

COMPARISON OF SENSORY CHARACTERISTICS, FATTY ACID PROFILES,  
PROXIMATE ANALYSIS, AND SHELF-LIFE STABILITY OF AKAUSHI BEEF,  
COMMODITY PRIME BEEF, AND TOP CHOICE BRANDED BEEF

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## ABSTRACT

Akaushi, prime and top choice branded beef (TCB) were evaluated for multiple quality attributes. Akaushi had the highest lipid oxidation followed by TCB, and prime ( $P < 0.0001$ ). Akaushi and prime had similar initial and sustained juiciness; both were juicier than TCB ( $P < 0.05$ ). Akaushi was more tender than TCB ( $P \leq 0.05$ ). Evaluating flavor intensity and overall acceptability, Akaushi and prime were similar, and both more favorable than TCB. Akaushi and prime had lower protein percentage compared to TCB ( $P < 0.0001$ ). TCB had the highest moisture percentage, followed by Akaushi, and prime ( $P < 0.0001$ ). Prime had the highest fat and collagen percentages, followed by Akaushi, and TCB ( $P < 0.0001$ ). TCB had the highest polyunsaturated:saturated fatty acid ratio ( $P < 0.0001$ ). Akaushi had the highest monounsaturated:saturated fatty acid ratio ( $P < 0.0001$ ).

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## INTRODUCTION

Akaushi is a Japanese breed of cattle that was introduced into the United States in 1994 (Hawkes, 2013). The breed is known for marbling ability and more monounsaturated fatty acids (Ishmael, 2014). The first Akaushi cattle introduced were eight females and three bulls. This breed of cattle is categorized as *Bos taurus*, and look comparable to the *Bos taurus* cattle in the United States. Visually, Akaushi are red in coat color (Halladay, 2012; Hawkes, 2013; Honda et al., 2006). An attractive trait for producers that the Akaushi has is high quality beef that the breed produces, as well as not sacrificing production efficiencies on the live side of the beef production spectrum for the quality premiums (Ishmael, 2014).

Quality is one of the primary factors evaluated when the value of a beef carcass is determined. Premiums can be achieved from high quality beef; and on the contrary, poor quality beef will receive substantial discounts. According to the National Cattlemen's Beef Association (NCBA) (2012a), food safety was ranked first, and eating satisfaction was ranked second as areas needing improvement to increase the value of beef products. The principal two elements of eating satisfaction that every segment of the beef industry acknowledges is tenderness and flavor (NCBA, 2012a). Consumers are aware of tenderness, juiciness, and flavor when consuming beef. Consumers could detect greater flavor, tenderness, and juiciness in higher degrees of marbling, as well as lesser flavor, tenderness, and juiciness in lower degrees of marbling (O'Quinn et al., 2012). Consequently, consumer acceptance has been shown to be affected by the quality of a meat product (Miller et al., 1995; Savell et al., 1987; Savell et al., 1989; Smith et al., 1987). According to Miller et al.

(2001), consumers could distinguish differences in Warner-Bratzler shear force values and were agreeable to pay a premium for steaks of greater tenderness.

Tenderness has been studied abundantly over the past 20 years as it is significant to the eating satisfaction of beef. Multiple studies indicate that tenderness is the most imperative factor when analyzing palatability (Savell et al., 1987; Miller et al., 1995; Miller et al., 2001). Other authors feel flavor has been presented as equally or more important than tenderness in more than one study (Killinger et al. 2004; Neeley et al. 1998). More specifically, flavor has been acknowledged for its prominence in consumption of beef, and has been studied more extensively as of recent (Adhikari et al., 2011; Kerth and Miller, 2015; Miller et al., 2015). The American Akaushi Association (2014) claims that Akaushi beef is very juicy and flavorful as a result of increased intramuscular fat and fatty acid differences. Furthermore, Akaushi are said to have fat that contains a higher proportion of oleic fatty acid, conjugated linoleic acid, and composite monounsaturated fat (Hawkes, 2013; Gotoh et al., 2014; American Akaushi Association, 2014).

As stated by Overstreet (2015), a health-conscious consumer is becoming a more prominent portion of the consumer population. Two essential fatty acids, linolenic acid and linoleic acid are considered the only essential fatty acids for humans (Ghisolfi and Putet, 1992). Beef provides a portion of the essential fatty acids needed in the human diet; in addition, some polyunsaturated fatty acids found in beef can be converted to essential fatty acids in the human body (Whetsell et al., 2003). Furthermore, consumption of these types of fats are linked to health benefits including deterring many diseases such as heart diseases, atherosclerosis, cancer, body lipid content, and immune system deficiencies (Whetsell et al., 2003). Initial claims indicate Akaushi beef would be a very beneficial product not only from

a flavor standpoint, but also as an option for consumers wanting a less saturated fat profile in the diet. However, there is little research comparing this relatively new beef type to products presently marketed in the United States.

### **Objective**

The objective for this study was to compare Akaushi beef with commodity prime beef and top choice branded beef using trained sensory panel attributes, Warner-Bratzler Shear Force values, fatty acid profiles, lipid stability, and proximate analysis components.

## LITERATURE REVIEW

### **Akaushi**

The Akaushi breed of cattle, which originates in Japan, is a *Bos taurus* breed type that is known for its marbling ability and fat that has a healthier fatty acid profile (Ishmael, 2014). The Akaushi is a breed of cattle that is a relatively new breed to the United States. The breed has been developed over the past 100 years by the Japanese government due to selection utilizing extensive data sets (Shibu, 2011). The breed became official in 1944 (Twinwood Cattle Company, 2011). In 1994, eight Akaushi females and three bulls were introduced to the United States and all of the Akaushi cattle in the United States can be traced back to that nucleus of cattle (Hawkes, 2013). The cattle were allowed to be imported through a loophole in the Trade Act of 1992 between the United States and Japan (Twinwood Cattle Company, 2011). In 1920, two distinct strains were identified; the Kumamoto and the Kochi. The Kumamoto cattle were heavily influenced by Simmental and Devon genetics, with a small influence of Korean cattle. On the other hand, the Kochi is more influenced genetically by the original native Korean cattle (Hawkes, 2013; Honda et al., 2006; Twinwood Cattle Company, 2011).

Akaushi cattle are red in coat color, very similar in appearance to the American *Bos taurus* cattle, in addition to performing similarly in terms of beef production (Halladay, 2012; Hawkes, 2013; Honda et al., 2006). Japan has four major breeds of beef cattle, which include the Akaushi, or the Japanese Brown, Kuroishi or the Japanese Black, the Japanese Polled and the Japanese Shorthorn (Ishmael, 2014; Gotoh, et al. 2014). The Japanese Black is the most numerous breed of cattle in Japan and is similar to the Akaushi (Shibu, 2011). The Akaushi

breed claims to be a highly fertile breed of cattle that are correct in their skeletal structure, will gain and convert in the feedlot, grade and yield on the rail, and produce a high quality end product for the consumer (Halladay, 2012). High quality beef is the attribute that draws producers to the Akaushi, as well as the indication of sustaining efficiencies in production. Furthermore, in the United States, far more crossbred Akaushi cattle are used in beef production than full-bloods.

Heartbrand Beef® is the only branded beef marketing program for Akaushi influenced cattle in the United States. The USDA-Agriculture Marketing Service (AMS) has a schedule termed the G-schedule that contains all of the information and specifications needed for every certified branded beef program in the United States. Heartbrand Beef® currently has two G-schedule branded beef programs through USDA-AMS. The G98 program is specific to International trade, and the G99 program is for domestic trade. To initially be considered for the Heartbrand Beef® branded program, the cattle are required to be genetically traced to Akaushi parentage. Marketing the beef as Akaushi requires the cattle to meet all of the requirements including: at least 93.75% Akaushi, as well as traceable to two parents or four grandparents. Additionally, to be marketed as Akaushi cross, the cattle need to be at least 50% Akaushi and traceable to one parent or two grandparents (USDA-AMS, 2015a; USDA-AMS, 2015b). Once genetically verified, the cattle must meet all of the additional requirements: must be non-hormone treated; A or B maturity in regards to lean color, texture, firmness, and skeletal maturity; practically free of capillary rupture in the ribeye muscle; free of dark cutting characteristics; a hump two inches or less in height; and moderately thick or heavier muscled. Finally, according to the maturity scores, the cattle

need to have medium or fine marbling texture that is in the complete range of USDA Prime to USDA Choice (USDA-AMS, 2015a; USDA-AMS, 2015b).

### **Quality Grades**

Quality is an essential factor in determining the value of beef. United States Department of Agriculture (USDA) Quality Grades are composed of the components marbling and maturity. Beef quality can achieve a premium from consumers on the high end of the spectrum or a discount on the lower end. Therefore, quality is a very important factor to consider when assessing value in the beef business. Beef cattle in Japan, including the Akaushi, tend to produce a product with a much greater amount of marbling (American Wagyu Association, 2014). Quality factors that consumers are aware of are the elements tenderness, juiciness and flavor. Higher degrees of marbling have been correlated with greater flavor, tenderness and juiciness in consumers (Garmyn et al., 2011; Lorenzen et al., 2003; O'Quinn et al., 2012). Additionally, quality plays a vital role in consumer acceptance of a meat product (Miller et al., 1995; Savell et al., 1987; Savell et al., 1989; Smith et al., 1987). Tenderness and flavor are two of the most influential factors that affect palatability.

### **Yield Grades**

Along with quality, another large value determining attribute is USDA Yield Grade. USDA uses yield grades to specify the ratio of lean-to-fat that is expected from a carcass (USDA-FSIS, 2014). This lean-to-fat ratio will give the expected percentage of boneless, closely trimmed retail cuts from the wholesale primals of a beef carcass (USDA-AMS, 1997). The four factors of ribeye area (REA), carcass weight, fat thickness (FT), and percent kidney, pelvic and heart fat are utilized to determine the yield grade of the carcass. The REA

and FT are taken on the ribbed surface between the twelfth and thirteenth ribs (USDA-AMS, 1997). Although not directly related to the consumer, yields are exceptionally vital to beef processors.

### **Proximate**

The five primary components of meat are moisture, protein, fat, carbohydrates, and ash (Jensen, et al., 2004a). The amounts of moisture, protein and ash are common to be inversely correlated to the amount of fat in a sample (Jensen, et al., 2004a). According to Brackebusch et al. (1991), proximate values for beef carcasses identified as low, intermediate, or high marbling produced a linear regression for the values of fat, protein, and moisture. Low marbling carcasses were considered to be slight marbling or less; intermediate marbling carcasses were comprised of marbling scores small, modest and moderate; and high marbling carcasses were considered slightly abundant or greater (Brackebusch et al., 1991). Low marbling carcasses produced an average of 4.89% fat, 73.21% moisture and 22.13% protein (Brackebusch et al., 1991). Intermediate carcasses had an average of 8.37% fat, 70.04% moisture and 21.35% protein (Brackebusch et al., 1991). Lastly, high marbling carcasses yielded 12.57% fat, 66.60% moisture and 20.54% protein (Brackebusch et al., 1991).

### **Sensory**

The traits evaluated by panelists in a sensory analysis are tenderness, juiciness, flavor, and overall acceptance (AMSA, 1995). This is meant to give an idea to researchers what type of eating experience a consumer might have. Each of the components represented in a sensory panel are important to palatability as a whole for consumers.

## Palatability

Palatability is composed of elements that affect the senses which include appearance, tenderness, juiciness, flavor, and aroma (Aberle, et al., 2001f). Expected palatability is determined by USDA Quality Grades (USDA-AMS, 1997). The quality grades for beef are USDA Prime, Choice, Select, Standard, Commercial, Cutter, and Canner (USDA-AMS, 1997). Younger cattle qualify for the first four grades listed, but mostly the first three are seen by consumers (USDA-AMS 1997; Aberle, et al., 2001e). As the marbling score increases, the expected palatability increases as well (O'Quinn et al., 2012).

## Tenderness

Tenderness is subject to numerous factors including the amount and type of connective tissue, and protein tenderness. Consumers look at tenderness more from a perceived view as they consume the product. Perception of tenderness is comprised of ease of fragmentation, resistance to tooth pressure, softness to tongue and cheek, mealiness, adhesion, and residue after chewing (Aberle, et al., 2001f). Tenderness is also evaluated in two common objective analyses: Warner-Bratzler Shear Force (WBSF) and Slice Shear (SS) (Shackelford et al., 1999). The WBSF measures the amount of pressure it takes to sever a predetermined size of cooked meat. This is helpful by creating a measuring system that can allow uniform comparisons to be made between samples, utilizing objective measurements (Kerth, 2013b). Beef tenderness for WBSF can range from tough (> 5.7kg) to tender (< 3.0kg) (Miller, et al., 2001).

