

BEEF FLAVOR MYOLOGY

A Thesis

by

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ABSTRACT

Beef flavor is very complex and the most important driver for consumer acceptance. Cooking method, Quality grade (marbling level), and cooked internal temperature may affect beef flavor. In this study, 54 treatments were utilized, including three beef cuts (outside skirt, inside skirt, and flaps), two Quality grades (USDA Choice and Select), three cooking methods (pan fry, pan grill, and outside grill), and three internal cook temperature endpoints (58°C, 70°C, and 80°C) to better understand trained descriptive beef flavor and texture attributes, volatile flavor aroma compounds, and Warner-Bratzler shear force (WBSF) tenderness. Meat sources were purchased as subprimals in six reps for each cut and were fabricated into 10.16 cm wide steaks.

Generally across all three cuts, Quality grade significantly affected fat-like, cardboardy, juiciness, muscle fiber tenderness beef flavor and texture attributes ($P < 0.05$). Internal cook temperature endpoint significantly affected beef identity, brown, bloody/serummy, metallic, burnt, smokey charcoal, and juiciness ($P < 0.05$). Finally, the attributes generally affected by cooking method included beef identity, brown, metallic, smokey charcoal ($P < 0.05$).

Most flap treatments were clustered near butanoic acid, benzeneacetaldehyde, phenyl acetaldehyde, and 2,6-dimethyl-pyrazine – which are generally sweet, rancid, floral aromas. Generally, inside skirt treatments were related to acetic acid, sulfur dioxide, methyl-benzene, and 1-heptanol – which are sour, sulfur, and fruity aromas. Outside skirts are clustered around aromas such as dl-limonene, 2-acetyl-2-thiazoline, carbon disulfide, and undecanal –

citrus, soapy, buttery aromas.

Treatments had a significant effect on flap tenderness measured by WBSF – Choice steaks were more tender ($P < 0.05$) than Select flaps, those cooked to 80°C were less tender ($P < 0.05$) than other internal temperatures, and pan-grilled flaps were more tender ($P < 0.05$) than the other cooking methods. For inside skirts, there was no effect ($P > 0.05$) by Quality grade on tenderness measured by Warner-Bratzler shear force. However, inside skirts cooked to 58°C were more tender than other internal temperature endpoints, and pan-grilled skirts were more tender than other cooking methods ($P < 0.05$). Lastly, for WBSF measurements, treatments had no impact ($P > 0.05$) on outside skirt steaks.

DEDICATION

My thesis and graduate degree are dedicated to my daughter, Emma. You have become my greatest inspiration and treasure.

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Contributors

This work was supervised by a thesis committee consisting of Dr. Rhonda Miller [advisor] and Dr. Chris Kerth of the Department of Animal Science and Dr. Christine Alvarado of the Department of Poultry Science.

The data analyzed for Chapter III was provided by Dr. Rhonda Miller.

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NOMENCLATURE

cm	centimeter
GC/MS/O	gas chromatograph/mass spectrophotometer system with olfactory
h	hour/s
IMPS	Institutional Meat Purchase Specifications
kgf	kilogram-force
mm	millimeter
SPME	Solid-Phase Micro-Extraction
USDA	United States Department of Agriculture
WBSF	Warner-Bratzler shear force

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CHAPTER I

INTRODUCTION

Cooking method, marbling level, and cooked internal temperature endpoint affect beef flavor and beef flavor have been shown to be the most important driver of consumer acceptance (Adhikari et al., 2011; Robbins et al., 2003). However, beef cuts respond differently to cooking method and cooked internal temperature endpoint based on their inherent chemical characteristics (Mcbee and Wiles, 1967). Beef cuts differ in chemical characteristics based on muscle function in the live animal (Mcbee and Wiles, 1967). Extensive work was conducted through the Beef Checkoff to understand chemical and tenderness characteristics of beef cuts and is available as the Beef Myology website (<https://bovine.unl.edu/>). This website is used by university, industry, and government entities to understand inherent characteristics of individual beef cuts and how to maximize their value as a protein source. However, an understanding of how to maximize flavor of individual cuts, the influence of cooking method, marbling level and cooked internal temperature endpoint across beef cuts has not been fully characterized. Recent Beef Checkoff-funded research has examined relationships between different cooking methods, degree of doneness, cuts, and marbling scores on consumer, trained sensory descriptive sensory flavor, and aromatic volatile chemicals. From 2013 to 2015, consumers in Pennsylvania, Oregon, Kansas and Georgia were recruited. Consumer evaluations were conducted in 2013 as a Central Location Test where beef from 20 different treatments were presented to 240 consumers. Consumers rated flavor and overall liking using 9-point hedonic

scales for beef that varied extensively in cook method, marbling level and internal cook temperature endpoint. The same samples were evaluated by a trained descriptive attribute sensory panel using methods defined by AMSA (2015) and Warner-Bratzler shear force was determined. A second study conducted similarly used beef top loin and beef bottom round roasts cooked to two internal cook temperature endpoints. Additional data for foodservice cooking methods was conducted in the 1990s and while consumer data were not available, trained descriptive attribute flavor data similar to the Beef Lexicon was collected. These data sets provided a base for understanding the effect of cooking method, marbling level and internal cook temperature endpoint on beef flavor across cuts. However, new cooking methods have been developed that provide differences in heat transfer. With increases in technology in cooking devices, the foodservice industry has the ability to prepare and hold beef items differently. Specifically, combination ovens equipped with advanced computer systems allow a chef or cook to prepare a single beef item using multiple cookery methods in a cycle-like application. For example, a chef can grill/sear, braise, and hold or temper a steak in a single device and cooking cycle. This type of technology is currently being utilized to create various flavor profiles and eating experiences in foodservice. Additionally, beef fabrication procedures have changed so that individual muscles, especially from the round and chuck, are merchandized. There is a need to centralize information in a user friendly manner for each beef cut, how flavor is impacted by cooking method, how marbling level impacts flavor, and how internal cooked temperature endpoint affects beef flavor. However, there is a need to expand the information to include new cuts and new cooking methods. This project would incorporate existing data and generate new data to be used in the development of a Beef Flavor Myology tool for use by university, industry and government.

CHAPTER II

LITERATURE REVIEW

Biological Response to Flavor

Flavor has been defined as the sum of perceptions resulting from stimulation of the senses that are grouped together at the entrance of the alimentary and respiratory tracts (Meilgaard et al., 2007). Flavor of food is complex, multi-dimensional and more than the taste perceived on the tongue. The perception of flavor is comprised of the aroma detected by the olfactory bulb, the chemical feeling sensations, the taste perceived by the tongue, and an interaction of these sensations (Meilgaard et al., 2007; Laird, 2015).

Behavioral and perceptual responses to food depend on a convergence between gustatory, olfactory, and visual information (Rolls and Baylis, 1994). Taste is commonly confused with flavor. Flavor is the combined sensory experience of olfaction and gustation. Gustatory signals originate in sensory organs in the oral cavity (taste buds) and are triggered by water-soluble compounds (Chaudhari and Roper, 2010). Gustation is not only responsible for detecting the basic tastes, but also solubilization in water, oil, or saliva by the receptors on the tongue and ultimately by the brain (Meilgaard et al., 2007). The five basic tastes - sweet, salty, sour, bitter, and umami – are important contributors to meat flavor (Chaudhari and Roper, 2010). Sweetness has been associated with glucose, fructose, ribose and L-amino acids such as glycine, alanine, threonine, lysine, asparagine, glutamine, and several others (MacLeod, 1994). Sourness stems from aspartic acid, glutamic acid, several organic acids, and carboxylic acids (MacLeod, 1994). Inorganic salts and the sodium salts of glutamate and aspartate are responsible for

salty flavors; while bitterness may be derived from hypoxanthine, anserine, carnosine and other peptides, as well as particular amino acids (MacLeod, 1994). Umami has a flat, salty, and brothy taste according to the beef lexicon (Adhikari et al., 2011). Umami has a characteristic savory quality that is supplied by glutamic acid, monosodium glutamate (MSG), 5'-inosine monophosphate (IMP), 5' -guanosine monophosphate (GMP) and certain peptides with glutamate being the most important contributor (MacLeod, 1994). Other senses, such as astringency, mouthfeel, and juiciness may also play a part in how flavor is perceived (Farmer, 1994).

Taste is also commonly confused with somatosensory sensations (trigeminal senses) such as the cool of menthol used in mouthwashes or the heat from capsaicin found in chili peppers (Chaudhari and Roper, 2010). Chaudhari and Roper (2010) go on to describe that both of these products stimulate ion channels in somatosensory nerve fibers. Capsaicin and related compounds may stimulate interactions between somatosensory trigeminal nerve fibers in the tongue and taste buds. Other somatosensory influencers include texture and visual cues. For example, fatty taste, which is important for meat and meat products, lies at the intersection of somatosensory and gustatory perception, and may become recognized as another basic taste (Chaudhari and Roper, 2010). Taste buds are defined as clusters of neuroepithelial cells that form compact, columnar “islands” embedded in the oral cavity – in humans there are about 5,000 taste buds. Taste buds are situated on the superior surface of the tongue, on the palate, and on the epiglottis. Taste buds within the oral cavity perform similar functions, leading the concept of a “tongue map” to be discredited (Lindemann, 1999).

Flavor is enhanced by the olfactory and gustatory systems working together. The olfactory senses are used for the detection of the aroma during tasting. Olfactory neurons that detect the volatile compounds are responsible for aromatic sensation perceived by the brain (Meilgaard et al.,

2007). This system is able to discriminate among many different aromas and can identify hundreds or even thousands at a time (Breer, 2008; Farmer, 1994). As Breer (2008) describes, the sensory and hedonic evaluation of most food-related flavors is mainly dependent on olfactory perception. The sense of smell is able to recognize and discriminate myriad of airborne molecules with great accuracy and sensitivity. Odor compounds may reach the receptors in the nasal epithelium either through the nose via smelling or through the posterior nares at the back of the throat while chewing (Breer, 2008).

Chemical Development of Flavor

There are many compounds released during the cooking process of meat. The characteristic flavor of cooked meat derives from thermally-induced reactions occurring during heating, including the Maillard reaction, lipid thermal degradation, or the lipid-Maillard reactions. These reactions are important for the flavor compounds of cooked foods (Mottram, 1998). There are hundreds of compounds in meat that contribute to flavor and aroma, many of them are altered through storage and cooking (Calkins and Hodgen, 2007). Each compound by itself is unique, but when they are combined and released during cooking, meat will develop its characteristic flavor (Farmer, 1994).

Maillard Reaction

The Maillard reaction is an important contributor to the cooked flavor of beef and other meat products. This reaction is what creates the browning effect of steak and other foods when cooked. It is a type of non-enzymatic browning that involves the reaction of carbonyl groups with free amino acids when cooked at higher temperatures (Kerth and Miller, 2015). Browning of meat can also occur at room temperature, refrigeration (such as

dehydrated foods), or during caramelization of sugar, but neither of these are due to the Maillard reaction (Kerth and Miller, 2015). The complex nature of the Maillard reaction provides many attributes to flavor, off-flavor, aroma, and odor but the main flavors developed from the reaction are sweet and bitter (Hurrell, 1982). Other flavors developed by the Maillard reaction include roasted, browned, meaty, caramelized, and more (Kerth and Miller, 2015).

During the Maillard reaction, amino compounds condense with the carbonyl group of a reducing sugar in the presence of heat (Calkins and Hodgen, 2007). Calkins and Hodgen (2007) go on to describe that this produces glycosylamine which is rearranged and dehydrated to form furfural, furanone derivatives, hydroxyketones, and dicarbonyl compounds, all of which contribute to flavor. As the reaction progresses, the intermediates can react with other amines, amino acids, aldehydes, hydrogen sulfide, and ammonia through the Amadori rearrangement, Strecker degradation, and Schiff bases pathways. Once the reaction has progressed through the Schiff base, Strecker degradation, or other pathways, the reactions can lead to melanoidins (Calkins and Hodgen, 2007; Fay and Brevard, 2005). This produces products that are either pleasing or unacceptable aromas and flavors. The type of product that is created depends on the various types of sugar and amino groups. For example, cysteine and glucose produce mainly sulfur compounds whereas cysteine and glucose under oxidized conditions produce more pyrazines and furans (Calkins and Hodgen, 2007; Tai and Ho, 1997). Calkins and Hodgen (2007) discussed how there should be more extensive research performed to determine if there are differences among muscles, cooking methods, degrees of doneness, and how these factors may affect the flavor profile of cooked meat.

A subsequent stage of the Maillard reaction that further contributes to flavor development is the Strecker degradation of amino acids by dicarbonyl compounds formed in the Maillard reaction (Mottram, 1998). The amino acid is decarboxylated and deaminated forming an aldehyde, while the dicarbonyl is converted to an aminoketone or aminoalcohol (Mottram, 1998; Kerth and Miller, 2015). Heterocyclic compounds (especially those containing sulfur), derived from ribose and cysteine, seem to be particularly important for the characteristic aroma of meat as well as savory, meaty, roast, and boiled flavors. In meat, the main sources of ribose are inosine monophosphate and other ribonucleotides (Mottram, 1998).

Lipid Thermal Degradation

Lipid degradation can contribute to the desirable flavor of cooked meat in several ways; fatty acids undergo a thermal oxidative change producing compounds that can influence aroma (Mottram and Edwards, 1983). Fatty acids also may react with components from lean tissue to create different flavor compounds, or act as a solvent for aroma compounds accumulated during production, processing and cooking of meat (Mottram and Edwards, 1983). Thermal degradation of lipid highly influences the development of beef flavor by producing several hundred volatile compounds including aliphatic hydrocarbons, aldehydes, ketones, alcohols, carboxylic acids and esters (Mottram, 1998). Mottram describes how thermal degradation of lipid provides compounds which give cooked meats a fatty aroma and compounds which determine the varying flavors and aromas between meat products derived from different species. Lipid degradation products tend to contribute to flavor to a greater extent than Maillard reaction products due to the breakdown of lipids

instead of water-soluble compounds as in the Maillard reaction (Mottram, 1998). The lipid compounds tend to be more dominant in flavor development, with the exception of more Maillard reaction products during high-heat cooking methods that cause large amounts of browning (Mottram, 1998).

Lipid-Maillard Reactions

Lipid-Maillard reactions occur when volatiles produced from thermal lipid degradation interact with Maillard reaction products producing more volatile flavor compounds (Melton, 1999). These lipid oxidation products enter the Maillard reaction during the Strecker degradation, ending in other volatiles not formed by meat precursors. Melton (1999) stated that generally, phospholipids in meat contribute the fatty acids that interact with Maillard reaction products. Mottram and Edwards (1983) concluded that the removal of triacylglycerols from lean beef caused no significant chemical or sensory aroma differences, but removal of both triacylglycerols and phospholipids resulted in a less meaty, more roasted aroma, lower concentrations of oxidation products and higher levels of heterocyclic compounds. As stated simply by Kerth and Miller (2015), it is possible for lipid and Maillard compounds to interact, but these interactions result in mild volatiles compared to the intensity of each primary reaction. These interactions of Maillard and lipid degradation products have been confirmed and provide a mechanism, that enables both interaction products to be controlled by the cooking process (Elmore et al., 1999).

Gas Chromatography and Mass Spectrometry

Flavor analysis has been conducted for many years using a variety of methods in order to develop new products, understand existing products, examine shelf-life, and to provide quality foods and other products (Chambers and Koppel, 2013). There are two primary forms of flavor analysis, sensory and instrumental. Sensory descriptive methods that have been developed are highly reliable and able to determine the human perception of flavor (Chambers and Koppel, 2013). For odors, sensory is the preferred method for evaluation, but since it can be expensive and time consuming, instrumental evaluation is also used for quicker results.

