
4) Place core in WBS machine and observe maximum peak value
5) Crosshead speed should be 200-250 mm/ min

Procedure adapted from:

AMSA. 1995. Research guidelines for cookery, sensory evaluation and instrumental tenderness measurement of fresh meat. National Livestock and Meat Board, Chicago, IL.

Kerth, C.R, L.K. Blair-Kerth, and W.R. Jones. 2003. Warner-Bratzler Shear force repeatability in beef Longissimus steaks cooked with a convection oven, broiler, or clam-shell grill. J. Food Science. 68:668-670.

APPENDIX V:

Lipid Extraction for FAME Analysis

1) Thaw tissue and mince with scalpel (100 strokes in many directions). Place 2g into 80ml beaker which contains 40ml of 0.25mg/mL concentration of chloroform: methanol mixture (21.6ml cholorform: 5.0ml C19:0 standard: 13.3 methanol)
2) Homogenize for 5min. on ice with gas Nitrogen feed to flush out atmospheric oxygen
3) Filter homogenate into 50mL graduated cylinders using #4 Whatman Filter Paper. Rinse homogenizing container with 2:1 chloroform: methanol mixture then filter into graduated cylinder. Bring volume of homogenate up to 40ml.
4) Keep homogenate mixed using slow hand movements to insure little induce oxidation. Transfer 20ml into 2 20X125 Pyrex culture tubes.
5) Add 4.6ml of a 0.1M NaCl solution. Flush tube with Nitrogen and cap. Vortex for 5 min and centrifuge for 10 min at 2,000 rpm.
6) Remove top aqueous layer. Dry bottom organic layer with nitrogen evaporator at 40ºC. Reconstitute sample with 3ml Dichloromethane.
7) Weigh 2 empty 13X100ml pyrex culture tubes individually with silicone tape around the threaded type. Transfer sample in into weighed tubes and dry again with nitrogen evaporator. Reweigh sample.

APPENDIX VI:

Fatty Acid Methylation

1) Add 1ml Boron Trifluoride solution of 12% in methanol into each tube. Flush with nitrogen, cap, and vortex for 1 min.
2) Place tube into heating block at 110-115ºC for 30 min. Check periodically to ensure the caps are not leaking BF_3
3) Place tubes in ice bath for 5 min.
4) Add 2ml Pentane and 1ml DW, flush with nitrogen, cap, and vortex for 15s.
5) Centrifuge for 5min at 2,000 rpm
6) Collect top layer and put into pre-weighed 13X100 Pyrex culture tube, add 3ml Diochloromethane and dry on nitrogen evaporator
7) Reweigh dry sample in tube. Re-suspend in 200µl Diochloromethane for Gas Chromatography injection

APPENDIX VII:

Thiobarbituric Reactive Substance (TBA) Assay

Reagents:

1) TCA/TBA stock solution: 15% TCA and 20mM TBA(MW 144.15) reagent in DDW. Dissolve 2.88g TBA in warm dionized water first, add TCA (150g), and then add DW to the 1L mark. (1L last about 100 samples in duplicate)
2) BHA: Make 10% stock solution by dissolving in 90% ethanol. Make 500ml batches.
3) TEP standard: $1*10^{-3}$ 1,3,3-tetra-ethoxypropane in DW. Dilute 0.5ml TEP with 499.5ml DW, and dilute resulting solution 1:2.96 (TEP: DW) with DW

Procedure:

1) Mince 5g of meat and add to 15ml DW
2) Homogenize sample
3) Add 2ml homogenate to 16X100mm Pyrex culture tube along with 100µl BHA and 4ml TCA/TBA reagent. Vortex.
4) Cover with aluminum foil and heat for 15 min in boiling water.
5) Cool for 10 min in ice cold water
6) Vortex and filter with Whatman Paper
7) Read absorbance of supernatant at 531nm against blank that contains all the reagents minus sample

Malonaldehyde standard curve:

1) Label 6 16X100mm Pyrex culture tubes as 0,5,10,20,30,40,50

2) Add the following amounts to each tube:

	TEP	DW	Set pipettor on:
0	0µl	2000ml	1000 (twice)
5	10µl	1990ml	995 (twice)
10	20µl	1980ml	990 (twice)
20	40µl	1960ml	980 (twice)
30	60µl	1940ml	970 (twice)
40	80µl	1920ml	960 (twice)
50	100µl	1900ml	950 (twice)

3) Add 4ml TBA/TCA to each tube and vortex

4) Cover with aluminum foil and heat tubes in boiling water for 15min

5) Cool in ice cold water for 20min

6) Vortex

7) Read absorbance of the standard against a blank at 531nm

Procedure adapted from:

Assay of lipid oxidation in muscle samples
Methods in Enzymol. 52: 302, AP