Connective tissue is one of the principal factors that influences tenderness. In reference to tenderness, the background effect is attributed to the connective tissue type and



location in the muscle (Calkins and Sullivan, 2007). Connective tissue can be found in three places in the muscle system, the endomysium, perimysium and epimysium. Endomysium surrounds muscle fibers, perimysium surrounds muscle bundles, and epimysium surrounds whole muscles (Aberle, et al., 2001a). Endomysium is the easiest to remove of the three, as it is largest and most visible. Furthermore, connective tissue is classified into three groups known as elastin, reticulin, and collagen (Pearson and Gillett, 1996a).

Collagen is the most abundant protein found in the body and is relatively soluble in younger animals (Calkins and Sullivan, 2007). As an animal ages, the number and strength of intermolecular cross linkages increases. These collagen intermolecular cross linkages are insoluble and decrease the tenderness of meat (Calkins and Sullivan, 2007). Elastin is insoluble and is found in the cervical ligament, artery walls, and in some organs. In comparison to collagen, elastin, and reticulin are less numerous in skeletal muscle and carcasses (Aberle, et al., 2001a).

Protein tenderness is another major factor that impacts tenderness. Protein tenderness is composed of the contractile state as well as the integrity of the muscle fiber matrix (Kerth, 2013b). Contractile state is seen through the actomyosin effect and is due to the state of the sarcomeres. A sarcomere is the small contractile element of muscle (Jensen, et al., 2004c). Longer sarcomeres produce a more tender product and shorter, more contracted sarcomeres produce a tougher meat product (Kerth, 2013b). Chilling carcasses at ideal temperatures, in addition to ageing carcasses or wholesale cuts is the easiest way to increase protein tenderness (Calkins and Sullivan, 2007). This degradation of the matrix integrity is primarily seen through ageing and proteolytic enzymes (Kerth, 2013b).

Fiber type is a factor that indirectly influences tenderness. In some research, the fiber type of beef cattle has been correlated with tenderness and marbling (Calkins et al., 1981). Muscle fiber type in meat animals is composed of two major fiber types, in addition to two intermediate types. The two major types are fast twitch also referred to as white muscle fiber types and slow twitch referred to as red muscle fiber types. White muscle fiber types are more glycolytic in metabolism type (Kerth, 2013a). Red muscle fibers are more oxidative in nature and are further predestined for the deposition of marbling (Aberle, et al., 2001a). In today's industry, white muscle fiber types are visually selected for in livestock, as animals with a heavier muscled phenotype are higher in the proportion of white muscle fibers (Kerth, 2013a). High growth and conversion efficient meat animals are not associated with tenderness, juiciness and taste as compared to slower growing and less conversion efficient animals (Jensen, et al., 2004c). This has in some cases decreased the quality of meat produced (Lee, et al., 2010). The metabolism of the fiber type has been associated with marbling and tenderness (Joo et al., 2013). The increased ability for these cattle to deposit intramuscular fat could be related to the predominant muscle fiber type of the cattle. Muscle fiber typing has not been studied in the Akaushi breed and could be very beneficial. This factor can be utilized when looking at producing quality meat (Schreurs, et al., 2008).

### Juiciness

Juiciness is an underlying factor that imparts initial and overall impressions on a consumer (Aberle, et al., 2001f). Juiciness is composed of fat and water that is in turn correlated with the amount of intramuscular fat present and water holding capacity (Aberle et al., 2001f). Degree of intramuscular fat in conjunction with maturity corresponds with USDA

Quality Grades (USDA-AMS, 1997). The maturity grades are A, B, C, D, and E. Young cattle are eligible for A and B, while cattle greater than around 42 months are only eligible for C, D, and E (USDA-AMS, 1997). Water holding capacity is the quantity of water that will be retained in the meat when acted upon with outside forces such as cutting, grinding, heating, or pressure (Aberle, et al., 2001c). The most tightly held form of water is known as bound water. Free water is not held in the muscle very easily and usually takes the form of weep or purge. Lastly, immobilized water is loosely held by charges in the muscle (Aberle, et al., 2001c).

## Flavor

Flavor of meat is determined by a multitude of factors, many of which are still not understood including: species, age of animal, cooking method, addition of spices and ingredients, amount and type of fat present, postmortem ageing, and pre-slaughter feeding (Pearson and Gillett, 1996b). The type of fat present is very characteristic to the flavor of meat. This is especially evident in the comparison of red meat species (beef, lamb, and pork) (Pearson and Gillett, 1996b). In addition, the animal's diet can affect the type of fat deposited. This can be seen in the different flavor profiles between grain and grass fed beef (Wood et al., 2008). Another factor to consider is volatile compounds. Volatiles are the oxidized products of lipids that impart powerful influences on flavor (Kerth, 2013c). Volatile compounds also have an effect on the flavor of meat, mostly seen negatively (Kerth, 2013c). The initial product of oxidation is hydroperoxides, which tend to be temporary, colorless and flavorless; in addition, they are measured utilizing peroxide values (Kerth, 2013c). The more significant portion of secondary oxidation is the formation of ketones, aldehydes,

hydrocarbons and alcohols (Kerth, 2013c). These are the products that are heavily associated with the negative flavors. Flavor is a component of palatability that is dependent on many factors, and each of these factors needs to be assessed as a whole to dictate how flavor will dictate palatability (Aberle, et al., 2001f).

### **Fatty Acids**

Another element to the Akaushi breed is that the fat found in Akaushi beef is claimed to include a higher proportion of oleic fatty acid, conjugated linoleic acid and monounsaturated fat (Hawkes, 2013; Gotoh et al., 2014; American Akaushi Association, 2014). These unique characteristics are not found in any other red meat animals, and are shown to be more beneficial to human health (Gazdziak, 2014; Gilmore, et al., 2013). These fatty acid features have been shown to be healthier for the human diet by reducing low density lipoprotein, which can pose problems to heart health (Gilmore et al., 2013). A greater amount of more beneficial fat to the human body is seen when the proportion of low density lipoprotein (LDL) is reduced, without reducing the number of high density lipoprotein (HDL) (Gilmore, et al., 2013). Some of the common fatty acids found in beef muscle include oleic, palmitic, stearic, palmitoleic, myristic, and linoleic (Kerth, 2013c). The saturated fatty acids include palmitic, stearic and myristic (Kerth, 2013c). The monounsaturated fatty acids are oleic and palmitoleic (Kerth, 2013c). Lastly, the polyunsaturated fatty acids include linoleic,  $\alpha$  linolenic and arachidonic (Kerth, 2013c). The common ratio of monounsaturated fatty acids to saturated fatty acids (MUFA:SFA) for beef is 1:1; however, Japanese Black cattle raised in Japan achieve a ratio of 2.6:1 (Jensen, et al., 2004b).

## **Shelf-Life**

Shelf-life is known as the amount of time from packaging a product until the product is undesirable to a consumer; therefore, it remains an important factor to consider in the meat industry (Brooks, 2007; Delmore, 2009). There are many factors that influence the shelf-life of a meat product, some of which are packaging, temperature, time, amount of fat and fat composition (Delmore, 2009). Relatively speaking, packaging type, temperature, and time are easier to influence as opposed to fat composition. The degree of saturation is an aspect to consider, when speaking of oxidation. Polyunsaturated fatty acids (PUFA) oxidize faster than saturated fatty acids (SFA) (Wood et al., 2008). Therefore, a product with a higher degree of saturated fats would be more beneficial from a shelf-life standpoint. However, health concerns are inverse as the 2015-2020 Dietary Guidelines for Americans recommends a reduced intake of SFA and an increased intake of PUFA and Monounsaturated Fatty Acids (MUFA) (U.S. Department of Health and Human Services and U.S. Department of Agriculture, 2015). There are many other elements to consider when looking at extending the shelf-life of increased PUFA and MUFA meat products such as the utilization of Vitamin E antemortem or various antioxidants postmortem (Aberle et al. 2001b, 2001d).

## **Economics**

There are many sectors in the beef industry producing cattle destined for slaughter, and with each sector come different economic conditions. Many different factors come into play with each sector and vary from operation to operation; in Table 1, wholesale beef prices are compared for consistency purposes. Beef is sold as boxed beef, which contains several cuts of the same anatomical origin; additionally, beef is sold on a value base, therefore

grouping together cuts of similar quality and trim level (Savell et al., 1995). Because of this, major price differentials can be seen between different quality levels, branded names, cuts, and trim levels of boxed beef. The price of the product varies depending if there is a value added component of quality or if it belongs to a branded beef program. For example, USDA Prime product will have a premium over USDA Choice product. Table 1 has a snapshot of prices of several cuts for each of the beef type treatments utilized in the project.

**Table 1.** Snapshot of pricing per pound for multiple middle meats

Beef Type Treatment	Quality Grade	Ribeye Roll #112A <sup>b</sup>	Strip Loin #180 <sup>b</sup>	Tenderloin # 189A <sup>b</sup>	Sirloin #184 <sup>b</sup>
Akaushi <sup>a</sup>	Upper 2/3 Choice	\$10.90	\$9.90 <sup>c</sup>	\$18.90	\$5.50 <sup>d</sup>
Prime <sup>e</sup>	Lower 1/3 Prime	\$9.42	\$9.83	\$12.12	\$3.61
TCB <sup>ef</sup>	Upper 2/3 Choice	\$6.73	\$4.90	\$9.49	\$3.31

<sup>a</sup>Akaushi prices were retrieved from a wholesale representative.

<sup>b</sup>Cut numbers represent specific cuts utilizing the IMPS system.

<sup>c</sup>Akaushi price refers to a 1x1, whereas TCB and Prime refer to a 0x1

<sup>d</sup>Akaushi price refers to a Top Butt Cap-off, versus a whole Top Butt.

<sup>e</sup>Prime and TCB prices were retrieved from [ams.usda.gov](http://ams.usda.gov), on September 19. National weekly Boxed Beef Cuts and Branded Product and National Weekly Boxed Beef Cuts for Prime Product were used.

<sup>f</sup>TCB=top choice branded beef

### Value of Proposed Research

For the first time since its inception in 1991, the 2011 National Beef Quality Audit (NBQA) evaluated end-users “willingness to pay”. The NBQA 2011 found that the two most important factors found among the end-users were food safety and eating satisfaction

(NCBA, 2013a). According to the 2011 NBQA, quality as defined by cattle producers, of which were dominated by cow-calf producers, is the production of safe, wholesome and healthy animals and beef products (NCBA, 2013b). A majority of these producers utilized vaccination programs as well as stockmanship and animal handling skills to positively affect quality (NCBA, 2013b). On the other hand, industry segments closer tied to the consumers place a larger emphasis on food safety and eating satisfaction rather than carcass weight, carcass cutability and animal genetics (NCBA, 2012b). USDA Quality Grades for beef are a prediction of palatability traits such as tenderness, juiciness, and flavor (USDA-AMS, 1997). The 2011 NBQA identified a list of barriers to continued improvement in the beef industry, one of which included differing definitions of quality and value across segments. More specifically, the recommendations to reduce these barriers and in turn help to improve beef profitability are to increase funding to improve eating satisfaction and the use of genetics to optimize cutability and palatability (NCBA, 2012b). Quality grade improvement starts at the cow-calf level with the control of breed and genetic inputs (NCBA, 2013a). Moreover, when emphasis is placed on improving quality grade, this will positively affect the overall quality and consistency of the beef supply (NCBA, 2013a). Two main components of eating satisfaction common to every segment of the beef market include tenderness and flavor (NCBA, 2012a).

Looking further into the fatty acid profile and intramuscular fat content claims can further validate the potential differences found in Akaushi. Also, analysis of sensory attributes, such as tenderness, juiciness and flavor of the Akaushi can help to verify the breed is additionally a very high quality beef breed.

The Akaushi genetics have the power and capability to conceivably provide a positive change to the foundation genetics of the national cowherd. This would most likely increase the quality and consistency of beef United States producers strive for. This would ultimately create a stronger value for Akaushi influenced cattle for the producers, as other segments will recognize profitable cattle and beef. Akaushi are claimed to increase the quality grade of the half-blood progeny by double (Payne, 2011). The crossbred Akaushi cattle are evaluated less than their fullblood counterparts, and in the United States, the crossbreds are considerably used more in beef production. A deeper look into the Akaushi crossbred cattle could give the association as well as the producer the opportunity to maximize quality and profitability of market cattle.