In foods and beverages, headspace analysis is an option for instrumental determination of volatile compounds in a sample as the headspace contains volatiles that are responsible for the odor sensation (Chambers and Koppel, 2013). Two common methods in instrumental volatile compound measurement are gas chromatograph/mass spectrometer (GC/MS) and a GC/MS coupled with an olfactometric port or a sniff port (Chambers and Koppel, 2013). Gas chromatograph/mass spectrometer systems are used to identify flavor aroma and compounds in flavor research. The GC/MS system uses four steps to evaluate the compounds: collection of volatiles, separation of volatile compounds, identification of each compound, and quantification of each compound (Chambers and Koppel, 2013). This instrumental technique is often used and accepted in flavor studies of muscle foods (Shahidi, 1994). There are several options to isolate and concentrate the volatile compounds from the matrix, such as steam distillation/extraction or supercritical CO₂ extraction or the solid phase microextraction (Chambers and Koppel, 2013). Solid phase microextraction is a popular technique used in flavor analysis as it is a simple, low-cost, solvent-free and sensitive

technique for the analysis of volatile compounds with a wide boiling point range (Ma et al., 2013). The volatiles are collected in the headspace of a container, then the SPME is injected into the GC/MS and desorbed. The volatiles are separated by the GC into individual compounds as the MS identifies the compounds (Laird, 2015). There are thousands of compounds that can be identified even though some may not be aromatic.

In recent years, the addition of the GC/O has modernized flavor research and determines which compounds have aromas by allowing for identification of aroma-active compounds. Shahidi (1994) explained that the volatiles are separated by the GC column then transported to the olfactory port, where they are combined with humidified air to prevent nasal passages from drying out when sniffed. Panelists that sniff for the aromatic compounds are thoroughly trained to identify volatiles of cooked meat, and the GC/O is there to help identify volatiles that are odor-active as well as non-odor-active from panelist detection (Laird, 2015). As the odor-active volatiles travel through the column, the panelist is able to record the smell and its intensity creating an aromagram. The compounds are also being recorded through the MS creating a chromatogram, then the aromagram and chromatogram are compared to determine which compounds are producing an odor. The identified volatile compounds can be used to correlate with sensory panels and which volatile aroma compounds correlate with overall consumer like and dislike.

Factors Influencing Flavor

Robbins et al. (2003) reported tenderness, flavor, and juiciness were the most important factors influencing consumer's eating satisfaction of beef. Flavor was an important eating characteristic when meat products were served (Ma et al., 2013; Behrends et al., 2005). Meat flavor is thermally derived, because uncooked meats have little or no aroma and only possess what is described as "a blood-like taste". During cooking, the volatile compounds generated between non-volatile components of lean and fatty tissues of meat through a complex series of thermally induced reactions, contribute to the aroma attributes and characteristic flavors of meat (Ma et al., 2013).

Muscle Comparison

Hundreds of compounds contribute to meat aroma and flavor because their complex interactions influence the perception of meat flavor. Flavor of meat can be influenced by lipid content, oxidation, animal's diet, pH, and myoglobin (Calkins and Hodgen, 2007). While all these factors influence flavor, Calkins and Hodgen (2007) reported that new research revealed that there was a relationship between certain muscles within a single beef carcass and flavor. They stated that this animal effect included the presence of off-flavors. Also, the part of the animal's diet that can contribute to an off-flavor was a diet high in polyunsaturated fatty acids.

Most research that compared muscles dealt with tenderness because there was high variation between muscles in tenderness compared to flavor (Shackelford et al., 1995; Wulf and Page, 2000; Calkins and Hodgen, 2007). Calkins and Hodgen (2007) reported the rankings of muscles for flavor intensity (most intense to least/bland) from different studies.

In a study by Jeremiah (2003a; 2003b), the diaphragm (skirt) was ranked the most intense and the *M. obliquus abdominis internus* (flap) was ranked the seventeenth most intense out of twenty-five muscles. However, statistically the differences in beef flavor intensity between muscles were relatively small.

Muscle flavor will also vary depending on muscle composition and structure. Intramuscular fat deposited between muscle bundles disrupts the endomysium's honeycomb structure and thin perimysial connective tissue fibers that increases tenderness (Jeremiah et al., 2003b). When meat is cooked, the fat will melt and create moisture within the product. Moisture influences juiciness directly and tenderness indirectly, but both influence overall flavor (Jeremiah et al., 1978, 2003b). Two components determine the tenderness of muscle: the myofibrillar component and the stromal component (Jeremiah et al., 1978, 2003b). Postmortem conditions before, during, and after the onset of rigor mortis are the main influences of the myofibrillar component; cooking and animal characteristics are the primary influencers of the stromal component (Jeremiah et al., 2003b). In raw meat, the connective tissue is influenced by gender and age, while gender, breed, and animal age influence the collagen solubility (Jeremiah et al., 2003b).

In the US, beef fajitas have become a popular dish served in Mexican restaurants (Huerta-Montauti, 2008; Huerta Sanchez, 2009). Since this dish has become so desired, the demand for both inside (*M. transversus abdominus*) and outside skirt (*M. diaphragma pars costalis*) steaks, the principal muscles prepared as fajitas, has dramatically increased. With the increased demand of fajita meat, research has been conducted on the flavor of skirts and other thin muscles to come up with alternatives. Tenderness is an important factor for flavor and consumer acceptance. Smith et al. (1979) suggested that it is likely that trained sensory

panel members rate all palatability traits higher if tenderness is adequate; tough samples may be perceived as generally unsatisfactory in all palatability traits (Huerta Sanchez, 2009).

The specific cuts in this study include outside and inside skirt steaks, and flap steaks. Beef skirt steaks prior to the 1980s were used for ground beef production. However, in the 1980s, demand for these cuts increased dramatically (Recio et al., 1988). Recio et al. (1988) explained that outside skirt steaks (diaphragm muscle) became more valuable due to the Japanese market having created a demand. In the late 80's, the inside skirt (internal abdominal oblique muscle) as well as the outside skirt increased in value due to the rising popularity of the beef "fajita" in the United States. Recio et al. (1988) obtained beef inside and outside skirts (n = 120) from 30 Choice and 30 Utility beef carcasses from two maturity groups (A and D maturity). They evaluated anatomical location, grade/maturity group, and method of mechanical tenderization on the palatability of cooked "fajita" beef. The skirts were vacuum-packaged and aged for ten days at 2°C. The skirt steaks then were removed from their packages and assigned randomly to one of twelve treatments (n = 10 per treatment). Treatment combinations included blade tenderization, cubing, and no tenderization. All treatments were tempered to 4°C and broiled to an internal temperature of 80°C for sensory evaluation and chemical determinations. Using a trained panel, steaks were evaluated for juiciness, muscle fiber and overall tenderness, connective tissue amount, off-flavor, and overall palatability. Recio et al. (1988) found that ratings for juiciness, muscle fiber tenderness, overall tenderness, and overall palatability were higher for outside skirts than for inside skirts. Skirt steaks, inside and outside, from Choice carcasses were juicier, more tender overall, and more palatable overall than skirt steaks from Utility carcasses.

Cubing, as a method of tenderization, was more useful for skirts from Utility carcasses than for skirt steaks from Choice carcasses.

Internal Temperature Endpoint

Raw meat has been described as weak, salty, and blood-like in flavor and as the degree of doneness (internal temperature endpoint) increases, the desirable characteristic beefy flavors evolves (Crocker, 1948). The temperature of the heating element and the method of cooking affect the rate of cooking (Crocker, 1948). The rate and extent of chemical reactions are impacted by the method of cooking combined with the final degree of doneness (Kerth, 2013). Cooking method and final temperature greatly affect what flavor volatiles may develop from the flavor compounds that are present in raw beef (Miller and Kerth, 2012). According to Glascock (2014), as degree of doneness (internal endpoint temperature) increased, beef flavor identity increased.

A range of internal endpoint temperatures is acceptable for beef cookery. The National Livestock and Meat Board described temperature endpoints for beef as very rare, 55°C; rare, 60°C; medium rare, 65°C; medium, 70°C; well done, 75°C; and very well done, 80°C (Bowers, 1987). Meat cooked to the various endpoint temperatures has different characteristics. Textural changes that occur as muscle is heated have been well documented by instrumental measurements or evaluation by sensory panels (Bowers, 1987; Bouton and Harris, 1981; Draudt, 1972; Martens et al., 1982; Penfield and Meyer, 1975). Flavor changes that occurred as temperature increased have not been characterized. In other studies, the “desirability” of flavors of beef cooked to various endpoints has been evaluated, but specific flavor characteristics have not been analyzed. By varying cooking methods and internal

temperatures, Calkins et al. (2007) created different flavors ranging from bland to strong meaty notes, some with high grill-like flavor, and others were noticeably roasted. The color change of meat as it is heated is used as an indicator of degree of doneness (Bowers, 1987).

Quality Grade

Quality characteristics of muscle foods are influenced by muscle structure, chemical composition and environment, interaction of chemical constituents, postmortem changes, stress and preslaughter effects, product handling, processing and storage, microbiological populations, and meat cookery (Miller, 1994). Even though muscle food quality is influenced by the physical characteristics of the muscle, quality is product-specific and is a measurement of consumer acceptability (Miller, 1994). Smith et al. (1983) described Quality grade as a predictor for consumer palatability (tenderness, juiciness, and flavor) by indirectly assessing the extent to which flavor and aroma producing compounds and precursors are likely to be present in beef.

The two main factors used to determine Quality grade are degree of marbling and degree of maturity (Philip, 2011). Marbling refers to the amount and distribution of intramuscular fat within the ribeye. Beef cuts with high levels of marbling are expected to be more tender, juicy and flavorful than cuts with lower levels of marbling (Tatum, 2007; Philip, 2011). Carcasses are also evaluated for maturity, which refers to the age of the animal. There are five maturity groupings (A, B, C, D, and E) that are determined by the evaluation of the size, shape, and ossification of the carcass's bones and cartilage, and by evaluating the color and texture of the ribeye (Tatum, 2007). Beef from older cattle was more intense in flavor than younger cattle, but their meat was tougher due to the increase in

insoluble collagen linkages (Miller, 1994). Carcasses were assigned to one of the eight USDA Quality grades after these factors were determined; the eight Quality grades for beef are Prime, Choice, Select, Standard, Commercial, Utility, Cutter, and Canner (USDA, 1996). Consumers generally prefer beef that is categorized as Prime, Choice, and Select (Philip, 2011). These three grades are generally more flavorful. Several studies have been conducted to understand palatability differences between Quality grades (Smith et al., 1987; Miller et al., 1997). Results of these studies indicated that Prime steaks were more palatable than the rest of the USDA Quality grades (Smith et al., 1987) and Choice steaks had a higher flavor intensity than Select steaks (Miller et al., 1997). According to a study done by Behrends et al. (2005), Choice carcasses were significantly rated higher in overall like, tenderness, juiciness, and flavor than High Select carcasses.

Marbling is a key factor of determining Quality grade and largely impacts beef flavor. According to Miller (2001), the minimum level of intramuscular fat is 3% for consumer acceptance but beyond 7.3%, perception of flavor and acceptability is negatively effected. McBee and Wiles (1967) and Smith et al. (1983) showed flavor desirability increased as marbling score increased from practically devoid to moderately abundant. Smith et al. (1983) also concluded concentrations of flavor and aroma in beef ultimately evaluated the marbling score; Smith et al. (1983) also found that a higher marbling score decreased the presence of off-flavors. Finally, Miller (1997) found that all sensory scores for steaks from Choice carcasses were higher than Select carcasses.

Cooking Method

The most important extrinsic factor that impacts volatile aroma compounds is cooking method (Kerth and Miller, 2015). Cooking significantly affects the flavor and tenderness of muscle foods. Cooking method will affect both temperature and moisture content which controls chemical reactions such as lipid degradation and Maillard reactions (Aberle et al., 2001). The type of cooking method, specifically the difference between moist-heat and dry-heat cookery, will drastically change flavor development (Aberle et al., 2001). Moist-heat cooking includes methods that cook meat in a closed or partially closed system such as braising, stewing, boiling, or simmering (Laird, 2015). Another example of moist-heat cooking includes enclosed containers such as crockpots. Moist-heat cookery means the beef will be cooked at low temperatures that will prevent the surface of the beef to reach a high enough temperature for the Maillard reaction to occur, which would dehydrate the beef (Kerth and Miller, 2015). Dry-heat cooking such as grilling, broiling, or pan-frying uses higher temperatures which will cause the surface to turn brown or black due to the Maillard reaction (Kerth and Miller, 2015).

Cooking method used can also influence degree of doneness or internal temperature as a part of the process. As the degree of doneness and internal temperature increases, the length of time that you have to cook the meat will also increase. There will be a difference in flavor because of the difference in the physical characteristics of the inside of the product as well as the outside of the product because of the changes in flavor profile (Kerth and Miller, 2015).

Lorenzen et al. (1999) studied cooking method effects on top loin steaks with outdoor grilling being the most liked overall and Choice top loins had the highest flavor intensity

liking. Also, in some cities, pan-frying created juicier steaks. Neely et al. (1999) found two interactions for overall like. USDA Quality grade x degree of doneness and degree of doneness x cooking method. Higher ratings were generally given to steaks cooked to medium rare or less. Neely et al. (1999) found that moist heat cookery methods had higher liking ratings.

Tenderness

Ramsbottom and Strandine (1948) defined tenderness as the state of being easily comminuted or masticated. Tenderness is the most important factor that influences consumer satisfaction for beef palatability (Dikeman, 1987; Savell et al., 1989). Consumer research by Glascock (2014), Luckemeyer (2015), and Laird (2015) has shown that tenderness and juiciness are important texture attributes, however, flavor is a greater driver of consumer overall liking. It has been found that consumers in both home and restaurant settings could differentiate among steaks varying in WBSF values (Miller et al., 1995). Several factors, such as animal age and gender, rate of glycolysis, amount and solubility of collagen, sarcomere length, ionic strength, degradation of myofibrillar proteins, and breeding and feeding conditions, contribute to variations in tenderness (Koohmaraie, 1992; Crouse et al., 1991; Wheeler et al., 1990; Shackelford et al., 1991). Clearly there are many factors within the muscle that can effect tenderness but not all variation in tenderness is accounted for (Luckemeyer, 2015). Dikeman (1987) explained that it was evident how important tenderness is because the *psaos major* muscle has a bland flavor but is the most valuable muscle per unit weight due to it being consistently very tender.

Marbling has a large impact when predicting tenderness (Luckemeyer, 2015). There are four marbling theories that detail and visualize marbling's effect on tenderness (Berry et al., 1974; Savell and Cross, 1988). The four theories include: lubrication, bulk density, insulation, and strain. Lubrication theory details how fat melts during heating which causes a lubrication effect around the muscle fibers that make it easier to bite through. This indicates juiciness is also associated with tenderness, as the more fat that melts, the juicier the steak will be. The bulk density theory indicates that fat is less dense than lean tissues which causes soft pockets, so the more fat within the muscle the easier it will be to bite through. The insulation theory works as insurance, the more fat within the lean portions, the more the steak will be protected from heat during cooking, therefore it is less likely to be overcooked or burn. Finally, the strain theory suggests that the strength of connective tissue may be decreased by the deposition of intramuscular fat. This means that the fat will loosen the structure of the connective tissue fibers enough to aid in heat penetration (Carpenter, 1962).

In more recent years, there has been more research performed on post mortem effects on meat tenderness (Koochmaraie, 1992, 1996; Wheeler et al., 2000). A large contributor to meat tenderness is the calpain system. Post-mortem tenderization is due to enzymatic degradation of key myofibrillar and other proteins (Koochmaraie, 1996). Researchers improved meat tenderness during post-mortem storage of beef carcasses at refrigerated temperatures due to aging (Koochmaraie, 1992). Aging meat is an important part of beef production to improve tenderness. Calpains (m-calpain and μ -calpain) are the enzymes that enable the weakening of the Z disks as they assist in breaking down the muscle fiber's structural proteins such as titin, desmin, and nebulin. Koochmaraie (1996) reported that calpains were most likely the only proteases that were directly involved with the meat

tenderization process. Calpastatin, calpain's protein inhibitor, and Ca^{2+} regulate calpain activity in living cells (Koohmaraie, 1992). Both m-calpain and μ -calpain undergo autolysis in the presence of sufficient calcium with the eventual loss of activity (Koohmaraie, 1992). Autolysis in the presence of higher free calcium concentration is likely the reason why μ -calpain decreases in activity post mortem (Koohmaraie, 1992).