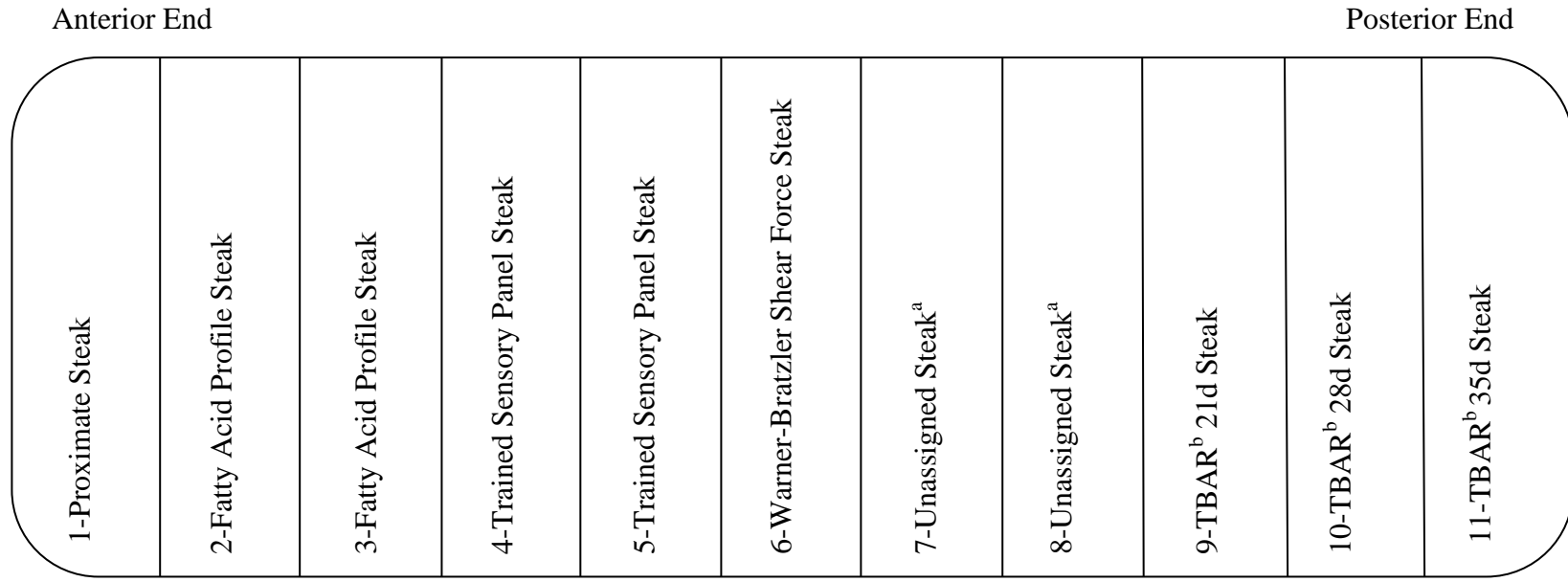
## MATERIALS AND METHODS

### **Collection and Fabrication**

Analyses including human subjects were approved by the Angelo State University Institutional Review Board (BRA-100115). Prior to sensory panels, participants were required to read and sign a consent form; participation was dependent on the participant signing the form. Striploins (NAMP# 180; n=106) were collected from two commercial beef harvesting plants (Hereford, Texas and Friona, Texas). Beef type served as treatment; these included Akaushi top choice (Akaushi) (n=36), top choice branded (TCB) (n=36), and commodity low prime (prime) (n=34). Striploins were selected with the criteria of modest to moderate marbling for the top choice products and slightly abundant marbling for the prime product. Additionally, care was taken to select samples that were devoid of defects. Akaushi product utilized was from the Certified Beef Program G-99 (USDA-AMS, 2015b). Guidelines for TCB were derived from the USDA Certified Beef Program G-1 and the guidelines were: modest or moderate marbling, medium or fine marbling texture, A maturity, 10-16 square inch ribeye, 1050 pound hot carcass weight or less, less than one inch backfat at the 12/13<sup>th</sup> rib interface, superior muscling, practically free of capillary rupture, no dark cutters, and no neck hump exceeding two inches (USDA-AMS, 2015c). Striploins were transported to Angelo State University Food Safety and Product Development Laboratory and maintained below 4°C within refrigeration range, where they were wet aged under vacuum for 21 days from the fabrication date.

At 21 days postmortem, loins were fabricated into 2.54 cm steaks starting from the anterior end. Steaks were serially assigned from the anterior end to proximate analysis, fatty

acid analysis (2 steaks), trained sensory analysis (2 steaks), Warner-Bratzler shear force, unassigned (2 steaks; held in contingency), thiobarbituric acid reactive substances assay (TBAR) 21 day, TBAR 28 day, and TBAR 35 day (Figure 1). The TBAR steaks 21 day, 28 day and 35 day aged are referred to as 21d, 28d, and 35d, respectively. All steaks anterior to the 21d TBAR were vacuum packaged and frozen at  $-12^{\circ}\text{C}$  for subsequent analyses. Remaining portions of strips were repackaged in vacuum bags under full vacuum and refrigeration. Portions were then aged for the appropriate amount of time (28 days and 35 days). After the allotted ageing time, a steak was removed and the remaining portion was repackaged. Sample identity was maintained throughout the entire process.



**Figure 1.** Steak assignment for laboratory analysis of strip loins used in study

<sup>a</sup>Unassigned steaks held in contingency

<sup>b</sup>TBAR = Thiobarbituric Reactive Substance Assay

## **Shelf-Life and Color**

Steaks assigned to TBAR 21d were allowed to bloom for approximately 20 minutes then placed in an overwrap package. Subsequently, steaks were placed on foam trays with an absorbent pad and covered with polyvinylchloride wrap. Objective color readings using a HunterLab MiniScan XE Plus (Hunter Associates Laboratory, Inc. Reston, VA) set to colorimeter color, numeric standard, and D65/10° were taken daily starting on d0 to d4 of display. Numerical readings recorded in the form of L\*, a\*, and b\* were the average of three readings taken in different areas of the longissimus lumborum. Steaks were displayed for four days in an atmosphere consistent with a commercial retail case, and maintained under lighting, using Promolux (Safe Spectrum T8 Platinum, Shawnigan Lake, BC. Canada), with illumination intensity kept at approximately 1900 lux and temperature was maintained below 4°C, within refrigeration range. This procedure was replicated for the 28d and 35d steaks once aged for the respective time durations under vacuum. After four days of display, samples were utilized for TBAR.

## **Thiobarbituric Acid Reactive Substance Assay (TBAR)**

### Reagent Preparation and Standards

The TBAR method utilized for lipid oxidation is a modified version of Buege and Aust (1978) as described by Luque et al. (2011). Trichloroacetic acid/ Thiobarbituric acid (TCA/TBA) stock solution, Butylated Hydroxyl Anisole (BHA) and 3M 1,1,3,3-tetraethoxypropane (TEP; 95% purity; MW = 220.31) (TEP) solutions were prepared one day prior to the initial analysis. After seven days, any remaining reagents were discarded and replaced for subsequent procedures. Prior to analysis each day, a duplicate of (TEP) solution

standard was prepared in order to create a standard curve. Readings were obtained utilizing an Evolution 201 UV spectrophotometer (Thermo Scientific, Waltham, MA) set at a wavelength of 531 nm.

#### TBAR Procedure

The retail like shelf atmosphere utilized in the project included continuous fluorescent lighting and packaging composed of a foam tray, absorbent pad and polyvinyl chloride overwrap. After the four days, external fat, connective tissue, and muscles were removed and the longissimus lumborum was cubed small enough to adequately facilitate powdering. The sample was submerged in liquid nitrogen until sufficiently frozen and then blended to a homogenous powder utilizing a commercially available food blender. Powdered samples were then stored at  $-80^{\circ}\text{C}$  in whirl pack bags until analysis. The remainder of the procedure was completed in duplicate. Duplicate 10 gram powdered samples were homogenized with 30 mLs of cold deionized water utilizing a Power Gen 1000 Homogenizer (Fisher Scientific, Waltham, MA). Homogenate was placed in a 50mL polypropylene conical vial and centrifuged at 1850 xG (3000 rpm) for 10 minutes. Following, 2 mLs of supernatant was removed and placed in a 15 mL polypropylene conical vial containing 4 mLs of TCA/TBA solution. Next, 100  $\mu\text{L}$ s of BHA was added to the vial. The vials were vortexed thoroughly, placed in a hot water bath ( $100^{\circ}\text{C}$ ) for 15 minutes, then submerged in an ice water bath for 10 minutes. Samples were centrifuged at 1850 xG (3000 rpm) for 10 minutes. The supernatant was then analyzed twice in the spectrophotometer utilizing a sipper cell to determine absorbance levels. The averages of the two values were used and were reported in mg of Malondialdehyde per kg muscle.

### **Trained Sensory Panel Analysis**

Steaks were thawed for 10-14 hours at 2-5°C, so as to allow the internal temperature to reach 2-5°C. The steaks were then cooked according to the clam-shell style grill procedure method outlined by Kerth et al. (2003). Raw weight and raw temperature were recorded before cooking. Steaks were threaded with a thermocouple probe to read the geometric center of the steak. Steaks were cooked on a Calphalon® (Southern Pines, NC) set to 350°F with the lid closed until an internal temperature of 71°C was reached. Final internal temperature and post cooking weight of the steaks were recorded. External fat and heavy connective tissue were then removed. The remaining portion of steak was cut into uniform 1cm<sup>3</sup> pieces utilizing the sensory grid. The pieces were immediately placed in sample holding pans containing temperature maintaining sand beneath the sample cups. The trained sensory panel was conducted according to AMSA (1995) procedures. A red-light sensory environment was utilized, and the panelists were instructed to cleanse their pallet before panels and between samples. The pallet cleanse consisted of a rinse of chilled filtered water, as well as two taste bud refreshers, apple juice and unsalted crackers. Each sample was given to every panelist, and values were recorded for initial and sustained juiciness, first impression and overall tenderness, flavor intensity, off flavor and overall acceptability. The mean value of 8-10 panelists was the value used for analysis of each attribute listed for the sensory analysis.

### **Warner-Bratzler Shear Force Analysis**

Steaks were allowed 10-14 hours to thaw at 2-5°C, for the internal temperature to reach 2-5°C. The clam-shell style grill procedure utilized in the sensory portion of the

project was duplicated for the Warner-Bratzler shear force (WBSF) analysis. The raw and cooked temperatures and weights were collected and recorded during the cooking portion of the analysis. After cooking, steaks were placed on a clean tray for chilling. The full trays were overwrapped with polyvinyl chloride (PVC) film and chilled for 24 hours at 2-5°C. After the chill period, the steaks were removed from the cooler, and utilizing a knife, steaks were trimmed to remove all external fat, muscles, and connective tissue. The orientation of muscle fibers in the steak was visually located and steaks were cored parallel to the muscle fibers. Each core was sheared once with a Warner-Bratzler Shear Force machine (Manhattan, KS) and the peak value for each core was recorded in kg. The mean value of six cores for each steak was utilized in the WBSF analysis.

### **Proximate Analysis**

Steaks utilized for the proximate analysis were trimmed of external fat, muscles, and connective tissue. Then, steaks were vacuum packaged and frozen at -12°C. Samples for the proximate analysis were thawed for 12-24 hours at 2-5°C. Steaks were then transported to Texas Tech University Animal and Food Science Laboratory for analysis and maintained at 4°C. The analysis was completed utilizing procedures outlined by Luque et al. (2011). The samples were cubed into pieces and using a domestic electric meat grinder, ground through a ¼ inch plate. The samples were then packed into plates and analyzed using a Foss® Foodscan™ (Eden Prairie, MN), following manufacture's guidelines. Values were reported as a percent of 100.

### **Gas Chromatography (GC) Fatty Acid Analysis**

Initial Preparation

Steaks were vacuum packaged and frozen at  $-80^{\circ}\text{C}$ , until subsequent analysis. The steaks were then thawed at  $2-5^{\circ}\text{C}$  for approximately 12 hours and transported to Texas Tech University Animal and Food Science Laboratory for analysis. The steaks were then cubed, frozen in liquid nitrogen, and powdered utilizing a Robot Coupe® food processor (Ridgeland, MS). The powdered samples were then stored in whirl pack bags at  $-80^{\circ}\text{C}$  prior to lipid extraction.