As previously mentioned, connective tissue contributes to the toughness of meat. In a study done by Cross et al. (1973) using a sensory panel, the percent soluble collagen related connective tissue to toughness. Collagen is a large contributing factor to variation in meat tenderness and texture since it is an abundant connective tissue protein. Collagen is the most abundant protein within the beef carcass (Aberle et al., 2001); their molecules are bound together through intermolecular crosslinks that provide structure and strength (Weston et al. 2002).

Tenderness differs among muscles from various anatomical locations due to the variation between the traits responsible for tenderness, such as myofibrillar protein degradation or connective tissue amount and type (Cross et al., 1973). Locomotive muscles, muscles with higher use, have more connective tissue than support muscles, or lower use muscles. They also will vary in the amount of soluble collagen. The presence of connective tissue within muscle is amazingly variable, depending on developmental stage, muscle position/function, animal breed, nutrition, exercise and injury (Purslow, 2005).

Aging

Meat aging influences flavor and is widely accepted that flavor improves with age then begins to degrade and turn rancid after a certain time (Touraille and Girard, 1985).

According to Gorraiz et al. (2002), beef aged up to 14 days increased in fatty flavor and other positive notes such as beefy and brothy, but aging beyond 14 days created negative flavors such as painty, cardboard, bitter, and sour. Flavor changes during aging resulted from proteolytic and lipolytic enzymes causing alteration of different compounds such as peptides, free amino acids, and fatty acids. The increase of off-flavors was also due to the increased carbonyl amounts that came with lipid oxidation of aged meat. Aging for more than 21 days decreased flavor identity and aging for 35 days resulted in an increase in metallic flavor (Yancey et al., 2005).

Aging environment has also proven to affect the flavor of the meat. Beef that is aged in high-oxygen environments develop burnt flavors (Rowe, 2002). Other studies have shown that dry-aged beef, or beef that is aged with controlled humidity without packaging, had increased beef flavor compared to those aged in vacuum or carbon dioxide packaging (Campbell et al., 2001; Sitz et al., 2006; Jeremiah and Gibson, 2003). In a study by Campbell et al. (2001), 14- and 21-day dry-aged steaks had higher beef intensities, dry-aged flavor, as well as brown/roasted aromas compared to wet-aged steaks. Dry-aged meat tends to create more off flavors due to its contact with oxygen and dehydration from moisture loss during dry aging.

Conclusion

Flavor is a complex and multidimensional concept. There are my intrinsic and extrinsic factors that will influence the flavor of beef. The way that you cook meat will affect the various attributes as well as how the muscle functioned in the live animal. This study will build on previous research to determine the importance of each factor.

Our objective is to evaluate the trained descriptive beef flavor and tenderness attributes, volatile flavor aromatic compounds, and Warner-Bratzler shear force in cooking method, degree of doneness, and Quality grade for inside skirts, outside skirts, and flap beef cuts.

Our hypothesis is that the flavor and tenderness attributes vary based on cooking method, degree of doneness, and Quality grade for inside skirts, outside skirts, and flap beef cuts.

CHAPTER III

MATERIALS AND METHODS

Product Selection and Preparation

USDA upper two-thirds Choice and USDA Select inside skirts (IMPS 121D), outside skirts (IMPS 121C), and flap boneless (IMPS 185A) subprimals were bought from Ruffino's Meats in Bryan, Texas. The inside and outside skirts were cut into 10.16 cm wide steaks and one steak was randomly assigned to cooking and internal temperature endpoint treatments for trained panel sensory evaluation while another steak was assigned for Warner-Bratzler shear force (WBSF). Flap steaks were cut in half and one half was randomly assigned to trained panel sensory evaluation with the other portion to WBSF. Each treatment used a different subprimal. The steaks were vacuum-packaged (B2470, Cryovac Sealed Air Corporation, Duncan, SC) in film with an oxygen transmission rate of 3-6 cc at 4°C (m², 24 h atm @ 4°C, 0% RH) and a water vapor transmission rate of 0.5-0.6 g at 38°C (100% RH, 0.6 m², 24 h). The steaks were aged for 14 d, frozen and stored at -40°C until evaluated. For each analysis, individual steaks were randomly selected and thawed in refrigerated (4°C) storage for 12 to 24 h.

Cooking

The steaks were cooked using a pan fry (Signature Enameled Cast Iron Skillet, 11 ¾ in, Le Creuset of America, Inc., Early Branch, SC), pan grill (Signature Enameled Cast Iron Square Skillet Grill, Le Creuset of America, Inc., Early Branch, SC), or outside grill methods

(Performance 4-Burner Liquid Propane Gas Grill with 1-side Burner, Char-Broil LLC. Columbus, GA). Steaks that were pan-fried were cooked using one tablespoon of canola oil. Pans were wiped clean between each steak being cooked.

The steaks were cooked using their respective pan fry, pan grill, or outside grill methods. The grills were preheated for 15 minutes to an approximate temperature of 177°C. Grill temperature was monitored using an infrared reader and steaks were only placed in pockets at the correct temperature. All steaks were turned upon reaching half of their desired internal cooked temperature. Steaks cooked to 58°C were flipped at approximately 29°C, steaks cooked to 70°C were flipped at approximately 35°C, and steaks cooked to 80°C were flipped at approximately 40°C. The steaks were cooked to an internal temperature of either 58, 70 or 80°C to represent medium rare, medium and well done steaks. Internal temperatures were monitored by iron-constantan thermocouples (Omega Engineering, Stamford, CT) inserted into the geometric center of each steak. Sensory was conducted as defined by AMSA (2015) and Meilgaard et al. (2007). Sensory evaluations were approved by the Institutional Review Board for Use of Humans In Research at Texas A&M University (IRB2016-0609M).

Expert, Trained Descriptive Meat Flavor Analysis

The samples (n = 324) were evaluated by an expert trained meat descriptive attribute panel that helped develop and validate the beef lexicon. This panel (n = 5) was retrained using similar beef for ten days as defined in this study and as defined by Adhikari et al. (2011). Beef flavor attributes (n = 23) were measured using a 16-point scale within each attribute (0 = none and 15 = extremely intense) as defined in Table 1. After training was

complete, panelists were presented twelve samples per day, over a period of 28 days. Each sensory day was divided into two sessions ten minutes apart. Prior to the start of each trained panel evaluation day, panelists were calibrated using one orientation or “warm up” sample that was evaluated and discussed orally. After evaluation of the orientation sample, panelists were served the first sample of the session and asked to individually rate the sample for each beef flavor lexicon attribute. Double distilled water, sparkling water and unsalted saltine crackers were available for cleansing the palette between samples. During evaluation, panelists were seated in individual breadbox-style booths separated from the preparation area, and samples were evaluated under red lights. Samples were served four minutes apart with six samples evaluated in a session.

After cooking, samples were cut into 1.27 cm cubes. Three cubes per sample were served in 59 mL clear, plastic soufflé cups previously tested to assure that they did not impart flavors on the samples. Samples were identified with random three-digit codes and served in random order. Samples were cut and served immediately to assure samples are approximately 37°C upon time of serving. When a sample finished cooking before time to serve, the steak was wrapped in foil, placed in Alto Sham, and held as needed for a maximum of 20 minutes.

Cooked Meat Volatile Flavor Evaluation

Volatiles were captured from the same steaks evaluated by the trained panelists at Texas A&M University. After samples were prepared for panelists, approximately 75g of 1.25 cm beef cubes were randomly selected and placed in foil with a new tag from half of the meat samples. Samples were placed in liquid nitrogen and frozen to -196°C. Samples were

stored at -80°C until volatile analysis. Volatiles were evaluated using the Aroma Trax gas chromatograph/mass spectrophotometer system with dual sniff ports for characterization of aromatics. This technology separates individual volatile compounds, identifies their chemical structure and characterizes the aroma/ flavor associated with the compound. Samples were placed in heated glass jars (473 mL) with a Teflon lid under the metal screw-top to avoid off-aromas and then set in a water bath at 60°C and thawed within the jar, then the headspace was collected with a Solid-Phase Micro-Extraction (SPME) Portable Field Sampler (Supelco 504831, 75 µm Carboxen/ polydimethylsiloxane, Sigma-Aldrich, St. Louis, MO). The headspace above each meat sample in the glass jar was collected for 2 h for each sample after the sample reached 60°C. The SPME was then injected in the injection port of the GC, where the sample was desorbed at 280°C. The sample was then loaded onto the multi-dimensional gas chromatograph into the first column (30m X 0.53mm ID/ BPX5 (5% Phenyl Polysilphenylene-siloxane) X 0.5 µm (SGE Analytical Sciences, Austin, TX). Through the first column, the temperature started at 40°C and increased at a rate of 7°C/minute until reaching 260°C. Upon passing through the first column, compounds were sent to the second column ((30m X 0.53mm ID; BP20, Polyethylene Glycol) X 0.50 µm (SGE Analytical Sciences, Austin, TX). The gas chromatography column then split into three different columns at a three-way valve with one going to the mass spectrometer (Agilent Technologies 5975 Series MSD, Santa Clara, CA) and two going to the two humidified sniff ports that were heated to a temperature of 115°C with glass nose pieces. The sniff ports and software for determining flavor and aroma were a part of the AromaTrax program (MicroAnalytics-AromaTrax, Round Rock, TX). Panelists were trained to accurately use the AromaTrax software, after they had also been trained according to the beef lexicon aromas.

Warner-Bratzler Shear Force Evaluation

Steaks were thawed 24 h prior to cooking in a 4°C cooler. The steaks were cooked using their respective pan fry, pan grill, or outside grill methods. The grills were preheated for 15 minutes to an approximate temperature of 177°C. All steaks were turned upon reaching half of their desired internal cooked temperature. Steaks cooked to 58°C were flipped at approximately 29.15°C, steaks cooked to 70°C were flipped at approximately 35°C, and steaks cooked to 80°C were flipped at approximately 40°C. Upon reaching their desired internal temperature, steaks were removed and cooled for approximately 4 h or until reaching room temperature. Internal temperature was monitored with a thermometer (Omega™ HH501BT, Stamford, CT) using a 0.02 cm diameter, iron-constantan Type-T thermocouple wire.

After cooling, steaks were trimmed of visible connective tissue in order to expose muscle fiber orientation. At least six 1.3 cm cores were removed from each muscle. Cores were removed parallel to the muscle fibers and sheared once, perpendicular to the muscle fibers, on a United Testing machine (United 5STM-500, Huntington Beach, CA) using an 11.3 kg load cell, and a WBSF attachment. The peak force (N) needed to shear each core was recorded, and the mean for each steak was used in statistical analysis.

Statistical Analyses

The trained panel descriptive flavor attributes, volatile compounds, and WBSF were analyzed using Proc Means and Proc GLMMIX procedures of SAS (version 9.4, SAS Institute, Cary, NC). A predetermined alpha of 5% was used in all analyses. For trained panel data, data was averaged across panelists for all data. Sensory day and order served were

defined as random variables. Data was analyzed within a each cut. Cooking treatment, Quality grade, internal cook temperature endpoint and their interactions were included as main effects. Least squares means were calculated and the pdiff function of SAS was used to determine differences between least squares means when significance was defined in the Analysis of Variance. For WBSF, shear day was included as a random variable. Principal component analysis (PCA) and partial least squares regression (PLS) were conducted using XLSTAT (v2013, Microsoft Corporation, Redmond, WA). Data is presented in bi-plots.

CHAPTER IV

RESULTS AND DISCUSSION

Expert, Trained Descriptive Meat Flavor Analysis

The beef flavor attributes, definition and reference standards used in this study are outlined in Table 1 (Adhikari, 2011). The juiciness and tenderness attributes were also included in Table 1 (AMSA, 2015). Descriptive sensory attributes were evaluated using 0 to 15 point scales where 0 = none and 15 = extremely intense. Animal hair, barnyard, green hay-like, chemical, chocolate, cooked milk, dairy, fishy, floral, leather, soapy, warmed over, medicinal, painty, petroleum, refrigerator stale, and smoky wood were not found in the meat samples and data were not presented. Positive beef flavors were identified such as beef identity, brown, roasted, bloody/serummy, fat-like, sweet, salty, and umami; negative flavors such as metallic, liver-like, sour, musty/earthy, and bitter were also identified in the treatments (Glascocock, 2014; Laird, 2015; Luckemeyer, 2015).

Flaps

Sensory data were examined to determine the effect of Quality grade, internal temperature endpoint, and cooking method in flap steaks (Table 2). Quality grade did not affect ($P \geq 0.05$) beef identity, brown, roasted, bloody/serummy, metallic, sweet, sour, salty, bitter, burnt, heated oil, smokey charcoal, or overall tenderness. However, Choice flap steaks were significantly higher in fat-like ($P = 0.0002$), umami ($P = 0.01$), buttery ($P < 0.0001$),

juiciness ($P = 0.0001$), and muscle fiber tenderness ($P = 0.04$) than Select flap steaks.

Cardboardy was higher in Select flap steaks compared to Choice flap steaks ($P = 0.004$).

Since Quality grade is a predictor for consumer palatability (tenderness, juiciness, flavor) according to Smith et al. (1983), it is expected that positive flavor attributes would be more prevalent in the Choice Quality grade. Beef cuts with high levels of marbling are expected to be more tender, juicy and flavorful than cuts with lower levels of marbling (Tatum, 2007; Philip, 2011). These results were expected as tenderness has been shown to be impacted by marbling, muscle fiber tenderness, and connective tissue (Berry et al., 1974; Koohmaraie, 1996). Attributes such as bloody/serumy, burnt, smokey charcoal, and roasted will depend more on degree of doneness and cooking method than Quality grade (Glascock, 2014; Laird, 2015; Luckemeyer, 2015).

Internal temperature endpoint did not affect ($P \geq 0.05$) beef identity, fat-like, umami, sweet, sour, salty, bitter, cardboardy, buttery, heated oil, muscle fiber tenderness, or overall tenderness in flap steaks. Brown was found to be the highest in flap steaks cooked to 80°C and lowest in flap steaks cooked to 58°C ($P = 0.03$). Roasted was also found to be the highest in flap steaks cooked to 80°C, lower in those cooked to 70°C, and lowest in steaks cooked to 58°C ($P < 0.0001$). These results were expected as the higher the internal cooked temperature, the more time there is for Maillard reactions to occur, which results in brown and roasted flavors. For bloody/serumy and metallic, there was no difference between steaks cooked to 58°C and 70°C, but steaks cooked to 80°C had significantly less bloody/serumy ($P < 0.0001$) and metallic ($P = 0.007$) flavors. According to Adhikari et al. (2011), metallic and bloody/serumy are closely related so it was to be expected that they differ in similar treatments. Steaks cooked to lower internal cook temperature endpoints will tend to have

higher bloody flavors as there is more of a raw meat taste associated with the steak (Glascock, 2014; Laird, 2015; Luckemeyer, 2015). There was significantly more burnt flavor in steaks cooked to 80°C than steaks cooked to other internal temperature endpoints ($P = 0.02$). Smokey charcoal was higher ($P = 0.04$) in steaks cooked to 80°C than those cooked to 58°C. Flap steaks cooked to 80°C had longer cook times. Finally, flap steaks cooked to 80°C were less juicy ($P < 0.0001$) than those cooked to 58°C or 70°C. The development of burnt and smokey charcoal flavors can be attributed to the Maillard reaction (Ames, 1992) and most likely contributed to cooked meaty aromas (MacLeod, 1994). The Maillard reaction is a non-enzymatic browning reaction that occurs when a carbonyl compound and a compound possessing an amino acid react (Ames, 1992). It should be noted that the thickness of the flap caused it to be on the grill longer to reach the desired internal temperature than other cuts, so the outer portions were exposed to heat for a longer time and would be expected to have more burnt flavor.