### Lipid Extraction

Initially, 1 gram ( $\pm 0.2$ ) of sample powder was weighed and placed into 50 mL polypropylene conical vials. The vials were stored at  $-80^{\circ}\text{C}$ . First, 7 mLs of methanol (HPLC grade, Fisher Scientific, Waltham, MA) was added to the tube and the sample was homogenized for 30 seconds using a Polytron PT-2100® (Kinematica AG, Switzerland). Following, 14 mLs of chloroform (HPLC grade, Fisher Scientific, Waltham, MA) was added to the tube and homogenized for another 30 seconds. The sample was then filtered through a Whatman no. 40 (ashless, 110 mm diameter) filter paper into another 50 mL polypropylene conical vial. The tube was then rinsed and homogenized with 12 mLs of a 2:1 (v/v) mixture of chloroform/methanol solution. The rinse solution was then filtered through the same filter paper. Between samples, the polytron was rinsed with a separate 2:1 (v/v) of chloroform/methanol for 5 seconds, then with deionized water for 5 seconds. The probe was then visually checked, wiped and dried with a disposable laboratory tissue. The rinse solutions were changed every four samples. After filtration, 8 mLs of 0.88% KCl (ACS grade, Fisher Scientific, Waltham, MA) was added to the conical vials and placed on a wrist action shaker for 10 minutes. The vials were then centrifuged at room temperature (21.0-



24.0°C) for 5 minutes at 1000 xG. The upper layer was then aspirated into a waste container, and the sample solution was placed into a 15 mL glass test tube with a Teflon-lined screw cap. Samples were reduced under nitrogen (UHP grade). The conical vial was then rinsed with 2 mLs of 2:1 (v/v) chloroform/methanol and the solution was added to the glass test tubes. The solution in the glass test tubes was reduced to approximately 3 mLs and flushed with nitrogen (UHP grade) before storing in -80°C.

### Lipid Separation

Initially, tubes were evaporated to dryness under nitrogen (UHP grade). Following evaporation, 2 mLs of chloroform were added to the test tube, and the tube was vortexed for 5 seconds. The solution was added to a prepared Restek® SPE cartridge (3mL volume, normal phase absorbent). Then, 2 more mLs of chloroform were added to the tube and again vortexed for 5 seconds and added to the cartridge. The vacuum was adjusted to approximately 0.5 mL per second. Lastly, 6 mLs of chloroform was added to the tube, vortexed for 5 seconds and added to the cartridge. Once the cartridge was drained to dryness, the tubes were removed and labeled as the neutral lipid portion. The neutral lipids were then reduced to dryness under nitrogen (UHP grade) for storage at -80°C. For the polar lipid fraction, 15 mLs of methanol was added to the test tube in 5 mL increments, vortexed for 5 seconds, and added to the cartridge. The portion that was collected once the cartridge was drained to dryness was the polar lipid fraction and was then reduced to dryness under nitrogen (UHP grade) for storage at -80°C. Between samples, cartridges were washed with 10 mLs of methanol, and then 10 mLs of chloroform. Cartridges were used for no more than

five samples. Immediately before use each time, cartridges were rinsed with 3 mLs of chloroform.

#### Base-Catalyzed Transesterification of Neutral and Polar Lipids

Method for transesterification was a modification of Christie (1993). First, 1 mL of 0.4 mg/mL internal standard (methyl heneicosanoate, Nu-Chek Prep, Inc., Elysian, MN) in toluene was added to the glass test tube containing the sample. Following, 2 mLs of 0.5M sodium methoxide (ACS grade, Sigma-Aldrich, St. Louis, MO) was added, and the tube was vortexed thoroughly. Polar lipid samples were placed in a hot water shaker bath (50°C at 150 rpm) for 5 minutes and neutral lipid samples were placed in a hot water shaker bath (50°C at 150 rpm) for 10 minutes. Next, 100 µLs of Glacial Acetic Acid was then added to samples, followed by 5 mLs of nano-pure water. Samples were then vortexed thoroughly, and then 5 mLs of hexane (HPLC grade, Fisher Scientific, Waltham, MA) was added. Samples were shaken on a wrist action shaker for 5 minutes, then centrifuged for 10 minutes at 1850 xG (3000 rpm). The top organic layer was removed and placed into a 15mL polypropylene conical vial. Another 5 mLs of hexane was added to the glass test tube and shaken for 5 minutes and then centrifuged at 1850 xG (3000 rpm). A second upper organic layer was removed and added to the 15 mL conical vial. Approximately, 1g of sodium sulfate (ACS grade, Sigma-Aldrich, St. Louis, MO) was added to tubes. The liquid portion was then transferred to a new 15 mL polypropylene conical vial and contents were reduced under nitrogen (UHP grade) to 4 mLs, resulting in a final concentration of 0.1 mg/mL internal standard. Tubes were then stored at -20°C.

## GC Preparation

A 1 mL aliquot of sample was added to gas chromatography (GC) vials (2mL amber autosampler vial). Samples were capped and stored at -20°C until analyzed. The GC system used to analyze fatty acid methyl esters (FAME) was an Agilent Technologies 6890N series (Santa Clara, CA) and equipped with a flame ionization detector and a HP-88 capillary column (100mm x 0.25 mm i.d., 0.20 µm film thickness, Agilent Technologies, Santa Clara, CA). The inlet temperature was set to 260°C with a split ratio of 15:1. Helium (UHP grade) was used as the carrier gas at a flow rate of 1 mL/min. The initial oven temperature was held for 5 minutes at 140°C then increased at a rate of 4°C/minute until a final temperature of 240°C was reached and held for 15 minutes. The detector temperature was 280°C. A standard mixture of 28 fatty acid methyl esters (FAMES) (GLC-462, Nu-Chek Prep, Inc., Elysian, MN) dissolved in hexane at five different concentration levels was used for calibration and to determine linearity and limit of detection (LOD). The LOD was calculated as 3 times the standard deviation of the baseline noise divided by the slope of the calibration curve for each analyte. The limit of quantification (LOQ) was defined as the lowest standard for each analyte (0.02 mg/mL). The calibration curve and internal standard were used to quantify FAMES. The FAMES that were analyzed include C10:0, C12:0, C14:0, C16:0, C18:0, C20:0, C22:0, C24:0, C12:1, C14:1, C16:1, C18:1n7, C18:1n9, C20:1, C22:1, C24:1, C18:2, C18:3n3, C18:3n6, C20:2, C20:3n3, C20:3n6, C20:4, C20:5, C22:2, C22:4, C22:5n3, and C22:6. Saturated fatty acids included C10:0, C12:0, C14:0, C16:0, C18:0, C20:0, C22:0, and C24:0. Monounsaturated fatty acids included C12:1, C14:1, C16:1, C18:1n7, C18:1n9, C20:1, C22:1, and C24:1. Polyunsaturated fatty acids included C18:2, C18:3n3, C18:3n6,

C20:2, C20:3n3, C20:3n6, C20:4, C20:5, C22:2, C22:4, C22:5n3, and C22:6. Values were reported as percent of total fatty acid.

### **Statistical Analysis**

All data was analyzed using the mixed model (PROC MIXED) procedure of SAS (SAS Inst. Inc., Cary, NC). Response variables analyzed in the models included trained sensory panel attributes, cook loss percent, fatty acid profiles, proximate components, lipid stability (TBAR), L\*a\*b\* values and Warner-Bratzler Shear Force values. When significant differences were found, the least squares means were separated utilizing the PDIFF option and Tukey-Kramer adjustment of SAS. Also, DDFM = satterwaithe statement was used to control for degrees of freedom. In the color analysis, steak was used as a repeated measure. All comparisons were analyzed using a predetermined of  $\alpha$  of 0.05. When looking at fatty acid profile composition, ratios of n-6 to n-3, polyunsaturated to saturated, and monounsaturated to saturated were calculated using means generated in SAS.

## RESULTS AND DISCUSSION

### Color

#### L\*

Color was measured objectively by a HunterLab Miniscan XE. Color values were reported as L\*, a\*, and b\*. When evaluating for each of the color attributes (L\*, a\*, and b\*), there was an interaction present between the main effects (beef type treatment, age, display day) ( $P < 0.0001$ ). Means are provided in Table 2. Scale for the color attribute L\* is 0=black and 100=white. When comparing L\* results of the three different beef type treatments (prime, top choice branded beef (TCB), and Akaushi) for the d0 of the 21d ageing period, means for Akaushi and TCB were similar and less than prime ( $P \geq 0.05$ ); moreover, a large numerical range for L\* was present, with treatments producing values of 44.60 (prime), 40.11 (TCB), and 38.75 (Akaushi). According to Holman et al. (2016), consumer acceptability increased linearly with L\* when b\* was constant. When assessing L\* values in the 21d ageing period for d0 and d4, values were similar ( $P \geq 0.05$ ) for each of the respective beef type treatments. Lighter colored, less fat, and fresher steaks were correlated with consumer's perception of better meat quality (Acebron and Dopico, 2000). Therefore, steaks with those attributes maintained greater consumer appeal over the display day timeframe. When comparing the 35d age, d0 steaks to the 21d age, d0 steaks within beef type treatments, TCB was not different ( $P = 0.99$ ), but prime 35d age was darker than 21d age ( $P = 0.01$ ) and Akaushi 35d age was lighter than 21d age ( $P < 0.0001$ ). This is not typically seen in beef color, and more replication of color analysis should be completed to gain confidence in the trend. When looking from a retail standpoint, maintaining color, or even improving

color with increased aging days is positive for the retailer. This would mean that cuts could remain in vacuum bags for a longer period of time without negative monetary effects of possible discounted meat products. Treatments were similar ( $P \geq 0.05$ ) when comparing d0 and d4 within the 35d age and respective beef type treatments. While comparing 21d age and 35d age at d4 and holding treatment constant, steaks were similar ( $P = 1.00$ ) for each. According to Carpenter et al. (2001), color did not affect taste of steaks or patties in consumers.

a\*

Scale for the color attribute a\* ranges from negative values=green to positive values=red. When evaluating a\* values, the day with the greatest range of values was d0 from the 21d age period with Akaushi (29.49) as the most red, followed by prime (27.08), and TCB (21.91); each treatment was different from one another ( $P \leq 0.05$ ). While holding beef type treatment and age treatment (21d) constant and comparing d0 and d4, all three beef types were more green on d4 ( $P < 0.0001$ ). When comparing 21d and 35d aging treatments and looking at d0 for each of the respective treatments, the steaks were more green in color on d0 of the 35d aging period ( $P < 0.0001$ ) for all beef types. When looking at the 35d aging period for each of the respective beef types, steaks on d0 were more red ( $P < 0.0001$ ) than steaks on d4. Furthermore, when comparing 21d and 35d aging periods on d4 for each of the respective treatments, steaks were more green on 35d for each treatment ( $P < 0.0001$ ). Historically, a\* values were utilized as an indicator of consumer acceptance; however, more recently b\* values have been more highly evaluated. Holman et al. (2016) and Morales et al. (2013), found that red color intensity did not have significant effects on consumer opinions.

b\*

Scale for the color attribute b\* is as follows: negative values=blue and positive values=yellow. When comparing values for b\*, the greatest range was in the 21d aging period on d0. Each of the beef type treatments were different from another ( $P \leq 0.05$ ), with Akaushi (24.55) reading the most yellow, followed by prime (21.22), and TCB (19.61). Over the 21d age period from d0 to d4, steaks became more blue in color for every beef type treatment ( $P < 0.0001$ ). While holding day and beef type treatment the same when comparing 21d and 35d aging periods for d0, steaks from the 21d period were more yellow ( $P = 0.0003$ ) for each of the beef type treatments. Over the display period for the 35d aging treatment, from d0 to d4 steaks became more blue in color for each of the beef type treatments ( $P \leq 0.0001$ ). As the aging period increased from 21d to 35d when looking at d4 steaks, values were more yellow with the 21d age treatment than the 35d age treatment for Akaushi and TCB ( $P < 0.0001$ ), and was the same for prime ( $P = 1.00$ ). Holman et al. (2016), suggested a possible acceptable b\* value range as it related to consumer acceptability; however, this acceptable b\* range is also dependent on L\* values. Holman et al. (2016) found unacceptable consumer scores were associated with b\* values of less than 16. All of the b\* values observed in this study fell at or above 16.

**Table 2.** Least squares means ( $\pm$  SEM) for Hunter color values (lightness L\*, redness a\*, yellowness b\*) of Akaushi, top choice branded, and prime steaks over three different ageing periods and a four day display time

Color Attribute	Day	Akaushi			TCB <sup>a</sup>			Prime		
		21d	28d	35d	21d	28d	35d	21d	28d	35d
L*	0	38.75 $\pm$ 0.47 <sup>y</sup>	40.05 $\pm$ 0.47 <sup>y</sup>	40.77 $\pm$ 0.47 <sup>x</sup>	40.11 $\pm$ 0.47 <sup>y</sup>	39.95 $\pm$ 0.47 <sup>y</sup>	40.87 $\pm$ 0.47 <sup>x</sup>	44.60 $\pm$ 0.48 <sup>x</sup>	42.75 $\pm$ 0.48 <sup>x</sup>	42.99 $\pm$ 0.48 <sup>x</sup>
	1	39.81 $\pm$ 0.47 <sup>y</sup>	40.92 $\pm$ 0.47 <sup>y</sup>	40.36 $\pm$ 0.47 <sup>y</sup>	40.79 $\pm$ 0.47 <sup>y</sup>	41.67 $\pm$ 0.47 <sup>xy</sup>	42.38 $\pm$ 0.47 <sup>xy</sup>	45.05 $\pm$ 0.48 <sup>x</sup>	43.78 $\pm$ 0.48 <sup>x</sup>	43.95 $\pm$ 0.48 <sup>x</sup>
	2	40.08 $\pm$ 0.47 <sup>y</sup>	39.52 $\pm$ 0.47 <sup>y</sup>	38.45 $\pm$ 0.47 <sup>y</sup>	40.52 $\pm$ 0.47 <sup>y</sup>	41.26 $\pm$ 0.47 <sup>y</sup>	41.29 $\pm$ 0.47 <sup>x</sup>	43.54 $\pm$ 0.48 <sup>x</sup>	44.13 $\pm$ 0.48 <sup>x</sup>	43.56 $\pm$ 0.48 <sup>x</sup>
	3	39.51 $\pm$ 0.47 <sup>y</sup>	39.52 $\pm$ 0.47 <sup>y</sup>	40.37 $\pm$ 0.47 <sup>x</sup>	38.73 $\pm$ 0.47 <sup>y</sup>	40.42 $\pm$ 0.47 <sup>y</sup>	41.50 $\pm$ 0.47 <sup>x</sup>	44.62 $\pm$ 0.48 <sup>x</sup>	43.35 $\pm$ 0.48 <sup>x</sup>	42.19 $\pm$ 0.48 <sup>x</sup>
	4	39.27 $\pm$ 0.47 <sup>y</sup>	39.14 $\pm$ 0.47 <sup>z</sup>	39.69 $\pm$ 0.47 <sup>y</sup>	40.39 $\pm$ 0.47 <sup>y</sup>	40.75 $\pm$ 0.47 <sup>xy</sup>	40.43 $\pm$ 0.47 <sup>xy</sup>	43.30 $\pm$ 0.48 <sup>x</sup>	43.37 $\pm$ 0.48 <sup>x</sup>	42.69 $\pm$ 0.48 <sup>x</sup>
a*	0	29.49 $\pm$ 0.27 <sup>x</sup>	22.80 $\pm$ 0.27 <sup>x</sup>	21.99 $\pm$ 0.27 <sup>x</sup>	21.91 $\pm$ 0.27 <sup>z</sup>	21.60 $\pm$ 0.27 <sup>x</sup>	19.12 $\pm$ 0.27 <sup>y</sup>	27.08 $\pm$ 0.28 <sup>y</sup>	21.32 $\pm$ 0.28 <sup>x</sup>	20.63 $\pm$ 0.28 <sup>xy</sup>
	1	25.39 $\pm$ 0.27 <sup>x</sup>	22.95 $\pm$ 0.27 <sup>x</sup>	22.14 $\pm$ 0.27 <sup>x</sup>	23.33 $\pm$ 0.27 <sup>y</sup>	22.01 $\pm$ 0.27 <sup>x</sup>	19.68 $\pm$ 0.27 <sup>y</sup>	25.39 $\pm$ 0.28 <sup>x</sup>	21.53 $\pm$ 0.28 <sup>x</sup>	20.92 $\pm$ 0.28 <sup>xy</sup>
	2	22.36 $\pm$ 0.27 <sup>x</sup>	25.95 $\pm$ 0.27 <sup>x</sup>	19.53 $\pm$ 0.27 <sup>x</sup>	21.47 $\pm$ 0.27 <sup>x</sup>	20.56 $\pm$ 0.27 <sup>y</sup>	17.80 $\pm$ 0.27 <sup>y</sup>	21.87 $\pm$ 0.28 <sup>x</sup>	19.90 $\pm$ 0.28 <sup>y</sup>	19.57 $\pm$ 0.28 <sup>x</sup>
	3	20.62 $\pm$ 0.27 <sup>xy</sup>	18.70 $\pm$ 0.27 <sup>y</sup>	16.66 $\pm$ 0.27 <sup>y</sup>	21.11 $\pm$ 0.27 <sup>x</sup>	18.18 $\pm$ 0.27 <sup>y</sup>	16.11 $\pm$ 0.27 <sup>y</sup>	19.30 $\pm$ 0.28 <sup>y</sup>	23.18 $\pm$ 0.28 <sup>x</sup>	18.21 $\pm$ 0.28 <sup>x</sup>
	4	20.62 $\pm$ 0.27 <sup>x</sup>	19.22 $\pm$ 0.27 <sup>x</sup>	16.14 $\pm$ 0.27 <sup>xy</sup>	19.05 $\pm$ 0.27 <sup>y</sup>	17.43 $\pm$ 0.27 <sup>y</sup>	15.30 $\pm$ 0.27 <sup>y</sup>	19.31 $\pm$ 0.28 <sup>xy</sup>	18.69 $\pm$ 0.28 <sup>xy</sup>	17.23 $\pm$ 0.28 <sup>x</sup>
b*	0	24.55 $\pm$ 0.20 <sup>x</sup>	19.21 $\pm$ 0.20 <sup>x</sup>	20.08 $\pm$ 0.20 <sup>x</sup>	19.61 $\pm$ 0.20 <sup>z</sup>	18.85 $\pm$ 0.20 <sup>x</sup>	17.76 $\pm$ 0.20 <sup>y</sup>	21.22 $\pm$ 0.21 <sup>y</sup>	19.95 $\pm$ 0.21 <sup>x</sup>	19.94 $\pm$ 0.21 <sup>x</sup>
	1	22.96 $\pm$ 0.20 <sup>x</sup>	20.75 $\pm$ 0.20 <sup>x</sup>	21.34 $\pm$ 0.20 <sup>x</sup>	19.67 $\pm$ 0.20 <sup>y</sup>	19.23 $\pm$ 0.20 <sup>y</sup>	18.26 $\pm$ 0.20 <sup>z</sup>	20.05 $\pm$ 0.21 <sup>y</sup>	19.23 $\pm$ 0.21 <sup>y</sup>	19.55 $\pm$ 0.21 <sup>y</sup>
	2	19.98 $\pm$ 0.20 <sup>y</sup>	20.25 $\pm$ 0.20 <sup>x</sup>	19.32 $\pm$ 0.20 <sup>x</sup>	19.16 $\pm$ 0.20 <sup>y</sup>	19.44 $\pm$ 0.20 <sup>xy</sup>	17.45 $\pm$ 0.20 <sup>y</sup>	21.17 $\pm$ 0.21 <sup>x</sup>	18.98 $\pm$ 0.21 <sup>y</sup>	19.64 $\pm$ 0.21 <sup>x</sup>
	3	18.52 $\pm$ 0.20 <sup>y</sup>	17.91 $\pm$ 0.20 <sup>xy</sup>	16.98 $\pm$ 0.20 <sup>y</sup>	19.71 $\pm$ 0.20 <sup>x</sup>	16.84 $\pm$ 0.20 <sup>y</sup>	16.23 $\pm$ 0.20 <sup>y</sup>	18.16 $\pm$ 0.21 <sup>y</sup>	18.47 $\pm$ 0.21 <sup>x</sup>	18.63 $\pm$ 0.21 <sup>x</sup>
	4	20.09 $\pm$ 0.20 <sup>x</sup>	19.27 $\pm$ 0.20 <sup>x</sup>	17.71 $\pm$ 0.20 <sup>x</sup>	18.05 $\pm$ 0.20 <sup>y</sup>	17.17 $\pm$ 0.20 <sup>y</sup>	16.21 $\pm$ 0.20 <sup>y</sup>	18.94 $\pm$ 0.21 <sup>y</sup>	19.42 $\pm$ 0.21 <sup>x</sup>	18.62 $\pm$ 0.21 <sup>x</sup>

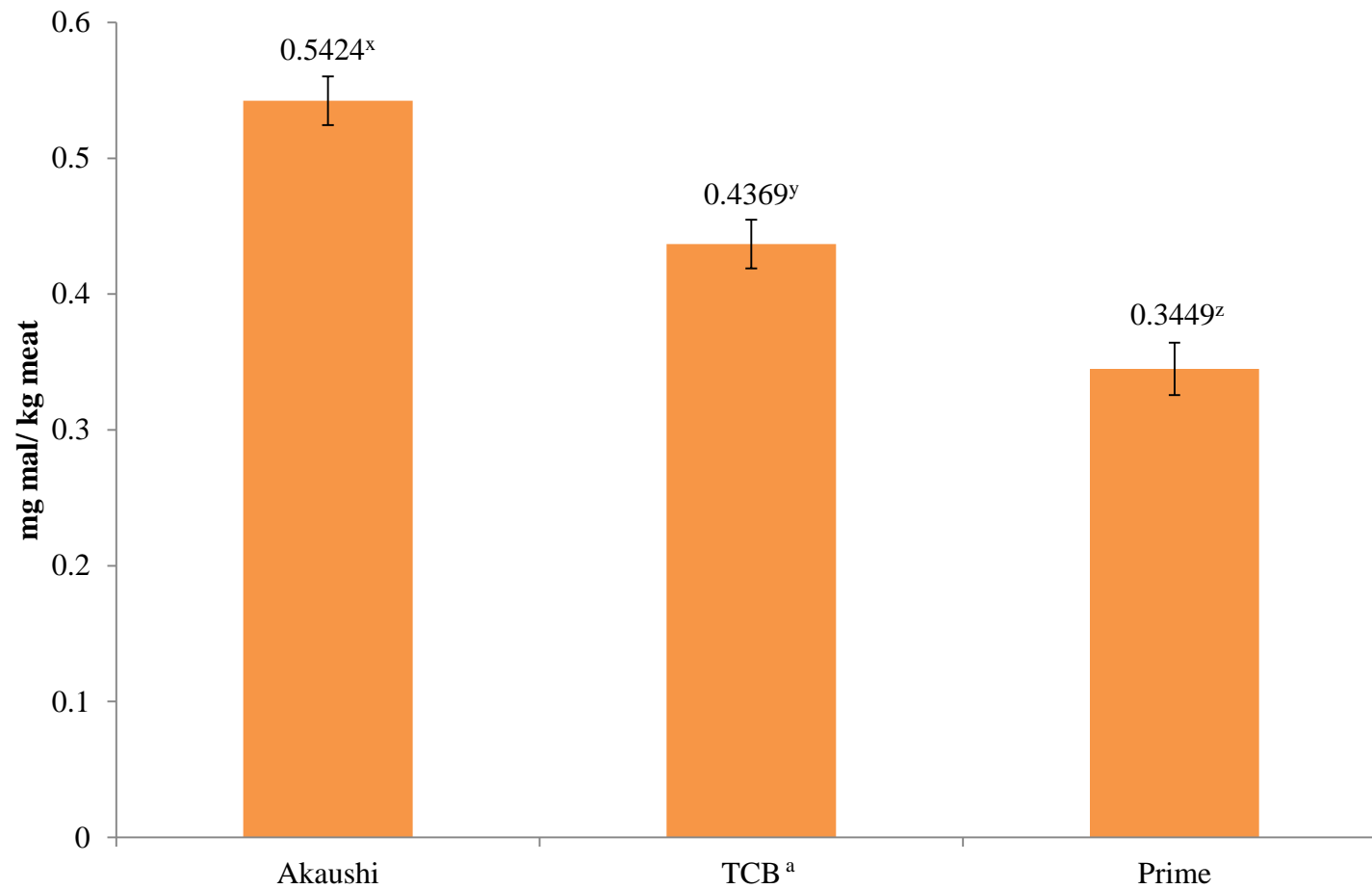
<sup>a</sup>TCB = top choice branded beef

<sup>x,y,z</sup> values within a color attribute, sharing a common day and ageing period, which have different superscripts differ ( $P \leq 0.05$ )