Cooking method did not influence ($P \geq 0.05$) umami, sweet, sour, salty, bitter, cardboardy, burnt, buttery, heated oil, muscle fiber tenderness, and overall tenderness sensory attributes. Flap steaks cooked on the outside grill were higher in beef identity ($P = 0.01$), brown ($P = 0.03$), and roasted ($P < 0.0001$) beef flavor attributes than pan fry and pan-grilled flap steaks. Flap steaks cooked on the outside grill were lower in bloody/serummy ($P < 0.0001$), fat-like ($P = 0.0007$), and metallic ($P < 0.0001$) beef flavor attributes than pan fry and pan-grilled flap steaks. Smokey charcoal flavor was higher for flap steaks cooked on the outside grill ($P = 0.01$). Lastly, juiciness was higher in pan-fried and pan-grilled ($P < 0.0001$) flap steaks.

Through previous research, it has been reported that consumers prefer grilling steaks outside rather than pan frying (Laird, 2015). Because of this, positive flavor attributes were likely to be associated with outside grilling. It was expected that beef identity, brown, roasted, and smokey charcoal, flavor attributes were higher in outside-grilled flap steaks. Since the flap steaks cooked using the pan-grilled and pan-fried cooking methods were in their own juices, and the excess fat drips away from the meat in the outside grill, steaks grilled outside lose moisture which causes steaks to have a lower cook yield and be less juicy.

Interactions for Quality grade by internal cook temperature endpoint was significant ($P = 0.01$) for green flavor (Table 3). Internal cook temperature endpoint by cooking method interactions were reported for musty/earthy flavor and connective tissue amount (Table 4). Select flap steaks cooked to the lowest internal cook temperature endpoint had slightly higher green flavor ($P = 0.01$). Flap steaks cooked using the pan fry method had slightly more green flavor than other flap steaks ($P = 0.01$). This result indicates that green flavor may come from the amount of fat, low cooked internal temperature, and/or Quality grade. Since the Select steaks cooked to 58°C had much more green detected, it is likely the cause is a combination of these. Select steaks were significantly more musty/earthy than Choice steaks ($P = 0.0002$). However, connective tissue in Select and Choice steaks did not differ ($P = 0.39$). In the internal temperature endpoint by cooking method interaction, steaks pan-fried to 58°C were ranked the highest for musty/earthy ($P = 0.01$). Steaks pan-fried to 70°C had the lowest ($P = 0.01$) musty/earthy flavor. Connective tissue amount was higher in the pan-grilled flap steaks cooked to 80°C than for pan-grilled flap steaks cooked to 58°C ($P = 0.02$).

A principal component analysis for trained descriptive flavor attributes and treatments is shown in Figure 1. Umami basic taste, beef identity, browned, roasted, and smokey charcoal flavors were associated with outside grilling for Choice flap steaks. Select flap steaks cooking using the outside grill and Select pan-fried flap steaks cooked to the highest internal temperature endpoint were associated with burnt, bitter, and cardboardy flavor attributes. Salty flavor was closely related to Choice flap steaks cooked to 58°C on the outside grill. Fat-like, buttery, sweet flavors, and overall tenderness, muscle fiber tenderness, and juiciness tended to be clustered with Choice pan-fried or pan-grilled flap steaks. Juiciness, bloody/serummy, and metallic were closely clustered indicating that juicier flap steaks had higher bloody/serummy and metallic flavors. Select pan-fried and pan-grilled flap steaks tended to be associated with heated oil, green, and sour flavors.

The results from the principal component analysis indicate indicate that Choice pan-fried and pan-grilled flap steaks are associated with positive flavor and texture attributes. Fat-like has been more closely associated with consumer overall liking (Glascok, 2014; Laird, 2015; Luckemeyer, 2015). This is due to the Choice steaks having a higher fat content due to Quality grade scoring. Steaks that are higher in fat will develop more beefy flavors that consumers enjoy.

Inside Skirts

Sensory data was examined to determine the affect of Quality grade, internal temperature endpoint, and cooking method in inside skirt steaks (Table 5). Quality grade did not affect ($P \geq 0.05$) beef identity, brown, fat-like, green, umami, sweet, sour, salty, bitter, burnt, buttery, heated oil, smokey charcoal, juiciness, connective tissue, or overall

tenderness. Select inside skirt steaks were more cardboardy than Choice steaks ($P = 0.02$). In a study by Beavers (2017), products with a lower fat content were also higher in cardboardy. Choice inside skirt steaks had more muscle fiber tenderness than Select steaks ($P = 0.04$).

Internal temperature endpoint did not affect ($P \geq 0.05$) fat-like, green, umami, sweet, sour, salty, cardboardy, buttery, muscle fiber tenderness, connective tissue, or overall tenderness. Steaks cooked to 80°C had a significantly higher beef identity ($P = 0.001$) and bitter ($P = 0.02$) than those cooked to 58°C and 70°C. Similar to this study's results, Glascock (2014) concluded that beef identity increased as internal temperature endpoint increased. Inside skirts cooked to 80°C had the highest brown ($P < 0.0001$) and those cooked to 58°C had the lowest brown flavor. Skirts cooked to 80°C had the highest burnt flavor ($P = 0.005$) while steaks cooked to 58°C had the lowest. Steaks cooked to 70°C had the highest heated oil flavor ($P = 0.001$), followed by 80°C, and lastly 58°C. Steaks cooked to 70°C and 80°C were higher in smokey charcoal flavor ($P = 0.002$) than those cooked to 58°C. On the other hand, steaks cooked to 58°C were more juicy ($P = 0.001$) than those cooked to the other internal temperatures. These results are expected due to the Maillard reaction. As previously discussed, the Maillard reaction is a browning reaction that occurs when meat is cooked at higher temperatures (Kerth and Miller, 2015). The Maillard reaction occurred when steaks were cooked to 70 and 80°C which is why they were higher in brown and smokey charcoal flavors. When the meat is cooked to a lower temperature, not as many beefy flavors have evolved (Crocker, 1948).

Cooking method tended to not impact flavor attributes for inside skirt steaks. Cooking method did not affect ($P \geq 0.05$) beef identity, brown, fat-like, green, umami, sweet, sour, salty, bitter, cardboardy, burnt, buttery, heated oil, juiciness, and connective tissue. Inside

skirt steaks cooked on the outside grill demonstrated more of the smokey charcoal attribute ($P < 0.0001$) than the steaks cooked via pan fry and pan grill. Steaks cooked on pan grill had higher muscle fiber tenderness ($P = 0.005$) than the ones cooked on the outside grill. Steaks cooked on pan grill were significantly more tender overall ($P = 0.006$) than pan fry and outside grill. It has been shown that cooking method significantly affects the flavor and tenderness of muscle since it will affect temperature and moisture content (Aberle et al., 2001).

Table 6 shows cooking method interactions with roasted and bloody/serumy as well as Quality grade by internal temperature endpoint interactions for the same two attributes. Cooking method did not have an effect on roasted ($P = 0.05$) or bloody/serumy ($P = 0.53$). When evaluating Quality grade by internal temperature endpoint interactions, Choice inside skirt steaks cooked to 80°C had the highest roasted flavor ($P = 0.01$), followed by Choice cooked to 70°C and Select cooked to 80°C. Choice steaks cooked to 58°C had the lowest roasted flavor. Generally for both Quality grades, the higher the internal temperature endpoint, the higher the roasted. On the other hand, Choice steaks cooked to 58°C had the highest bloody/serumy flavor ($P = 0.03$). Those cooked to higher temperatures had less bloody/serumy flavor interactions. Glascock (2014) and Luckemeyer (2015) also concluded that as roasted flavor was lower and bloody/serumy was higher when meat was cooked to lower final internal temperatures. These conclusions are due to when a steak is cooked to a lower degree of doneness and internal temperature, it will appear more bloody and will be more rare. The steak will not have as much roasted flavor if it is not cooked to a higher temperature.

The metallic flavor attribute had two interactions ($P < 0.05$), Quality grade by internal cook temperature endpoint and internal cook temperature endpoint by cooking method (Table 7). Choice steaks cooked to 58°C and Select steaks cooked to 58°C and 70°C had the highest metallic flavor ($P = 0.02$). Choice steaks cooked to 70°C and 80°C and Select steaks cooked to 80°C had low metallic flavor compared to the rest of the treatments. These results agree with a study by Belk et al. (1993) that reported meat cooked to a low degree of doneness had higher metallic flavors. There were slight differences between internal temperature endpoint by cooking method interaction. All three cooking methods cooked to 58°C and pan grill cooked to 70°C were higher ($P = 0.03$) than outside grill cooked to 70°C and pan grill and outside grill cooked to 80°C.

Musty/earthy flavor attribute had slight differences between Quality grade and internal temperature endpoint by cooking method interaction ($P < 0.05$; Table 8). Select inside skirt steaks had slightly higher levels of musty/earthy than Choice inside skirt steaks ($P = 0.02$). Steaks grilled outside to 58°C were lowest and those pan-fried to 70°C were the highest ($P = 0.03$) in the internal temperature endpoint by cooking method interaction. Musty/earthy is considered to be a negative off-flavor indicating that these treatments tend to be less palatable (Glascock, 2014)

A principal component analysis for trained descriptive flavor attributes and treatments is shown in Figure 3. Green, musty/earthy, and heated oil were related to Select inside skirts pan-fried to 70°C. Metallic and sour were related to Choice inside skirts pan-fried to 58°C. Burnt, roasted, and smokey charcoal were related to Choice inside skirt steaks grilled outside to 80°C. Select inside skirts grilled to 80°C were related to umami basic taste. Liver, salty, and bitter were related to Choice inside skirt steaks grilled outside to 70°C. Beef identity was

related to Choice inside steaks pan-grilled to 80°C. Select inside skirt steaks grilled outside to 70°C were closely related to cardboardy. Overall sweetness and buttery were related to Choice inside skirt steaks cooked on the pan grill to 58°C. Results from the principal component analysis were similar to what we gathered from the least squares means.

Outside Skirts

Sensory data to examine the effect of Quality grade, internal temperature endpoint, and cooking method on outside skirt steaks was examined (Table 9). Quality grade did not affect ($P \geq 0.05$) beef identity, brown, metallic, liver-like, green, sweet, sour, salty, bitter, buttery, heated oil, musty/earthy, muscle fiber tenderness, or connective tissue. Choice outside skirt steaks were significantly more fat-like ($P = 0.04$) and juicy ($P = 0.02$) than Select steaks. On the other hand, Select outside skirt steaks were significantly more cardboardy ($P = 0.02$) than Choice outside skirt steaks. A study reported by Legako et al. (2016) concluded that as Quality grades increased, so did the presence of sweetness, but in the study we did not come to that conclusion for any of the cuts.

Internal temperature endpoint did not affect ($P \geq 0.05$) fat-like, liver-like, green, sweet, salty, bitter, cardboardy, buttery, heated oil, musty/earthy, muscle fiber tenderness, or connective tissue. Outside skirt steaks cooked to 70°C and 80°C were higher in beef identity ($P < 0.0001$) and brown ($P = 0.0004$) than 58°C. Outside skirts cooked to 58°C were highest in metallic flavor ($P < 0.0001$) and those cooked to 80°C had the least metallic flavor. Outside skirts cooked to 58°C had the most sour flavor ($P = 0.03$) and those cooked to 80°C had the least. Steaks cooked to 70°C had no difference in sour flavor from other internal temperature endpoints. These results agree with a study by Belk et al. (1993) that reported

meat cooked to a low internal temperature endpoint had higher metallic and sour flavors. Lastly, steaks cooked to 58°C were more juicy ($P < 0.0001$) than the other two treatments and steaks cooked to 70°C were more juicy than those cooked to 80°C. Lorenzen et al. (1999) stated that as degree of doneness increased, juiciness decreased and our results agree with this statement.

Cooking method did not affect ($P \geq 0.05$) fat-like, liver-like, sweet, sour, salty, bitter, cardboardy, buttery, heated oil, musty/earthy, juiciness, muscle fiber tenderness, or connective tissue. Steaks grilled outside were significantly higher in beef identity ($P < 0.0001$) and brown ($P = 0.0002$) flavor attributes than pan-fried and pan-grilled steaks. Outside skirt steaks that were pan-grilled had more metallic flavor ($P = 0.01$) than outside-grilled steaks. Steaks that were pan-fried had more green flavor ($P = 0.006$) than those that were grilled outside. The definition of metallic is the impression of slightly oxidized metal, such as iron, copper, and silver spoons (Table 1). The metallic flavor found in pan-grilled steaks is likely due to the pans being cast iron. The definition of green is sharp, slightly pungent aromatics associated with green/plant/vegetable matter such as parsley, spinach, pea pod, fresh cut grass, etc (Table 1). The green flavor that was indicated in the pan-fried steaks is likely due to the canola oil used when frying the steaks.

There were two types of interactions in the outside skirt treatment, internal temperature endpoint by cooking method and Quality grade by internal temperature endpoint. Burnt and smokey charcoal flavor attributes displayed an internal temperature endpoint by cooking method interaction presented in Table 10. Quality grade did not have a significant effect on burnt ($P = 0.53$) or smokey charcoal ($P = 0.67$). Overall, steaks that were grilled outside were higher in burnt ($P = 0.01$). Steaks that were grilled outside to 58°C and 70°C

were highest in smokey charcoal ($P = 0.001$), followed by those that were grilled outside to 80°C. Steaks that were pan-grilled and fried to 58°C and 70°C had the least detectable smokey charcoal flavors. These results are also due to the Maillard reaction that occurs when meat is cooked at higher temperatures (Kerth and Miller, 2011).

Umami and overall tenderness flavor and texture attributes had cooking method and Quality grade by internal temperature endpoint interactions presented in Table 11. Cooking method did not have a significant effect on umami ($P = 0.05$) or overall tenderness ($P = 0.75$). Choice outside skirt steaks cooked to 80 had the highest umami flavor ($P = 0.02$) and steaks cooked to 58°C had the lowest umami flavor. So, generally, steaks cooked for a longer period of time to a higher internal temperature endpoint, have a stronger umami flavor. Choice steaks cooked to 70°C and Select steaks cooked to 58°C were the highest in overall tenderness ($P = 0.02$), but Choice steaks cooked to 58°C were the least tender.

To understand relationships between descriptive sensory attributes and treatments for the inside skirt, a principal component analysis is presented in Figure 6. Select steaks pan-grilled to 70°C were closely related to green. Choice steaks cooked on an outside grill to 80°C and Select steaks cooked on an outside grill to 70°C were closely related to sweet, umami, beef identity, brown, connective tissue, and overall tenderness flavor and tenderness attributes. Choice steaks pan-grilled to 58°C and Choice/Select steaks pan-fried to 58°C were associated with juiciness, metallic, bloody/serummy, fat-like, sour and heated oil. Choice 70°C outside grill was most closely related to smokey/charcoal and burnt flavor attributes. Outside grilling of outside skirt steaks was associated with positive flavor attributes. Select outside skirt steaks pan-fried to either 80°C or 70°C were most closely clustered with the off-flavors

of liver-like and cardboardy flavor attributes indicating that pan frying most likely induced off-flavors.

Although the cuts were unique to this study, overall, the beef attributes results were expected and comparable to recent beef flavor studies with descriptive panel results by Glascock (2014), Luckemeyer (2015), and Miller and Kerth (2012).

Cooked Meat Volatile Flavor Evaluation

Flaps

Volatile aromatic chemicals are reported in Table 12. Eighty volatile aromatic compounds were reported for the different cuts. To understand relationships between volatile aromatic compounds and descriptive sensory attributes for the flap, a partial least squares regression biplot is presented in Figure 2. Choice steaks pan-fried to 58°C were closely related to 3-ethyl-benzaldehyde, 3-dodecen-1-al, (E)-2-heptenal, and 1-octanol. Kerth and Miller (2015) describe 1-octanol as having a waxy, green, citrus aroma and (E)-2-heptenal as having a intense green, sweet, apple skin aroma. Select steaks pan-grilled to 58°C were closely related to pentanal, styrene, and acetic acid. Select steaks pan-grilled to 80°C were closely related to 2-methyl-butanal, a malty, green, fruity, musty aroma (Kerth and Miller, 2015). Select steaks pan-fried to 58°C were closely related to (E)-2-nonenal which is described as having a fatty, green aroma (Kerth and Miller, 2015). Lastly, Choice steaks pan-grilled to 58°C were closely related to octane and 1-octene.

Many aromas that were close to flap steaks contained aromas that have been described as green, sweet, or similar. All the treatments cooked to an internal temperature of 70°C and some cooked to 80°C were related to butanoic acid, benzeneacetaldehyde, phenyl

acetaldehyde, and 2,6-dimethyl-pyrazine. Butanoic acid is described to be sweaty and rancid (Kerth and Miller, 2015), benzeneacetaldehyde is a sweet, floral, honey, rosy aroma (Kerth and Miller, 2015), phenyl acetaldehyde is described as a sweet, honey, rose aroma (Kerth and Miller, 2015), and 2,6-dimethyl-pyrazine is a compound that results from the Maillard reaction and results in roasted and caramel-like odors (Xiao et al., 2014).

Inside Skirts

For the inside skirts, a partial least squares regression biplot is presented in Figure 4. Select inside skirt steaks pan-fried to 70°C were closely related to nonanal, styrene, carbon disulfide, tetradecanal, (E)-2-octenal, trans-2-undecenal, and (E,E)-2,4-decadienal.

According to Kerth and Miller (2015), nonanal is a citrus-like, soapy aroma and styrene is sweet, balsamic, floral, and extremely penetrating. A study by Pham et al. (2008) found carbon disulfide to be a sulfur aromatic that lowered consumer acceptability. Choice inside skirt steaks pan-fried to 80°C were closely related to octane, pentanal, heptane, 2-ethyl-6-methyl-pyrazine, trimethyl-pyrazine, and 3-ethyl-2,5-dimethyl-pyrazine. Pyrazines are also a product of the Maillard reaction and have a distinct roasted aroma (Glascock, 2014). Pentanal is described to have a winey, fermented, bready aroma while 3-ethyl-2,5-dimethyl-pyrazine is a peanut, caramel, coffee, popcorn aroma (Kerth and Miller, 2015). Select steaks pan-fried to 80°C were closely related to 3-dodecen-1-al as well as 1-pentanol which is described as fusel, fermented, bread, cereal (Kerth and Miller, 2015). Choice steaks pan-grilled to 80°C were closely related to 2-butanone as well as benzeneacetaldehyde which is known to produce a sweet, rosy aroma (Kerth and Miller, 2015). Choice inside skirt steaks pan-grilled

to 70°C were related to 1-octanol. Select inside skirt steaks pan-grilled to 58°C were related to methyl-pyrazine and dihydro-2(3H)-furanone.

The rest of the treatments were all closely related with acetic acid, sulfur dioxide, and methyl-benzene. Acetic acid is described as a sour, vinegar aroma (Kerth and Miller, 2015). Sulfur dioxide has been shown to greatly contribute to meaty aromas (MacLeod, 1986). Sulfur-containing compounds are products of the Maillard reaction which is why they have this meat-like aroma. Methyl-benzene, also known as toluene, has been described to have a paint-like aroma.

Outside Skirts

Partial least squares regression biplots were presented to show relationships between volatile aromatic compounds and trained descriptive attributes in Figure 7. Select steaks grilled outside to 70°C were closely related to methyl thioacetate, 3-methyl-butanal, 2-methyl-pyrazine, and 2-heptanone volatile aromatic compounds. 3-methyl-butanal is described to have a malty aroma, 2-methyl-pyrazine has a nutty, brown, musty, roasted aroma (Kerth and Miller, 2015). 2-heptanone has a very different aroma from the previous two – it is described to smell cheesy, banana, and fruity (Kerth and Miller, 2015). Choice steaks pan-fried to 70°C were closely associated with benzeneacetaldehyde, 2-ethyl-6-methyl-pyrazine and buttery volatile aromatic compounds and flavor attribute. As mentioned previously, benzeneacetaldehyde is a sweet, floral aroma. Select steaks pan-fried and pan-grilled to 58°C were closely related to 2-butanone, 2-(hexyloxy)-ethanol, acetic acid, 1-octene, liver like, bloody, metallic, juiciness, sour, and musty/earthy volatile aromatic compounds and descriptive flavor attributes. 2-butanone is described as being chemical-like

and fruity-green (Kerth and Miller, 2015). Choice steaks pan-fried to 58°C were closely related to 3-ethyl-2,5-dimethyl-pyrazine, 3-dodecen-1-al, (E)-2-heptenal, 1-octenal, 1-heptanol, and heated oil volatile aromatic compounds and flavor attribute. 3-ethyl-2,5-dimethyl-pyrazine has a peanut, caramel, coffee, popcorn aroma; highly contrasting, (E)-2-heptenal has an intense green, sweet, apple skin aroma (Kerth and Miller, 2015). 1-heptanol also has a fruity and apple aroma (Kerth and Miller, 2015). Choice steaks pan-fried to 58°C were also closely related to hexanal, which is a compound known to be a product of lipid oxidation (Mottram, 2007). Select steaks grilled outside to 58°C were closely related to ethynyl-benzene.

The rest of the treatments are clustered around Phenyl acetaldehyde, methanethiol, trans-2-undecenal, dl-limonene, 2-acetyl-2-thiazoline, carbon disulfide, and (E)-2-nonenal. Kerth and Miller (2015) describe phenyl acetaldehyde as a sweet, honey, rose aroma. Their review also describes methanethiol as vegetable oil, alliaceous, eggy, and creamy. Undecenal is described to be soapy and metallic. Limonene aromatic is lemon-like and citrus. Finally, Kerth and Miller (2015) describe (E)-2-nonenal as a fatty and green aromatic.

Warner-Bratzler Shear Force Evaluation

According to the criteria established by Bellew et al. (2003), beef cattle carcass muscles may be classified by their shear force as: WBSF values less than 3.2 kgf (kilogram-force) are very tender; WBSF values between 3.2 kgf and 3.9 kgf are tender; WBSF values between 3.9 kgf and 4.6 kgf are intermediate; and WBSF values above 4.6 kgf are tough. Results from Warner-Bratzler shear force evaluation can be found in Table 13.

Flaps

Within the flap cut, there was a significant differences between the various Quality grades, internal temperature endpoints, and cooking methods ($P < 0.05$). Within the Quality grades, Select flaps had a higher shear force measurement than Choice flaps ($P = 0.04$). This result indicates that overall, Choice flaps were more tender than Select. Within the internal temperature endpoints, flaps cooked to 80°C had higher shear force values than those cooked to 58°C and 70°C ($P = 0.0004$). This indicates that flaps cooked to 80°C were the least tender. Within the three cooking methods, flaps cooked on the outside grill and pan fry both had higher shear force values than flaps cooked on the outside grill ($P = 0.002$). These results indicate the that flaps cooked on the pan grill were the most tender.

According to the criteria, both Quality grades are very tender. Flaps cooked to 80°C are tender and those cooked to 58°C and 70°C are very tender. Pan-fried and pan-grilled flaps are considered very tender, but flaps cooked outside are tender.

The WBSF results agreed with the trained panel results for tenderness between Quality grades. For both, Choice flaps were more tender than Select flaps when looking at the muscle fiber tenderness attribute. For internal temperature endpoints and cooking method, trained panelists found no differences between treatments for tenderness.

Inside Skirts

Within the inside skirt cut, there was a significant difference between the internal temperature endpoints and cooking methods ($P < 0.05$) but the two Quality grades were not different ($P = 0.20$). Within the internal temperature endpoints, skirts cooked to 70°C and 80°C had higher shear force measurements than the skirts cooked to 58°C ($P = 0.0001$).

These results indicate that the less the skirt is cooked, the more tender the meat is. Within the cooking methods, inside skirt steaks cooked on a pan grill had lower shear force measurements than skirts cooked on a pan fry and the outside grill ($P = 0.002$). This indicates that the pan grill inside skirts are the most tender.

According to the criteria, both Quality grades are considered intermediate. Inside skirts cooked to 70°C and 80°C are also considered intermediate, but steaks cooked to 58°C are tender. Inside skirt steaks cooked on the pan grill are classified in the tender category but skirts cooked on pan fry and outside grill are intermediate.

The WBSF results were slightly different than the trained panel results. According to WBSF, there was no difference between Quality grades, however trained panelists picked up differences between them in the muscle fiber tenderness attribute. Trained panelists did not pick up the difference in tenderness for internal temperature endpoints like the WBSF. The WBSF results for cooking method agreed with the trained panel, both indicate pan grill is more tender than pan fry and outside grill.

Outside Skirts

Within the outside skirt cut, there was not a difference between the internal temperature endpoints ($P = 0.88$), Quality grades ($P = 0.34$), or cooking methods ($P = 0.71$). These results indicate that outside skirt tenderness is not affected by the treatments used. This is likely due to the outside skirt cut being very thin. According to the criteria, overall outside skirts are very tender.

The WBSF were different than the trained panel results. As discussed, the WBSF results did not show any tenderness differences between treatments for the outside skirts.

However, the trained panelists found differences in the juiciness attribute. Also, for overall tenderness, there were differences in Quality grade by internal temperature endpoint during trained panel.

CHAPTER V

CONCLUSIONS

Beef flavor is very complex and has been identified as a key component of beef demand. It is impacted by marbling level, cooking method, and cooked internal temperature endpoint. Consumer acceptability of beef flavor is important to the beef industry, so research has been conducted through the Beef Checkoff program to determine what factors impact beef flavor. The purpose of this research is to assist in the development of the Beef Flavor Myology tool that will assist industry personnel when determining factors that impact beef flavor across various cuts, cooking methods, marbling levels, and cooked internal temperature.

For inside skirt, outside skirt, and flap steaks, cooking method and internal temperature endpoint tended to impact beef flavor more than USDA beef Quality grades. Choice steaks tended to have more positive beef flavor attributes such as umami, beef identity, brown, and roasted. Pan frying tended to result in more off-flavor development, but outside grilling created more positive flavor attributes. Pan-grilled steaks tended to have an intermediate flavor when compared to those that were grilled outside or pan-fried. internal temperature endpoint tended to effect beef flavor attributes such as bloody/serummy, metallic, burnt, and smokey charcoal.

For flap steaks, Quality grade, internal temperature endpoint, and cooking method all had a significant effect on tenderness. Choice flaps, flaps cooked to 58°C and 70°C, and flaps cooked on a pan grill were more tender than Select flaps, flaps cooked to 80°C, and pan-fried

and outside-grilled flaps. For inside skirts, there was no effect on tenderness due to Quality grade. Inside skirts cooked to 58°C and skirts that were pan-grilled will more tender than those cooked to 70°C and 80°C, as well as those that were pan-fried or grilled outside. Finally, for outside skirt steaks, there was no effect on tenderness by Quality grade, internal temperature endpoint, or cooking method.

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APPENDIX A
TABLES AND FIGURES

Table 1. Definition and reference standards for beef descriptive flavor aromatics and basic taste sensory attributes and their intensities where 0 = none and 15 = extremely intense from Adhikari et al. (2011).

Attributes	Definition	Reference
Flavor		
Beef Flavor ID	Amount of beef flavor identity in the sample.	Swanson's beef broth = 5.0 80% lean ground beef = 7.0 Beef brisket (160 °F)= 11.0
Bitter	The fundamental taste factor associated with a caffeine solution.	0.01% caffeine solution = 2.0 0.02% caffeine solution = 3.5
Bloody/Serumy	The aromatics associated with blood on cooked meat products. Closely related to metallic aromatic.	USDA Choice strip steak (60 °C) = 5.5 Beef brisket = 6.0
Browned	Aromatic associated with the outside of grilled or broiled meat; seared but not blackened or burnt.	Steak cooked at high temperature (internal 137 °F, seared on outside)
Burnt	The sharp/acrid flavor note associated with over roasted beef muscle, something over baked or excessively browned in oil.	Arrowhead Mills Puffed Barley Cereal= 3.0
Buttery	Sweet, dairy-like aromatic associated with natural butter.	Land O'Lakes Unsalted butter = 7.0
Cardboardy	Aromatic associated with slightly oxidized fats and oils, reminiscent of wet cardboard packaging.	Dry cardboard (1 in. square) = 5.0 (a) Wet cardboard (1 in. square, 1 cup water) = 7.0 (a)

Fat-Like	The aromatics associated with cooked animal fat.	Hillshire farms Lit'l beef smokies = 7.0 Beef suet = 12.0
Green	Sharp, slightly pungent aromatics associated with green/plant/vegetable matter such as parsley, spinach, pea pod, fresh cut grass, etc.	Hexanal (50 mL) in propylene glycol (10 mL) at 5000ppm = 6.5 (a) Fresh parsley water (25 g) steeped in water for 15 min then drained) = 9.0
Heated Oil	The aromatics associated with oil heated to a high temperature.	Wesson Vegetable Oil (1/2 cup, 3 min microwaved) = 7.0 Lay's Potato Chips (4 chips in medium snifter) = 4.0 (a)
Liver-Like	The aromatics associated with cooked organ meat/liver.	Beef liver (broiled) = 7.5 Brauschweiger liver sausage = 10
Metallic	The impression of slightly oxidized metal, such as iron, copper, and silver spoons.	0.10% Potassium Chloride solution = 1.5 Select strip Steak (cooked to 60 °C internal) = 4.0 Dole Canned Pineapple Juice = 6.0
Musty-Earthy/Humus	Musty, sweet, decaying vegetation.	Mushrooms = 0 1000 ppm of 2,6-Dimethylcyclohexanol = 9.0 (a)
Roasted	Aromatic associated with roasted meat.	Precooked Roast
Salty	The fundamental taste factor of which sodium chloride is typical.	0.15% sodium chloride solution = 1.5 0.25% sodium chloride solution = 3.5
Smoky Charcoal	An aromatic associated with meat juices and fat dripping on hot coals, which can be acrid, sour, burned, etc.	Wright's Natural Hickory seasoning (1/4 tsp. in 100 ml of water) = 9.0 (a)
Sour	The fundamental taste factor associated with citric acid.	0.015% citric acid solution = 1.5 0.050% citric acid solution = 3.5
Sweet	The fundamental taste factor associated with sucrose.	2.0% sucrose solution = 2.0

Umami	Flat, salty, somewhat brothy. The taste of glutamate, salts of amino acids and other molecules called nucleotides.	0.035% Accent Flavor Enhancer solution = 7.5
Tenderness		
Juiciness	The amount of perceived juice that is released from the product during mastication.	Carrot = 8.5; Mushroom = 10.0; Cucumber = 12.0 Apple = 13.5; Watermelon = 15.0 Choice top loin steak cooked to 58°C = 11.0 Choice top loin steak cooked to 80°C = 9.0
Muscle fiber tenderness	The ease in which the muscle fiber fragments during mastication.	Select eye of round steak cooked to 70°C = 9.0 Select tenderloin steak cooked to 70°C = 14.0
Connective tissue	The structural component of the muscle surrounding the muscle fiber that will not break down during mastication	Cross cut beef shank cooked to 70°C = 7.0 Select tenderloin cooked to 70°C = 14.0
Overall tenderness	Average of muscle fiber tenderness and connective tissue amount when connective tissue amount is 6 or less.	If connective tissue amount is 12 to 15, then overall tenderness = the value of muscle fiber tenderness; If connective tissue amount is then overall tenderness is the average of connective tissue amount and muscle fiber tenderness.