## **Thiobarbituric Reactive Substance Assay**

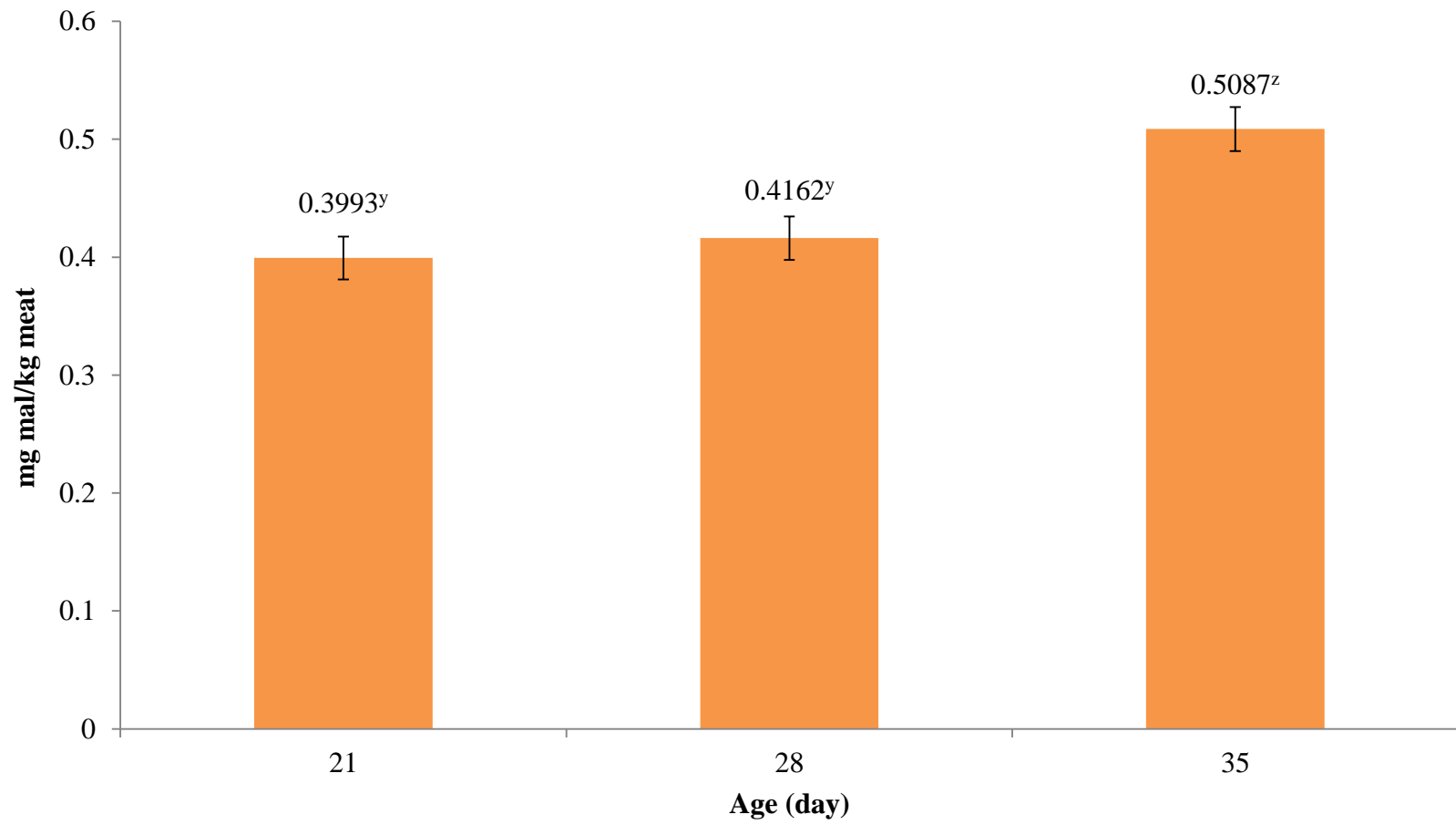
When evaluating oxidative rancidity, Thiobarbituric Reactive Substance Assay (TBAR) was utilized. TBAR measures the secondary oxidation product, aldehydes. TBAR values were recorded in mg of Malondialdehyde per kg muscle. Increased levels of oxidation will decrease shelf-life, as off flavors are produced, and this decreases consumer satisfaction. When evaluating lipid oxidation, there was not an interaction between the main effect of beef type treatment and ageing treatment ( $P = 0.19$ ). The TBAR values for all three beef type treatments were different ( $P < 0.0001$ ), (Figure 2). Akaushi (0.54) had the highest lipid oxidation followed by TCB (0.44), and prime (0.34). Akaushi produced the highest proportion of monounsaturated fatty acids to saturated fatty acids; therefore, this could attribute to the increased level of oxidation. Furthermore, TCB had the greatest proportion of polyunsaturated fatty acids to saturated fatty acids (Table 4.2). As the degree of saturation decreases, the affinity to oxidize increases (Kerth, 2013c); this could be a possible reason as to why there was increased oxidation in Akaushi and TCB. Additionally, there was a difference among aging treatments, ( $P < 0.0001$ ), with 21d (0.40) and 28d (0.42) being similar, while 35d (0.51) was more oxidized (Figure 3). This is comparable to Pouzo et al. (2016), who reported increasing TBAR values for increased aging periods measured after a 5d retail display time. Moreover, Pouzo et al. (2016) stated all three means for 3d, 14d, and 56d were different, but the largest consecutive difference was between 14d to 56d. Campo et al. (2006) suggested the value of 2.28 (mg Malondialdehyde per kg muscle) as a limit for consumer acceptability, and Greene and Cumuze (1981) suggested a detectable level of 0.6 to 2.0. The TBAR values for the three beef type treatments and three aging periods fell below both these suggested thresholds of consumer acceptability.



**Figure 2.** Least Squares Means of Thiobarbituric Reactive Substance Assay Values for Akaushi, top choice branded, and prime steaks  
Error bars represent the standard error of the least squares means

<sup>a</sup>TCB=top choice branded beef

<sup>x,y,z</sup>values within a color attribute which have different superscripts differ ( $P \leq 0.05$ )



**Figure 3.** Least Squares Means of Thiobarbituric Reactive Substance Assay values for 21d, 28d, and 35d ageing period steaks

<sup>a</sup>Error bars represent the standard error of the least squares means

## Trained Sensory Panel

Trained sensory panels evaluated initial juiciness, sustained juiciness, first impression tenderness, overall tenderness, flavor intensity, off flavor, and overall acceptability. The scales utilized in sensory were juiciness: 1- extremely dry, 8- extremely juicy, tenderness: 1- extremely tough, 8- extremely tender, flavor intensity: 1- extremely bland, 8- extremely intense, off flavor: 1- extreme off flavor, 4- none, and overall acceptability: 1- dislike extremely, 8- like extremely. Cook loss was calculated as  $((\text{raw weight}-\text{cooked weight})/\text{raw weight})\times 100$ .

Percentage cook loss was not different between treatments ( $P = 0.51$ ); Table 3 contains the values for cook loss. There was a treatment effect for initial juiciness ( $P = 0.003$ ); Akaushi (6.16), and prime (6.08) were similar to each other, but were juicier than TCB (5.74). Similarly, there was a main treatment effect for sustained juiciness ( $P = 0.0002$ ), and Akaushi (6.03) and prime (6.00) were not different, but juicier than TCB (5.55). First impression tenderness had a main treatment effect ( $P = 0.035$ ), with Akaushi (6.38) more tender than TCB (6.01). Moreover, there was a main treatment effect for overall tenderness ( $P = 0.05$ ); Akaushi (6.19) was more tender than TCB (5.83). There was a main treatment effect for flavor intensity ( $P = 0.007$ ), with Akaushi (6.22) and prime (6.22) measuring more intense than TCB (5.99). There was a main treatment effect for off flavor ( $P = 0.03$ ). Akaushi (3.99) had less of an off flavor compared to TCB (3.96). In the sensory component of overall acceptability, there was a main treatment effect present ( $P = 0.008$ ); Akaushi (6.22) and prime (6.14) were not different, but were greater than TCB (5.79). Results are similar to other studies that found that beef flavor has a greater relationship with

overall acceptability than tenderness or juiciness (Kerth and Miller, 2015; Killinger et al., 2004; Miller et al., 2015).

**Table 3.** Least squares means ( $\pm$  SEM) for trained sensory components and associated cook loss of Akaushi, top choice branded, and prime steaks

Traits	Akaushi	TCB <sup>a</sup>	Prime
	Steak	Steak	Steak
Initial Juiciness <sup>b</sup>	6.16 $\pm$ 0.09 <sup>y</sup>	5.74 $\pm$ 0.09 <sup>z</sup>	6.08 $\pm$ 0.09 <sup>y</sup>
Sustained Juiciness <sup>b</sup>	6.03 $\pm$ 0.09 <sup>y</sup>	5.55 $\pm$ 0.09 <sup>z</sup>	6.00 $\pm$ 0.09 <sup>y</sup>
First Impression Tenderness <sup>c</sup>	6.38 $\pm$ 0.10 <sup>y</sup>	6.01 $\pm$ 0.10 <sup>z</sup>	6.22 $\pm$ 0.10 <sup>yz</sup>
Overall Tenderness <sup>c</sup>	6.19 $\pm$ 0.10 <sup>y</sup>	5.83 $\pm$ 0.10 <sup>z</sup>	6.00 $\pm$ 0.10 <sup>yz</sup>
Flavor Intensity <sup>d</sup>	6.22 $\pm$ 0.06 <sup>y</sup>	5.99 $\pm$ 0.06 <sup>z</sup>	6.22 $\pm$ 0.06 <sup>y</sup>
Off Flavor <sup>e</sup>	3.99 $\pm$ 0.01 <sup>y</sup>	3.96 $\pm$ 0.01 <sup>z</sup>	3.98 $\pm$ 0.01 <sup>yz</sup>
Overall Acceptability <sup>f</sup>	6.22 $\pm$ 0.10 <sup>y</sup>	5.79 $\pm$ 0.10 <sup>z</sup>	6.14 $\pm$ 0.11 <sup>y</sup>
Cook Loss (%) <sup>g</sup>	10.20 $\pm$ 0.60	9.52 $\pm$ 0.60	10.48 $\pm$ 0.61

<sup>a</sup>TCB=top choice branded

<sup>b</sup>Sensory scale for juiciness was 1- extremely dry, 8- extremely juicy

<sup>c</sup>Sensory scale for tenderness was 1- extremely tough, 8- extremely tender

<sup>d</sup>Sensory scale for flavor intensity was 1- extremely bland, 8- extremely intense

<sup>e</sup>Sensory scale for off flavor was 1- extreme off flavor, 4- none

<sup>f</sup>Sensory scale for overall acceptability 1- dislike extremely, 8- like extremely

<sup>g</sup>Cook loss was calculated as ((raw weight-cooked weight)/raw weight)\*100

<sup>y-z</sup>values within a sensory trait which have different superscripts differ ( $P \leq 0.05$ )

## Fatty Acid Profile

Each of the three main beef type treatments were analyzed for fatty acid profile composition, and results were reported as a percentage of total fatty acid. Fatty acid percentages can be found in Table 4.1 and Table 4.2. When evaluating the profiles for composite saturated fatty acids, TCB (47.358) had the highest percent, followed by prime (46.590) and Akaushi (42.818) ( $P < 0.0001$ ). Again, when assessing composite monounsaturated fatty acid percentages, Akaushi (53.835) was the highest, proceeded by prime (49.727), and TCB (48.275) ( $P < 0.0001$ ). TCB (4.413) had the highest percent of composite polyunsaturated fatty acids followed by prime (3.720) and Akaushi (3.391) ( $P < 0.0001$ ) (Table 4.2). The 2015-2020 Dietary Guidelines for Americans recommends no more than ten percent of calories to come from saturated fatty acids (U.S. Department of Health and Human Services and U.S. Department of Agriculture, 2015). When looking at stearic acid, TCB (14.8676) had the highest percent, followed by prime (13.9364), then Akaushi (10.4761) ( $P < 0.0001$ ). When analyzing arachidic acid, TCB (0.1162) had the highest percent, followed by Akaushi (0.0913) and prime (0.0820) ( $P < 0.0001$ ). When analyzing C14:1 (myristoleic acid), there was a main treatment effect ( $P = 0.0006$ ); Akaushi (1.2144) had a higher percent than prime (1.0536) and TCB (0.9602). Evaluation of palmitoleic acid revealed a treatment effect ( $P < 0.0001$ ); Akaushi (5.5259) had a higher percent than both prime (4.2810) and TCB (4.0204). Moreover, when considering differences for vaccenic acid there was a main treatment effect; ( $P < 0.0001$ ); Akaushi (1.7648) had a higher percent than both prime (1.2400) and TCB (1.0780). There was a main treatment effect ( $P < 0.0001$ ) for oleic acid; Akaushi (44.6567) contained the highest percentage, followed by prime (42.4230) and TCB (41.6087). When evaluating percent nervonic acid, Akaushi (0.1045) and prime