Table 2. Flap steak main effect least squares means for flavor and texture descriptive sensory attributes.¹

	Beef Identity	Brown	Roasted	Bloody/ Serummy	Fat-Like	Metallic	Umami	Sweet	Sour
<u>Quality grade</u>	0.50	0.87	0.62	0.12	0.0002	0.84	0.01	0.05	0.08
Choice	10.7 ± 0.11	10.9 ± 0.17	8.8 ± 0.18	2.3 ± 0.11	2.8 ^a ± 0.07	2.4 ± 0.08	4.0 ^a ± 0.11	1.9 ± 0.06	2.3 ± 0.05
Select	10.6 ± 0.11	10.9 ± 0.17	8.9 ± 0.18	2.1 ± 0.12	2.5 ^b ± 0.07	2.4 ± 0.08	3.7 ^b ± 0.11	1.8 ± 0.06	2.4 ± 0.05
<u>Internal Temp Endpoint</u>	0.33	0.03	<0.0001	<0.0001	0.08	0.007	0.37	0.41	0.25
58°C	10.5 ± 0.13	10.5 ^b ± 0.20	8.2 ^c ± 0.21	2.6 ^a ± 0.13	2.8 ± 0.08	2.5 ^a ± 0.09	3.7 ± 0.13	1.9 ± 0.07	2.5 ± 0.06
70°C	10.7 ± 0.13	10.9 ^{ab} ± 0.20	8.9 ^b ± 0.21	2.4 ^a ± 0.13	2.8 ± 0.08	2.4 ^a ± 0.09	4.0 ± 0.13	1.9 ± 0.07	2.4 ± 0.06
80°C	10.7 ± 0.13	11.2 ^a ± 0.20	9.4 ^a ± 0.20	1.8 ^b ± 0.13	2.6 ± 0.08	2.2 ^b ± 0.09	3.8 ± 0.13	1.8 ± 0.07	2.3 ± 0.06
<u>Cooking Method</u>	0.01	0.03	<0.0001	<0.0001	0.0007	<0.0001	0.11	0.75	0.16
Pan Fry	10.5 ^b ± 0.13	10.8 ^b ± 10.8	8.3 ^b ± 0.20	2.4 ^a ± 0.13	2.8 ^a ± 0.08	2.5 ^a ± 0.09	3.7 ± 0.13	1.9 ± 0.06	2.5 ± 0.06
Pan Grill	10.5 ^b ± 0.13	10.6 ^b ± 10.6	8.5 ^b ± 0.20	2.7 ^a ± 0.13	2.8 ^a ± 0.08	2.6 ^a ± 0.09	3.9 ± 0.13	1.9 ± 0.07	2.4 ± 0.06
Outside Grill	10.9 ^a ± 0.13	11.3 ^a ± 11.3	9.8 ^a ± 0.20	1.5 ^b ± 0.13	2.6 ^b ± 0.08	2.1 ^b ± 0.09	4.0 ± 0.13	1.9 ± 0.06	2.3 ± 0.06

^{abc}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 2 cont. Flap steak main effect least squares means for flavor and texture descriptive sensory attributes.¹

	Salty	Bitter	Cardboardy	Burnt	Buttery	Heated Oil	Smokey Charcoal	Juiciness	Muscle Fiber Tenderness	Overall Tenderness
<u>Quality grade</u>	0.37	0.52	0.004	0.70	<0.0001	0.56	0.64	0.001	0.04	0.10
Choice	2.2 ± 0.03	2.5 ± 0.09	1.2 ^b ± 0.10	1.0 ± 0.20	0.6 ^a ± 0.06	0.1 ± 0.03	1.2 ± 0.12	11.1 ^a ± 0.13	11.3 ^a ± 0.14	11.1 ± 0.16
Select	2.2 ± 0.04	2.6 ± 0.09	1.5 ^a ± 0.10	1.0 ± 0.20	0.3 ^b ± 0.06	0.1 ± 0.03	1.1 ± 0.12	10.5 ^b ± 0.13	11.0 ^b ± 0.15	10.8 ± 0.16
<u>Internal Temp Endpoint</u>	0.57	0.06	0.60	0.02	0.26	0.50	0.04	<0.0001	0.07	0.30
58°C	2.2 ± 0.04	2.5 ± 0.11	1.3 ± 0.12	0.7 ^b ± 0.23	0.5 ± 0.07	0.1 ± 0.04	0.8 ^b ± 0.15	11.3 ^a ± 0.15	11.3 ± 0.17	11.0 ± 0.19
70°C	2.2 ± 0.04	2.4 ± 0.11	1.3 ± 0.12	0.9 ^b ± 0.24	0.5 ± 0.07	0.1 ± 0.04	1.2 ^{ab} ± 0.15	11.1 ^a ± 0.15	11.3 ± 0.17	11.1 ± 0.19
80°C	2.2 ± 0.04	2.7 ± 0.11	1.5 ± 0.12	1.5 ^a ± 0.23	0.4 ± 0.07	0.1 ± 0.04	1.4 ^a ± 0.15	10.2 ^b ± 0.15	10.9 ± 0.17	10.7 ± 0.19
<u>Cooking Method</u>	0.27	0.34	0.10	0.29	0.10	0.28	0.01	<0.0001	0.10	0.26
Pan Fry	2.2 ± 0.04	2.5 ± 0.11	1.3 ± 0.12	1.1 ± 0.23	0.5 ± 0.07	0.1 ± 0.04	1.0 ^b ± 0.15	11.2 ^a ± 0.15	11.2 ± 0.17	11.0 ± 0.18
Pan Grill	2.3 ± 0.04	2.4 ± 0.11	1.3 ± 0.12	0.8 ± 0.23	0.5 ± 0.07	0.1 ± 0.04	0.9 ^b ± 0.15	11.3 ^a ± 0.15	11.3 ± 0.17	11.1 ± 0.19
Outside Grill	2.2 ± 0.04	2.6 ± 0.11	1.5 ± 0.12	1.2 ± 0.23	0.4 ± 0.07	0.1 ± 0.04	1.5 ^a ± 0.15	10.0 ^b ± 0.15	10.9 ± 0.17	10.7 ± 0.18

^{abc}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 3. Flap steak main effect and Quality grade by internal cook temperature endpoint least squares means for green flavor descriptive attribute.¹

Treatment	Green
<u>Cooking Method</u>	0.01
Pan Fry	0.2 ^a ± 0.04
Pan Grill	0.1 ^{ab} ± 0.04
Outside Grill	0.1 ^b ± 0.04
<u>Quality grade by Internal Cook Temp Endpoint</u>	0.01
Choice, 58°C	0.0 ^b ± 0.05
Choice, 70°C	0.1 ^b ± 0.06
Choice, 80°C	0.1 ^b ± 0.06
Select, 58°C	0.3 ^a ± 0.06
Select, 70°C	0.1 ^b ± 0.05
Select, 80°C	0.1 ^b ± 0.06

^{ab}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 4. Flap steak main effect and internal cook temperature endpoint by cook method least squares means for musty/earthy, and connective tissue descriptive attributes.¹

Treatment	Musty/Earthy	Connective Tissue
<u>Quality grade</u>	0.0002	0.39
Choice	1.2 ^b ± 0.09	11.3 ± 0.19
Select	1.6 ^a ± 0.09	11.2 ± 0.19
<u>Internal Cook Temperature Endpoint by Cooking Method</u>	0.01	0.02
Pan Fry, 58°C	1.9 ^a ± 0.16	11.1 ^{bc} ± 0.30
Pan Grill, 58°C	1.5 ^{ab} ± 0.16	10.7 ^c ± 0.30
Outside Grill, 58°C	1.2 ^{bc} ± 0.16	11.4 ^{abc} ± 0.30
Pan Fry, 70°C	1.1 ^c ± 0.17	11.5 ^{ab} ± 0.30
Pan Grill, 70°C	1.4 ^{bc} ± 0.16	11.3 ^{abc} ± 0.30
Outside Grill, 70°C	1.3 ^{bc} ± 0.17	11.5 ^{ab} ± 0.30
Pan Fry, 80°C	1.3 ^{bc} ± 0.16	10.8 ^{bc} ± 0.30
Pan Grill, 80°C	1.3 ^{bc} ± 0.16	11.9 ^a ± 0.30
Outside Grill, 80°C	1.5 ^{ab} ± 0.16	11.0 ^{bc} ± 0.30

^{abc}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹ 0 = none; 15 = extremely intense

Table 5. Inside skirt steak main effect least squares means for flavor and texture descriptive sensory attributes.¹

	Beef Identity	Brown	Fat-Like	Green	Umami	Sweet	Sour	Salty	Bitter
<u>Quality grade</u>	0.31	0.27	0.12	0.05	0.10	0.40	0.30	0.99	0.33
Choice	10.4 ± 0.09	10.6 ± 0.15	3.2 ± 0.10	0.1 ± 0.04	4.0 ± 0.09	2.0 ± 0.05	2.2 ± 0.05	2.2 ± 0.03	2.3 ± 0.07
Select	10.3 ± 0.09	10.5 ± 0.15	3.0 ± 0.10	0.1 ± 0.04	3.8 ± 0.09	1.9 ± 0.05	2.3 ± 0.05	2.2 ± 0.03	2.3 ± 0.07
<u>Internal Temp Endpoint</u>	0.001	<0.0001	0.33	0.21	0.21	0.50	0.07	0.14	0.02
58°C	10.3 ^b ± 0.11	10.1 ^c ± 0.17	2.1 ± 0.12	0.1 ± 0.04	3.8 ± 0.11	1.9 ± 0.06	2.4 ± 0.06	2.1 ± 0.04	2.2 ^b ± 0.08
70°C	10.1 ^b ± 0.11	10.5 ^b ± 0.17	2.9 ± 0.12	0.2 ± 0.04	3.8 ± 0.11	1.9 ± 0.06	2.2 ± 0.06	2.2 ± 0.03	2.3 ^b ± 0.08
80°C	10.7 ^a ± 0.11	11.1 ^a ± 0.17	3.2 ± 0.12	0.1 ± 0.04	4.0 ± 0.11	1.9 ± 0.06	2.2 ± 0.06	2.2 ± 0.03	2.4 ^a ± 0.08
<u>Cooking Method</u>	0.08	0.77	0.06	0.20	0.32	0.56	0.62	0.29	0.44
Pan Fry	10.2 ± 0.11	10.6 ± 0.17	3.2 ± 0.12	0.2 ± 0.04	3.8 ± 0.11	1.9 ± 0.06	2.2 ± 0.06	2.2 ± 0.03	2.4 ± 0.09
Pan Grill	10.5 ± 0.11	10.5 ± 0.18	3.2 ± 0.12	0.1 ± 0.04	4.0 ± 0.11	2.0 ± 0.06	2.3 ± 0.06	2.2 ± 0.03	2.4 ± 0.08
Outside Grill	10.4 ± 0.11	10.5 ± 0.17	2.9 ± 0.12	0.1 ± 0.04	3.8 ± 0.11	2.0 ± 0.06	2.3 ± 0.06	2.1 ± 0.03	2.3 ± 0.08

^{abc}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 5 cont. Inside skirt steak main effect least squares means for flavor and texture descriptive sensory attributes.¹

	Cardboardy	Burnt	Buttery	Heated Oil	Smokey Charcoal	Juiciness	Muscle Fiber Tenderness	Connective Tissue	Overall Tenderness
<u>Quality grade</u>	0.02	0.74	0.13	0.71	0.25	0.26	0.04	0.81	0.08
Choice	1.4 ^b ± 0.13	0.4 ± 0.13	0.7 ± 0.06	0.2 ± 0.04	1.1 ± 0.14	10.7 ± 0.16	9.7 ^a ± 0.20	9.9 ± 0.22	9.4 ± 0.19
Select	1.6 ^a ± 0.12	0.5 ± 0.13	0.6 ± 0.06	0.2 ± 0.04	0.9 ± 0.14	10.5 ± 0.16	9.2 ^b ± 0.20	9.9 ± 0.22	9.0 ± 0.19
<u>Internal Temp Endpoint</u>	0.22	0.005	0.31	0.001	0.002	0.001	0.23	0.34	0.25
58°C	1.3 ± 0.14	0.2 ^b ± 0.15	0.8 ± 0.08	0.1 ^b ± 0.05	0.7 ^b ± 0.16	11.1 ^a ± 0.19	9.7 ± 0.25	9.9 ± 0.25	9.4 ± 0.23
70°C	1.6 ± 0.14	0.4 ^{ab} ± 0.15	0.6 ± 0.08	0.3 ^a ± 0.05	1.1 ^a ± 0.16	10.5 ^b ± 0.19	9.2 ± 0.24	10.1 ± 0.24	8.9 ± 0.22
80°C	1.5 ± 0.14	0.8 ^a ± 0.15	0.6 ± 0.08	0.2 ^b ± 0.05	1.3 ^a ± 0.16	10.2 ^b ± 0.19	9.3 ± 0.24	9.7 ± 0.24	9.1 ± 0.22
<u>Cooking Method</u>	0.07	0.20	0.05	0.17	<0.0001	0.06	0.005	0.49	0.006
Pan Fry	1.6 ± 0.14	0.6 ± 0.15	0.6 ± 0.08	0.3 ± 0.05	0.8 ^b ± 0.16	10.4 ± 0.19	9.4 ^{ab} ± 0.24	9.8 ± 0.25	9.1 ^b ± 0.23
Pan Grill	1.3 ± 0.14	0.3 ± 0.15	0.8 ± 0.08	0.2 ± 0.05	0.6 ^b ± 0.16	10.9 ± 0.19	10.0 ^a ± 0.24	10.1 ± 0.24	9.7 ^a ± 0.22
Outside Grill	1.6 ± 0.14	0.5 ± 0.15	0.6 ± 0.08	0.2 ± 0.05	1.7 ^a ± 0.16	10.4 ± 0.19	8.9 ^b ± 0.24	9.8 ± 0.24	8.7 ^b ± 0.22

^{abc}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 6. Inside skirt steak main effect and Quality grade by internal temperature endpoint least squares means for roasted and bloody/serumy flavor descriptive attributes.¹

Treatment	Roasted	Bloody/serumy
<u>Cooking Method</u>	0.05	0.53
Pan Fry	8.7 ± 0.17	2.1 ± 0.13
Pan Grill	8.5 ± 0.16	2.4 ± 0.13
Outside Grill	9.0 ± 0.16	1.8 ± 0.13
<u>Quality grade by Internal Temperature Endpoint</u>	0.01	0.03
Choice, 58°C	7.7 ^d ± 0.22	2.9 ^a ± 0.16
Choice, 70°C	9.0 ^b ± 0.21	1.9 ^c ± 0.16
Choice, 80°C	9.7 ^a ± 0.22	1.7 ^c ± 0.16
Select, 58°C	8.3 ^c ± 0.21	2.4 ^b ± 0.16
Select, 70°C	8.8 ^{bc} ± 0.23	2.1 ^{bc} ± 0.17
Select, 80°C	9.1 ^b ± 0.22	1.8 ^c ± 0.16

^{abcd}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 7. Inside skirt steak Quality grade by internal cook temperature endpoint and internal cook temperature endpoint by cooking method least squares means for metallic flavor descriptive attribute.¹

Treatment	Metallic
<u>Quality grade by Internal Temperature Endpoint</u>	
Choice, 58°C	2.5 ^a ± 0.09
Choice, 70°C	2.1 ^b ± 0.09
Choice, 80°C	2.1 ^b ± 0.09
Select, 58°C	2.5 ^a ± 0.09
Select, 70°C	2.5 ^a ± 0.09
Select, 80°C	2.2 ^b ± 0.09
<u>Internal Temperature Endpoint by Cooking Method</u>	
Pan Fry, 58°C	2.5 ^{ab} ± 0.11
Pan Grill, 58°C	2.5 ^a ± 0.11
Outside Grill, 58°C	2.5 ^{abc} ± 0.10
Pan Fry, 70°C	2.3 ^{abcd} ± 0.11
Pan Grill, 70°C	2.5 ^{ab} ± 0.11
Outside Grill, 70°C	2.1 ^d ± 0.11
Pan Fry, 80°C	2.2 ^{bcd} ± 0.11
Pan Grill, 80°C	2.0 ^d ± 0.11
Outside Grill, 80°C	2.2 ^d ± 0.10

^{abcd}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹ 0 = none; 15 = extremely intense

Table 8. Inside skirt main effect and internal cook temperature endpoint by cooking method least squares means for musty/earthy flavor descriptive attribute.¹

Treatment	Musty/Earthy
<u>Quality grade</u>	0.02
Choice	1.4 ^b ± 0.09
Select	1.6 ^a ± 0.09
<u>Internal Temperature Endpoint by Cooking Method</u>	0.03
Pan Fry, 58°C	1.6 ^{ba} ± 0.18
Pan Grill, 58°C	1.6 ^{ab} ± 0.18
Outside Grill, 58°C	1.1 ^c ± 0.16
Pan Fry, 70°C	1.8 ^a ± 0.18
Pan Grill, 70°C	1.3 ^{bc} ± 0.17
Outside Grill, 70°C	1.5 ^{abc} ± 0.17
Pan Fry, 80°C	1.4 ^{abc} ± 0.16
Pan Grill, 80°C	1.4 ^{abc} ± 0.17
Outside Grill, 80°C	1.6 ^{ba} ± 0.17

^{abc}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 9. Outside skirt steak main effect least squares means for flavor and texture descriptive sensory attributes.¹

	Beef Identity	Brown	Fat-Like	Metallic	Liver-like	Green	Sweet	Sour	Salty
<u>Quality grade</u>	0.44	0.46	0.04	0.91	0.14	0.73	0.29	0.28	0.86
Choice	10.6 ± 0.11	10.6 ± 0.14	3.3 ^a ± 0.08	2.3 ± 0.07	0.4 ± 0.08	0.1 ± 0.03	2.1 ± 0.05	2.2 ± 0.04	2.2 ± 0.04
Select	10.5 ± 0.11	10.4 ± 0.13	3.1 ^b ± 0.08	2.3 ± 0.07	0.5 ± 0.08	0.1 ± 0.03	2.0 ± 0.05	2.3 ± 0.04	2.2 ± 0.04
<u>Internal Temp Endpoint</u>	<0.0001	0.0004	0.28	<0.0001	0.20	0.51	0.13	0.03	0.91
58°C	10.1 ^b ± 0.12	10.0 ^b ± 0.16	3.3 ± 0.09	2.6 ^a ± 0.08	0.4 ± 0.09	0.1 ± 0.03	1.9 ± 0.06	2.3 ^a ± 0.05	2.2 ± 0.04
70°C	10.7 ^a ± 0.13	10.7 ^a ± 0.16	3.3 ± 0.10	2.3 ^b ± 0.08	0.4 ± 0.10	0.1 ± 0.03	2.1 ± 0.06	2.2 ^{ab} ± 0.05	2.2 ± 0.04
80°C	10.7 ^a ± 0.12	10.8 ^a ± 0.16	3.1 ± 0.10	2.1 ^c ± 0.08	0.06 ± 0.09	0.1 ± 0.03	2.1 ± 0.06	2.2 ^b ± 0.05	2.2 ± 0.04
<u>Cooking Method</u>	<0.0001	0.0002	0.42	0.01	0.06	0.006	0.13	0.09	0.65
Pan Fry	10.3 ^b ± 0.13	10.4 ^b ± 0.16	3.2 ± 0.10	2.3 ^{ab} ± 0.08	0.6 ± 0.10	0.2 ^a ± 0.03	1.9 ± 0.06	2.2 ± 0.05	2.2 ± 0.04
Pan Grill	10.3 ^b ± 0.12	10.1 ^b ± 0.16	3.1 ± 0.10	2.5 ^a ± 0.08	0.5 ± 0.9	0.1 ^{ab} ± 0.03	2.0 ± 0.06	2.3 ± 0.05	2.2 ± 0.04
Outside Grill	10.9 ^a ± 0.13	11.0 ^a ± 0.1	3.1 ± 0.10	2.2 ^b ± 0.08	0.3 ± 0.10	0.0 ^b ± 0.03	2.1 ± 0.06	2.2 ± 0.05	2.2 ± 0.04

^{abc}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 9 cont. Outside skirt steak main effect least squares means for flavor and texture descriptive sensory attributes.¹

	Bitter	Cardboardy	Buttery	Heated Oil	Musty/ Earthy	Juiciness	Muscle Fiber Tenderness	Connective Tissue
<u>Quality grade</u>	0.58	0.02	0.81	0.47	0.36	0.02	0.91	0.80
Choice	2.3 ± 0.05	1.0 ^b ± 0.09	0.8 ± 0.07	0.3 ± 0.07	1.3 ± 0.08	11.2 ^a ± 0.14	11.9 ± 0.11	11.5 ± 0.15
Select	2.4 ± 0.05	1.2 ^a ± 0.09	0.7 ± 0.07	0.2 ± 0.07	1.4 ± 0.08	10.9 ^b ± 0.14	11.9 ± 0.11	11.5 ± 0.15
<u>Internal Temp Endpoint</u>	0.75	0.20	0.95	0.12	0.92	<0.0001	0.71	0.28
58°C	2.4 ± 0.07	1.0 ± 0.11	0.7 ± 0.08	0.4 ± 0.08	1.4 ± 0.10	11.5 ^a ± 0.15	11.9 ± 0.12	11.3 ± 0.17
70°C	2.4 ± 0.07	1.2 ± 0.11	0.8 ± 0.09	0.2 ± 0.08	1.3 ± 0.10	11.1 ^b ± 0.16	11.9 ± 0.13	11.5 ± 0.18
80°C	2.3 ± 0.07	1.2 ± 0.11	0.7 ± 0.08	0.2 ± 0.08	1.4 ± 0.10	10.7 ^c ± 0.15	11.8 ± 0.12	11.6 ± 0.17
<u>Cooking Method</u>	0.12	0.29	0.59	0.16	0.08	0.79	0.52	0.46
Pan Fry	2.3 ± 0.07	1.2 ± 0.11	0.8 ± 0.08	0.2 ± 0.08	1.5 ± 0.10	11.1 ± 0.16	11.9 ± 0.12	11.5 ± 0.18
Pan Grill	2.3 ± 0.07	1.0 ± 0.11	0.7 ± 0.09	0.3 ± 0.08	1.4 ± 0.10	11.1 ± 0.15	11.8 ± 0.12	11.3 ± 0.17
Outside Grill	2.5 ± 0.07	1.1 ± 0.11	0.7 ± 0.09	0.1 ± 0.08	1.2 ± 0.10	11.0 ± 0.16	12.0 ± 0.13	11.6 ± 0.18

^{abc}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 10. Outside skirt steak main effect and internal cook temperature endpoint by cooking method least squares means for burnt and smokey charcoal flavor descriptive attributes.¹

Treatment	Burnt	Smokey Charcoal
<u>Quality grade</u>	0.53	0.67
Choice	0.2 ± 0.11	0.8 ± 0.12
Select	0.3 ± 0.11	0.7 ± 0.11
<u>Internal Temperature Endpoint by Cooking Method</u>	0.01	0.001
Pan Fry, 58°C	0.1 ^b ± 0.21	0.3 ^{cd} ± 0.19
Pan Grill, 58°C	0.0 ^b ± 0.21	0.2 ^d ± 0.19
Outside Grill, 58°C	0.8 ^a ± 0.21	1.5 ^a ± 0.20
Pan Fry, 70°C	0.1 ^b ± 0.22	0.2 ^d ± 0.21
Pan Grill, 70°C	0.0 ^b ± 0.21	0.3 ^{cd} ± 0.20
Outside Grill, 70°C	0.9 ^a ± 0.22	1.9 ^a ± 0.20
Pan Fry, 80°C	0.2 ^b ± 0.21	0.6 ^{bcd} ± 0.20
Pan Grill, 80°C	0.3 ^{ab} ± 0.21	0.7 ^{bc} ± 0.19
Outside Grill, 80°C	0.0 ^b ± 0.21	1.0 ^b ± 0.19

^{abcd}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 11. Outside skirt steak main effect and Quality grade by internal cook temperature endpoint least squares means for umami and overall tenderness descriptive attributes.¹

Treatment	Umami	Overall Tenderness
<u>Cooking Method</u>	0.05	0.75
Pan Fry	3.8 ± 0.11	11.6 ± 0.13
Pan Grill	3.7 ± 0.11	11.4 ± 0.13
Outside Grill	4.1 ± 0.11	11.7 ± 0.13
<u>Quality grade by Internal Temperature Endpoint</u>	0.02	0.02
Choice, 58°C	3.5 ^c ± 0.15	11.2 ^b ± 0.17
Choice, 70°C	4.1 ^{ab} ± 0.16	11.8 ^a ± 0.18
Choice, 80°C	4.4 ^a ± 0.15	11.7 ^{ab} ± 0.17
Select, 58°C	3.6 ^c ± 0.15	11.8 ^a ± 0.17
Select, 70°C	3.8 ^{bc} ± 0.15	11.4 ^{ab} ± 0.18
Select, 80°C	3.8 ^{bc} ± 0.15	11.6 ^{ab} ± 0.18

^{abc}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

¹0 = none; 15 = extremely intense

Table 12. Volatile aromatic chemical compounds identified in the different cuts and their corresponding codes.

Code	Chemical Name
C1	2-Propanone
C2	Benzaldehyde
C3	Butanoic acid
C4	Carbon disulfide
C5	Hexanal
C6	Nonanal
C7	Octanal
C8	Heptanal
C9	Styrene
C10	Trimethyl-pyrazine
C11	Pentanal
C12	2-(Ethenyloxy)-propane
C13	2,6-Dimethyl-pyrazine
C14	Acetic acid
C15	Decanal
C16	Ethyl ester decanoic acid
C17	2-(Hexyloxy)-ethanol
C18	1-Heptanol
C19	2-Heptanone
C20	2-Methyl-butanal
C21	3-methyl-butanal
C22	Decane
C23	2-Pentyl-furan
C24	Heptane
C25	2-Ethyl-6-methyl-pyrazine
C26	3-Ethyl-2,5-dimethyl-pyrazine
C27	Methyl-pyrazine
C28	1-Octanol
C29	2-Decanone
C30	2-Decenal
C31	2-Heptenal
C32	2,4 Heptadienal
C33	3-Dodecen-1-al
C34	3-Ethyl-benzaldehyde
C35	Nonenal
C36	1-Octene
C37	2-Butanone
C38	Octane
C39	Methyl-benzene

C40	2-Ethyl-3,5-dimethyl-pyrazine
C41	2,5-Dimethyl-pyrazine
C42	2-Hexenal
C43	2-Nonenal
C44	2-Octenal
C45	2,4-Decadienal
C46	Cyclooctane
C47	trans-2-Undecenal
C48	Dihydro-2(3H)-furanone
C49	2-Undecenal
C50	Benzene
C51	Undecanal
C52	Methanethiol
C53	3-Hydroxy-2-butanone
C54	Hexanoic acid
C55	Sulfur dioxide(DOT)
C56	Tetradecanal
C57	Benzeneacetaldehyde
C58	2-methyl pyrazine
C59	Phenyl acetaldehyde
C60	Octanoic acid
C61	2-Ethyl-3-methyl-pyrazine
C62	Ethyl ester octanoic acid
C63	1-Octen-3-ol
C64	Undecenal
C65	2,4-Heptadienal
C66	2-Methyl-butane
C67	1-(4,5-Dihydro-2-thiazolyl)-ethanone
C68	1-Pentanol
C69	2-Docen-1-al
C70	Nonanoic acid
C71	Butyrolactone
C72	Tridecanal
C73	1-Hexanol
C74	Ethynyl-benzene
C75	Ethanol
C76	2,6-Diethyl-pyrazine
C77	Methyl thioacetate
C78	dl-Limonene
C79	2-Acetyl-2-thiazoline

Table 13. Warner-Bratzler shear force (kg) least square means.

	Flap	Inside Skirt	Outside Skirt
<u>Quality grade</u>	0.04	0.20	0.34
Choice	2.8 ^b ± 0.13	4.1 ± 0.18	2.2 ± 0.07
Select	3.1 ^a ± 0.13	4.2 ± 0.18	2.3 ± 0.07
<u>Internal Temp Endpoint</u>	0.0004	0.0001	0.88
58°C	2.6 ^b ± 0.15	3.6 ^b ± 0.20	2.2 ± 0.09
70°C	2.9 ^b ± 0.15	4.3 ^a ± 0.20	2.3 ± 0.09
80°C	3.3 ^a ± 0.15	4.6 ^a ± 0.20	2.3 ± 0.09
<u>Cooking Method</u>	0.002	0.002	0.71
Pan Fry	3.0 ^a ± 0.15	4.4 ^a ± 0.20	2.3 ± 0.09
Pan Grill	2.6 ^b ± 0.15	3.7 ^b ± 0.20	2.3 ± 0.09
Outside Grill	3.2 ^a ± 0.15	4.5 ^a ± 0.20	2.2 ± 0.09

^{ab}Mean values within a column and interaction followed by the same letter are not significantly different (P < 0.05).

Figure 1. Flap principal component analysis for descriptive sensory flavor attributes and the treatments

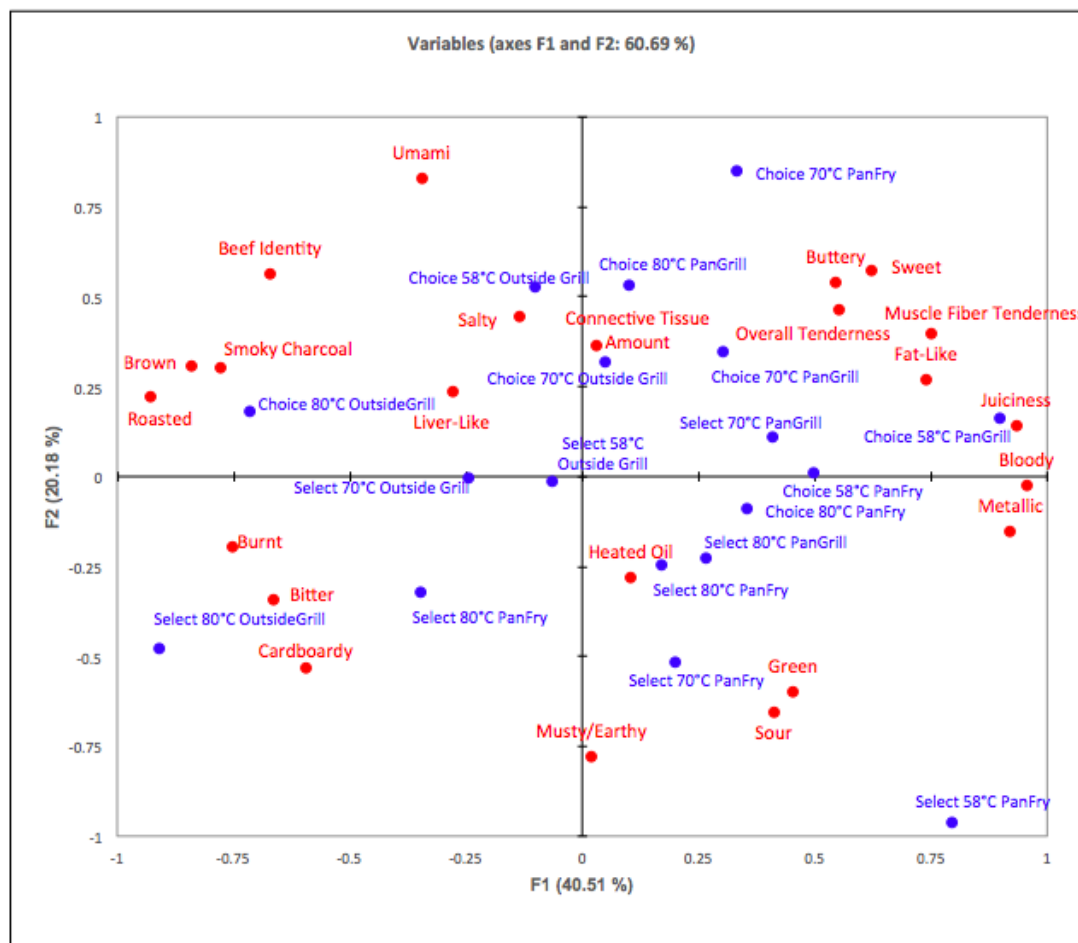


Figure 2. Flap partial least squares regression biplot for trained descriptive flavor, volatile aromatic compounds, and steak treatments.