(0.0956) were not different, but higher than TCB (0.0527) ( $P = 0.0016$ ). Linoleic acid is an essential fatty acid (EFA) to the human diet (Oregon State University, 2016). Humans cannot synthesize essential fatty acids, so the EFAs need to be consumed in the diet (Oregon State University, 2016). The human body can synthesize other long chain PUFAs into the EFAs; however, this is an inefficient method to obtain the needed EFAs (Oregon State University, 2016). Regarding percent linoleic acid there was a main effect ( $P < 0.0001$ ), with TCB (3.2743) higher than prime (2.9444) and then Akaushi (2.5565). The dietary intake recommended for female and male adults ranges from 11 to 16 grams per day (Oregon State University, 2016). When looking at C20:2, there was a main treatment effect ( $P = 0.0113$ ), prime (0.0763) had a higher percentage than Akaushi (0.0494) and TCB (0.0494). When analyzing C20:3n3, TCB (0.3413) had a higher percent than prime (0.1700) ( $P = 0.0007$ ). When looking at C20:4, there was a beef type treatment difference; TCB (0.1063) had a higher percentage than Akaushi (0.0363) ( $P = 0.0212$ ). Regarding C22:5n3, TCB (0.0895) had a higher percentage than Akaushi (0.0274) ( $P = 0.0001$ ). There was no treatment effect when looking at percentages of C10:0, C12:0, C14:0, C16:0, C22:0, C24:0, C12:1, C20:1, C22:1, C18:3n6, C20:3n6, C20:5, and C22:2 ( $P = 0.3604, 0.0758, 0.2370, 0.8085, 0.1594, 0.1453, 0.3502, 0.0655, 0.8520, 0.0624, 0.0877, 0.1453, \text{ and } 0.3559$ , respectively). When analyzing C22:4, there was a treatment difference ( $P = 0.0530$ ); however there was not a difference using Tukey-Kramer to separate least squares means. Additionally, there was not a difference between treatments ( $P = 0.0950$ ) for  $\alpha$ -linolenic acid, another EFA for the human body. The dietary recommendation for  $\alpha$ -linolenic acid for adult men and women ranges from 1.1 to 1.6 grams per day (Oregon State University, 2016). When analyzing fatty acids, C22:6 was not detected for any of the three treatments. TCB (0.094) had a higher ratio of



PUFA to SFA, than prime (0.080) and Akaushi (0.079) ( $P < 0.0001$ ). When evaluating the ratio of MUFA to SFA, Akaushi (1.262) had a higher ratio than prime (1.073) and TCB (1.025) ( $P < 0.0001$ ). A common ratio of MUFA:SFA for beef cattle is 1:1; however, the Japanese Black cattle raised using traditional practices in Japan achieve ratios of 2.6:1 (Jensen, et al., 2004b). The Akaushi achieved a 1.26:1 ratio, which is much closer to conventional beef; however, the Japanese Black is a different breed and Japan utilizes different production parameters compared to the United States.

A large surge of increasingly health-conscious consumers in today's food market was recognized by Overstreet (2015). Consumers are turning to healthier foods, which includes foods that contain healthier fat. When fats are more beneficial to the human body, the proportion of low density lipoprotein (LDL) to high density lipoprotein (HDL) is reduced, without reducing the number of high density lipoprotein (HDL) (Gilmore et al., 2013). Beef with increased proportions of oleic fatty acid, conjugated linoleic acid and monounsaturated fat have shown to be beneficial to human health (Hawkes, 2013; Gotoh et al. 2014; American Akaushi Association, 2014). These fatty acid features have reduced low density lipoprotein in studies, which can pose problems to heart health (Gilmore et al., 2013). Akaushi did have the highest proportion of MUFA:SFA, which could have positive health implications. The code of federal regulations requires a product to meet 20% or more of an individual's recommended daily intake (RDI) to be labeled as "high, rich in, or excellent source of" (Nutrient Content Claims-General Principles, 2016). Additionally, it is required that a product meet 10-19% of an individual's RDI to be labeled a "good source of, contains or provides" (Nutrient Content Claims-General Principles, 2016). The prime beef type treatment was utilized as an example for the nutritional evaluation, as it contained the greatest

concentration of linoleic fatty acid (as it had the highest percent fat). Furthermore, fatty acid percentages and proximate values were used to calculate nutritional values. If a steak produced 2.94 percent of linoleic acid per total fatty acid and was 12.19 percent intramuscular fat, it would only offer 8.3 percent of RDI (for a 11 g per day requirement) for linoleic acid when consuming a 255 g ( $\approx$  9 oz) steak; therefore, steaks in the current study would not be allowed any special labeling for essential fatty acid content. Moreover, the 2015-2020 Dietary Guidelines for Americans recommends 2-7 oz per day of protein depending on caloric intake (U.S. Department of Health and Human Services and U.S. Department of Agriculture, 2015).

**Table 4.1** Least squares means ( $\pm$  SEM) for fatty acid profile components for Akaushi, top choice branded, and prime steaks

Fatty Acids <sup>b</sup>	Chemical Name	Akaushi		TCB <sup>a</sup>		Prime		
		Total	SE	Total	SE	Total	SE	
Saturated Fatty Acids	Capric Acid	C10:0	0.0820	0.007	0.0867	0.007	0.0731	0.007
	Lauric Acid	C12:0	0.0887	0.005	0.0981	0.005	0.0803	0.006
	Myristic Acid	C14:0	3.6442	0.092	3.5743	0.092	3.7950	0.094
	Palmitic Acid	C16:0	28.3674	0.249	28.5348	0.249	28.5903	0.256
	Stearic Acid	C18:0	10.4761 <sup>z</sup>	0.213	14.8676 <sup>x</sup>	0.213	13.9364 <sup>y</sup>	0.219
	Arachidic Acid	C20:0	0.0913 <sup>z</sup>	0.004	0.1162 <sup>y</sup>	0.004	0.0820 <sup>z</sup>	0.004
	Lignoceric Acid	C22:0	0.0457	0.011	0.0488	0.011	0.0218	0.011
Monounsaturated Fatty Acids		C24:0	0.0027	0.004	0.0100	0.004	2.94E-18	0.004
	Lauroleic acid	C12:1	0.0000	0.0009	0.0000	0.0009	0.0016	0.0009
	Myristoleic Acid	C14:1	1.2144 <sup>y</sup>	0.046	0.9602 <sup>z</sup>	0.046	1.0536 <sup>z</sup>	0.047
	Palmitoleic Acid	C16:1	5.5259 <sup>y</sup>	0.112	4.0204 <sup>z</sup>	0.112	4.2810 <sup>z</sup>	0.115
	Vaccenic Acid	C18:1n7	1.7648 <sup>y</sup>	0.070	1.0780 <sup>z</sup>	0.070	1.2400 <sup>z</sup>	0.072
	Oleic Acid	C18:1n9	44.6567 <sup>y</sup>	0.394	41.6087 <sup>z</sup>	0.394	42.4230 <sup>z</sup>	0.405
		C20:1	0.2408	0.033	0.2182	0.033	0.3256	0.034
Polyunsaturated Fatty Acids		C22:1	0.3179	0.044	0.3340	0.044	0.2982	0.045
		C24:1	0.1045 <sup>y</sup>	0.011	0.0527 <sup>z</sup>	0.011	0.0956 <sup>y</sup>	0.011
	Linoleic Acid	C18:2	2.5565 <sup>z</sup>	0.073	3.2743 <sup>x</sup>	0.073	2.9444 <sup>y</sup>	0.075
	$\alpha$ -linolenic Acid	C18:3n3	0.2755	0.030	0.3473	0.030	0.2602	0.031
	$\gamma$ -linolenic Acid	C18:3n6	0.0400	0.007	0.0159	0.007	0.02111	0.008
		C20:2	0.0494 <sup>z</sup>	0.007	0.0463 <sup>z</sup>	0.007	0.0763 <sup>y</sup>	0.008
		C20:3n3	0.2453 <sup>yz</sup>	0.030	0.3413 <sup>y</sup>	0.030	0.1700 <sup>z</sup>	0.031
	Dihomo- $\gamma$ -linolenic	C20:3n6	0.1343	0.014	0.1545	0.014	0.1108	0.014
	Arachidonic Acid	C20:4	0.0363 <sup>z</sup>	0.018	0.1063 <sup>y</sup>	0.018	0.0613 <sup>yz</sup>	0.018
	Timnodonic Acid	C20:5	0.0027	0.004	0.0100	0.004	2.94E-18	0.004
	C22:2	0.0027	0.003	0.0059	0.003	1.45E-18	0.003	
	C22:4	0.0068	0.002	5.42E-20	0.002	4.58E-19	0.002	
	C22:5n3	0.0274 <sup>z</sup>	0.010	0.0895 <sup>y</sup>	0.010	0.0586 <sup>yz</sup>	0.010	
	C22:6	ND <sup>c</sup>		ND <sup>c</sup>		ND <sup>c</sup>		

<sup>a</sup>TCB = top choice branded beef<sup>b</sup>Values are reported as a percentage of total fatty acid<sup>c</sup>ND = not detected by gas chromatograph<sup>xyz</sup>values within a fatty acid type which have different superscripts differ ( $P \leq 0.05$ )

**Table 4.2** Least squares means ( $\pm$  SEM) for fatty acid profile components for Akaushi, top choice branded, and prime steaks

Fatty Acids <sup>a</sup>	Akaushi	TCB <sup>b</sup>	Prime
SFA <sup>c</sup>	42.818 $\pm$ 0.385 <sup>z</sup>	47.358 $\pm$ 0.385 <sup>y</sup>	46.590 $\pm$ 0.397 <sup>y</sup>
MUFA <sup>d</sup>	53.835 $\pm$ 0.385 <sup>x</sup>	48.275 $\pm$ 0.385 <sup>z</sup>	49.727 $\pm$ 0.397 <sup>y</sup>
PUFA <sup>e</sup>	3.391 $\pm$ 0.094 <sup>z</sup>	4.413 $\pm$ 0.094 <sup>x</sup>	3.720 $\pm$ 0.096 <sup>y</sup>
M:S <sup>f</sup>	1.262 $\pm$ 0.018 <sup>y</sup>	1.025 $\pm$ 0.018 <sup>z</sup>	1.073 $\pm$ 0.018 <sup>z</sup>
P:S <sup>g</sup>	0.079 $\pm$ 0.002 <sup>z</sup>	0.094 $\pm$ 0.002 <sup>y</sup>	0.080 $\pm$ 0.002 <sup>z</sup>
n-6/n-3 <sup>h</sup>	0.475 $\pm$ 0.050 <sup>y</sup>	0.264 $\pm$ 0.050 <sup>z</sup>	0.295 $\pm$ 0.052 <sup>z</sup>

<sup>a</sup>Values are reported as a percentage of total fatty acid

<sup>b</sup>TCB = top choice branded beef

<sup>c</sup>SFA = C10:0, C12:0, C14:0, C16:0, C18:0, C20:0, C22:0, C24:0

<sup>d</sup>MUFA = C12:1, C14:1, C16:1, C18:1n7, C18:1n9, C20:1, C22:1, C24:1

<sup>e</sup>PUFA = C18:2, C18:3n3, C18:3n6, C20:2, C20:3n3, C20:3n6, C20:4, C20:5, C22:2, C22:4, C22:5n3, C22:6

<sup>f</sup>M:S = composite monounsaturated/composite saturated

<sup>g</sup>P:S = composite polyunsaturated/composite saturated

<sup>h</sup>n-6/n-3 = C18:3n6, C20:3n6/ C18:3n3, C20:3n3, C22:5n3

<sup>xyz</sup>values within a fatty acid which have different superscripts differ ( $P \leq 0.05$ )

## **Warner-Bratzler Shear Force**

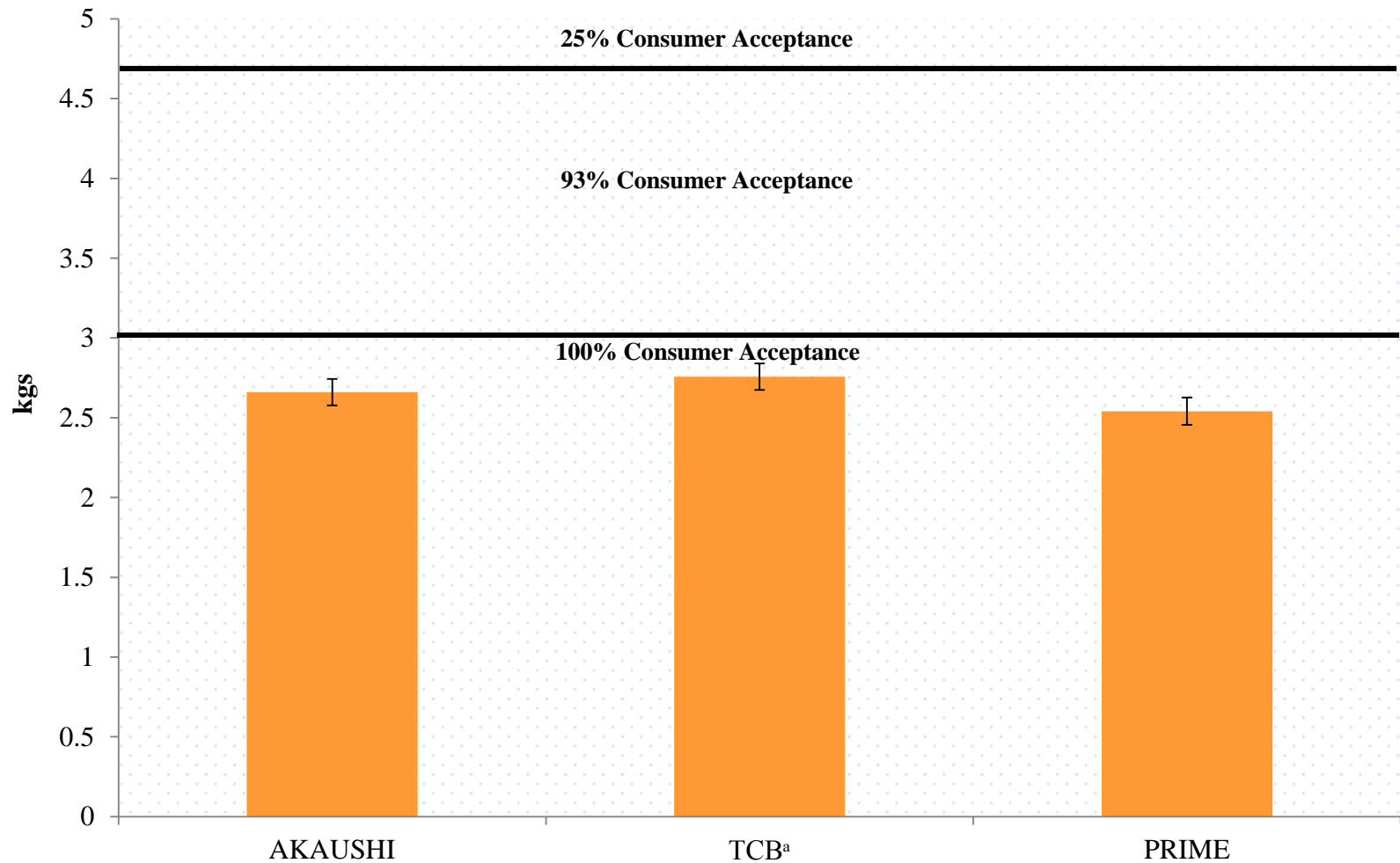
Warner-Bratzler Shear Force is an instrumental method used to analyze tenderness. It is thought that this method of measuring tenderness would be more objective, compared to human sensory panels. Cook loss was not different for the treatments ( $P = 0.34$ ). Additionally, average WBSF values were similar ( $P = 0.20$ ) for all of the treatments. Shear force values for the beef type treatments were as follows 2.76, 2.66, and 2.54 for TCB, Akaushi, and prime, respectively. Average Warner-Bratzler Shear Force (WBSF) values and cook loss averages are found in Table 5. According to Miller et al. (2001), all main treatment averages would fall into the category of 100% consumer acceptability, as values less than 3.0 kg were produced or categorized as very tender. Miller et al. (2001) suggested threshold levels of  $< 3.0$ ,  $3.0-4.6$ , and  $>4.6$  for 100%, 93%, and 25% consumer acceptance, respectively. Values from the present study can be seen in Figure 4 as compared to threshold values found in Miller et al. (2001). Additionally, Belew et al. (2003) suggested the scale of very tender- $< 3.2$  kg, tender- $3.2$  to  $3.9$  kg, intermediate- $3.9$  to  $4.6$  kg and tough-  $> 4.6$  kg. The mean shear force value for strip steak was reported as 3.40 kg, which was higher than what was observed in the present study; however, it should be noted that Belew et al. (2003) utilized USDA Choice and Select carcasses. Furthermore, Guelker et al. (2013) reported a mean value of 23.3N or 2.38 kg for top loin steaks in the 2012 National Beef Tenderness Survey. Lastly, Igo et al. (2015) identified tenderness values of 24.1N or 2.46 kg for choice steaks and 18.8N or 1.92 kg for prime steaks when steaks were grouped by quality grade. This is similar to the Akaushi and TCB values in the present study; however, prime was higher in this study.

**Table 5.** Least squares means ( $\pm$  SEM) for Warner-Bratzler Shear Force values and associated cook loss values for Akaushi, top choice branded, and prime steaks

	Akaushi		TCB <sup>a</sup>		Prime	
	Steak	SE	Steak	SE	Steak	SE
Shear Force (kg) <sup>b</sup>	2.66	0.08	2.76	0.08	2.54	0.09
Cook Loss (%) <sup>b</sup>	12.01	0.48	11.40	0.48	11.01	0.50

<sup>a</sup>TCB=top choice branded beef

<sup>b</sup>Values for Shear Force and Cook Loss did no differ ( $P > 0.05$ )



**Figure 4.** Least Squares Means ( $\pm$  SEM) of Warner-Bratzler Shear Force of Akaushi, top choice branded, and prime steaks

<sup>a</sup>TCB=top choice branded beef

<sup>b</sup>Line at 3.0 represent 100% consumer satisfaction according to Miller et al. (2001)

<sup>c</sup>Line at 4.6 represents 25% consumer satisfaction according to Miller et al. (2001)

<sup>d</sup>The area between the two lines represents 93% consumer satisfaction according to Miller et al. (2001)

## Proximate

Attributes measured in the proximate analysis included percent protein, moisture, fat, and collagen (Table 6). Proximate values were measured utilizing the Foss® Foodscan™ (Eden Prairie, MN), and reported as a percent of the whole. Akaushi (21.78) and prime (21.72) contained lower levels of protein than TCB (22.59) ( $P < 0.0001$ ). Moisture was different for all three treatments ( $P < 0.0001$ ); TCB (67.87) had the highest moisture content, followed by Akaushi (67.12), and prime (64.93). Fat levels were also different across the three treatments ( $P < 0.0001$ ), with prime (12.19) having the highest level, followed by Akaushi (9.00), and TCB (7.81). Similarly, collagen was different ( $P < 0.0001$ ) for all treatments, with prime (2.32) having the highest level, followed by Akaushi (1.97), and TCB (1.81). Results for protein, moisture and fat are comparable with O'Quinn et al. (2012) and Corbin et al. (2015) who reported similar protein levels for USDA Prime and USDA High Choice. Both Akaushi and TCB utilized were upper 2/3 Choice, which resulted in slightly lower means than reported High (upper 1/3) Choice values for O'Quinn et al. (2012) and Corbin et al. (2015). Corbin et al. (2015) also identified values for upper 2/3 Choice Holstein, which were similar to values of Akaushi and TCB, found in the present study. Proximate values obtained fell within expected ranges of the analysis, especially given the results of other associated analyses including perceived tenderness and overall acceptability.



**Table 6.** Least squares means ( $\pm$  SEM) for proximate component values for Akaushi, top choice branded, and prime steaks

Component	Akaushi	TCB <sup>a</sup>	Prime
Protein <sup>b</sup>	21.78 $\pm$ 0.09 <sup>z</sup>	22.59 $\pm$ 0.09 <sup>y</sup>	21.72 $\pm$ 0.09 <sup>z</sup>
Moisture <sup>b</sup>	67.12 $\pm$ 0.21 <sup>y</sup>	67.87 $\pm$ 0.21 <sup>x</sup>	64.93 $\pm$ 0.22 <sup>z</sup>
Fat <sup>b</sup>	9.00 $\pm$ 0.28 <sup>y</sup>	7.81 $\pm$ 0.28 <sup>z</sup>	12.19 $\pm$ 0.29 <sup>x</sup>
Collagen <sup>b</sup>	1.97 $\pm$ 0.03 <sup>y</sup>	1.81 $\pm$ 0.03 <sup>z</sup>	2.32 $\pm$ 0.03 <sup>x</sup>

<sup>a</sup>TCB=Top choice branded beef

<sup>b</sup>Values are reported as a percentage of the whole

<sup>x,y,z</sup>values within a proximate component which have different superscripts differ ( $P \leq 0.05$ )

## CONCLUSION

Akaushi, a Japanese breed of cattle, are marketed for their marbling ability and less saturated fat than other beef breeds. In the sensory portion of the present study, Akaushi and prime were similar for many of the attributes, and more favorable than TCB. Moreover, within the USDA Quality Grade of upper 2/3 Choice, the Akaushi appeared to hold some advantages in comparison to TCB in the organoleptic properties of juiciness, flavor, and overall acceptability, as Akaushi was more similar to prime. However, all of the treatments fell within the acceptable range of consumer satisfaction. The proximate component of the study fell within expected ranges, as related to averages of USDA Quality Grades for the beef type treatments. The fatty acid profile element was diverse, as TCB had the highest polyunsaturated:saturated ratio, and Akaushi had the most monounsaturated:saturated ratio. This seemed to carry over to the shelf stability portion, as prime was the most shelf stable, followed by TCB and then Akaushi. This has an implication for retail merchandizers, in that profiles containing higher levels of unsaturated fatty acid, a shelf-life disadvantage seems to exist. Additionally, the 35d aged product produced the higher oxidation levels than the 28d and 21d. Therefore, if expecting a retail shelf display life of four days, increased aging could result in discounted prices due to the effect of negative aromas, appearances, or the production of off flavors when cooked. There was an interaction in each of the color constituents L\*, a\*, and b\*, when evaluating the impact of beef type, ageing period, and display day. This indicates that the impact of one variable is influenced by another variable; therefore the various combinations of desired age, display day, and beef type treatment, will impact color differently. Lastly, when evaluating Warner-Bratzler Shear Force, values for

each of the treatments fell into the “very tender” category (Belew et al., 2003). Consumer acceptance of tenderness would be expected to be very high for steaks in the “very tender” category.

### Industry Avenues

The Akaushi breed could make an impact on the United States beef industry in several different ways. The breed brings different dynamics to the table, that could help the beef industry; however, this is not certain as many different breeds and genetics of livestock have fallen victim to varying aspects the industry.

The first possible impact could be integrating the genetics into the national cowherd. An example of this class of influence can be seen through the historical instance of the first Certified Beef Program, or better known as Certified Angus Beef®. The breed association organized a premier boxed beef program that in turn increased the demand for black cattle (Angus influenced) throughout the United States (Zimmerman and Schroeder, 2011). This has made a very large impact on the cattle bred, marketed, and harvested in the U.S.

A downfall that could arise in the situation would be that unlike the Angus product, a risk of decreasing profit is seen when the quantity of the product is increased; therefore, increasing the demand of the product is key to sustaining prices as the quantity would increase. Additionally, this type of a situation would be joined with the fact that the genetics would be watered down, and integrity would not be maintained. This leads into the second avenue that could be taken, a niche market. This would create the best opportunity for the owners of the cattle to make the greatest profit, as compared to other systems, by isolating the genetics to control quantity and who can generate profit. Unlike the initial system, in this

niche market only a few people would benefit from the breed both from the production side of the spectrum as well as the consumer side of the spectrum.

Each of the systems that the breed could take are unique to their strengths and weaknesses, and they would need to be addressed and applied to the situation to choose the best situation.

The Akaushi seem to hold some quality advantages, in addition to a higher ratio of monounsaturated:saturated fatty acids; however, these do come with a shelf-life stability disadvantage. Infusing these breed genetics into the current cattle population in the United States could have a positive effect on the quality of beef produced; however, this would only be possible if it is economically competitive with the industry. Additionally, further research should be done to discover the impact of the breed genetics with different breed combinations and ratios. Furthermore, for production and economic purposes, it would be very beneficial to further research Akaushi breed implications to the live phases of beef production.

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