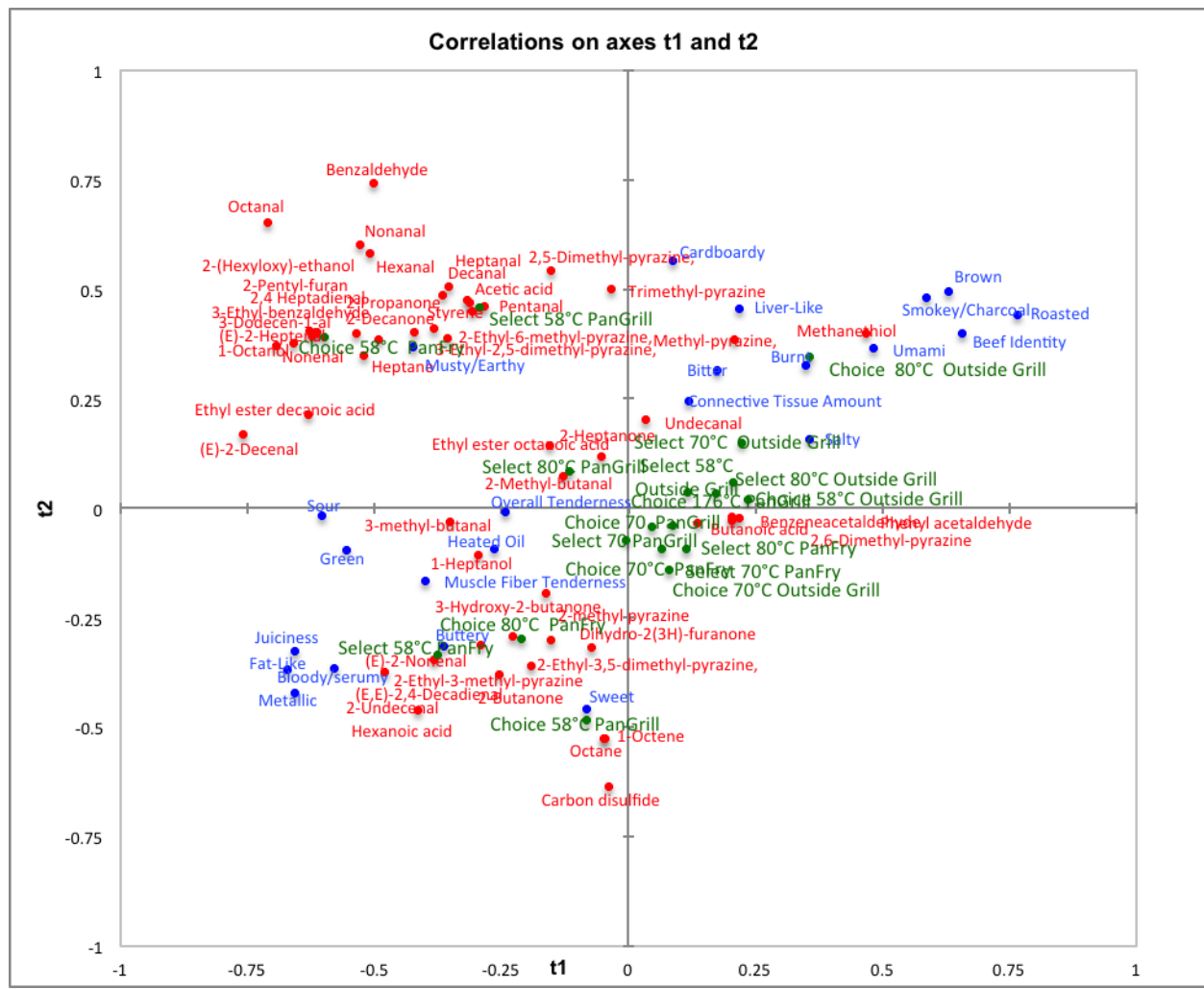


Figure 3. Inside skirt principal component analysis for descriptive sensory flavor attributes and the treatments.

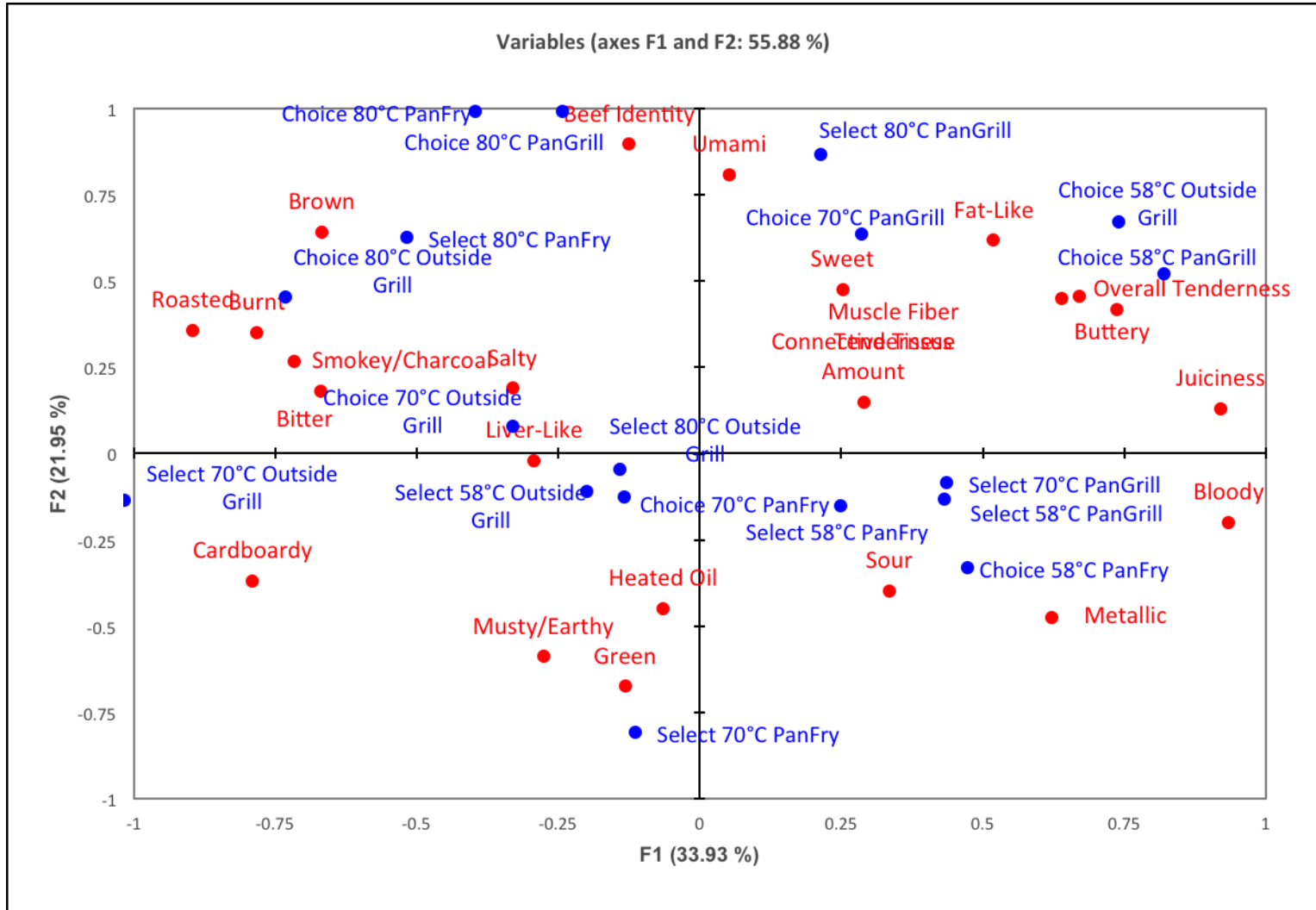


Figure 4. Inside skirt partial least squares regression biplot for trained descriptive flavor, volatile aromatic compounds, and steak treatments.

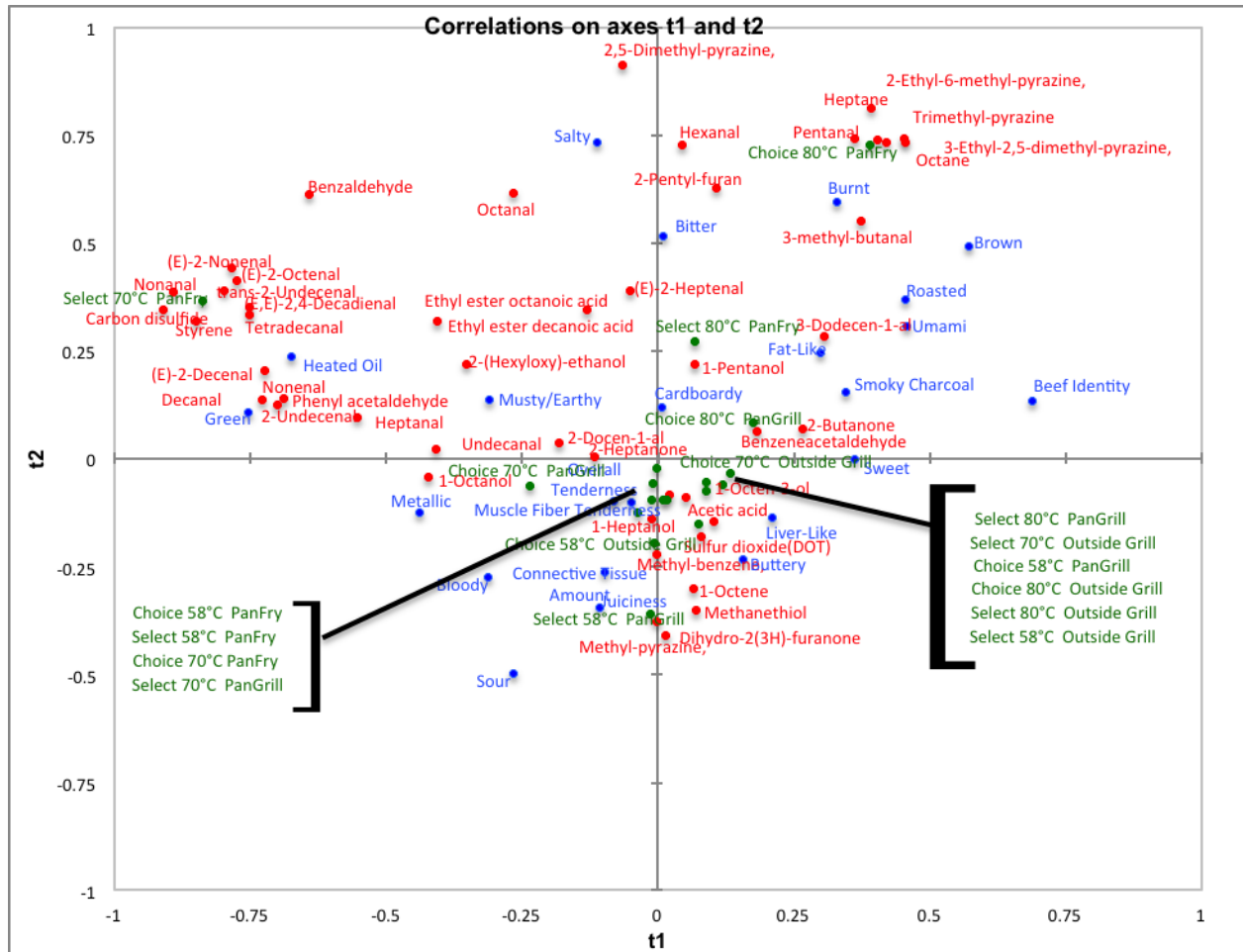


Figure 5. Outside skirt principal component analysis for descriptive sensory flavor attributes and the treatments.

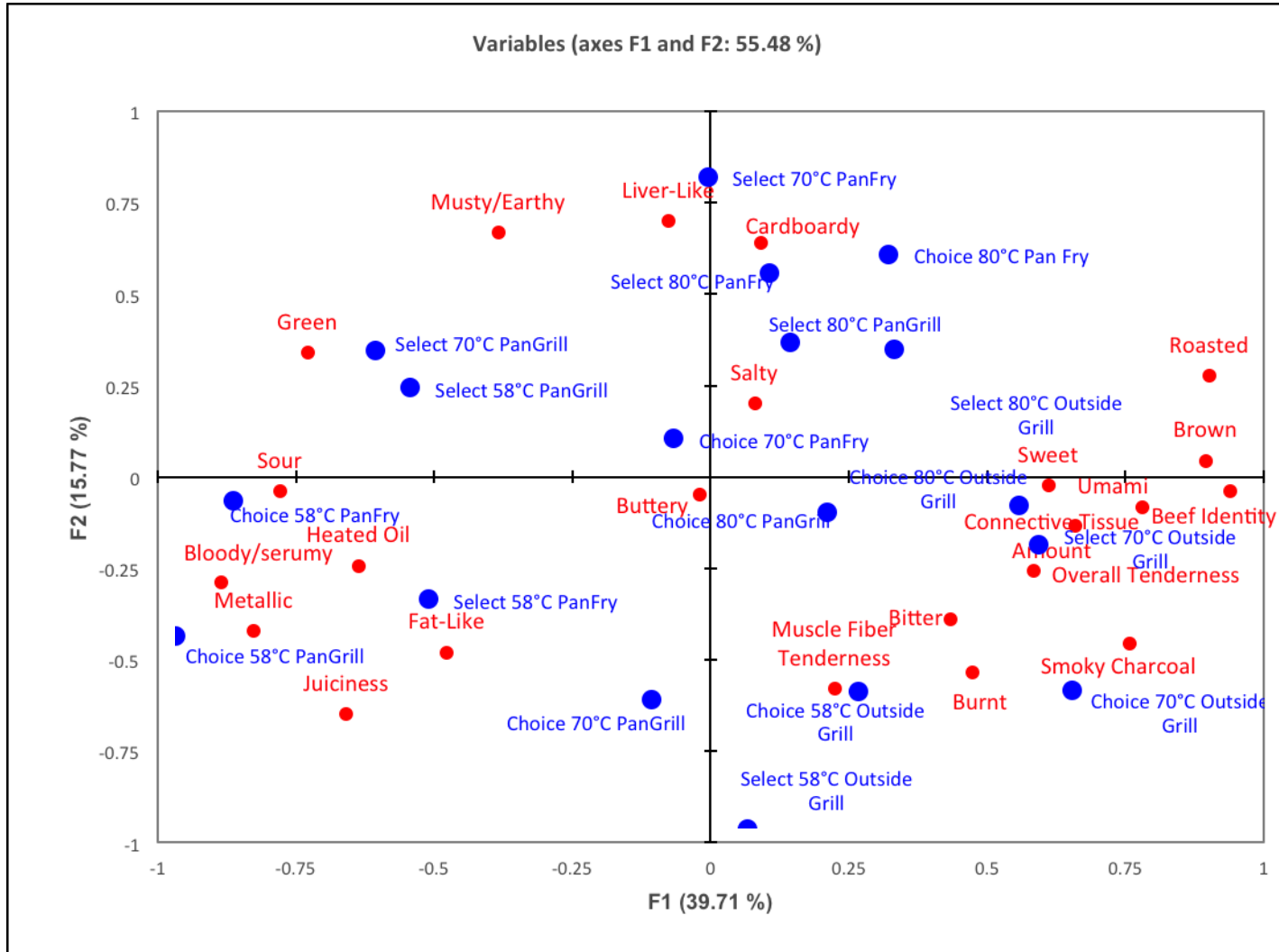
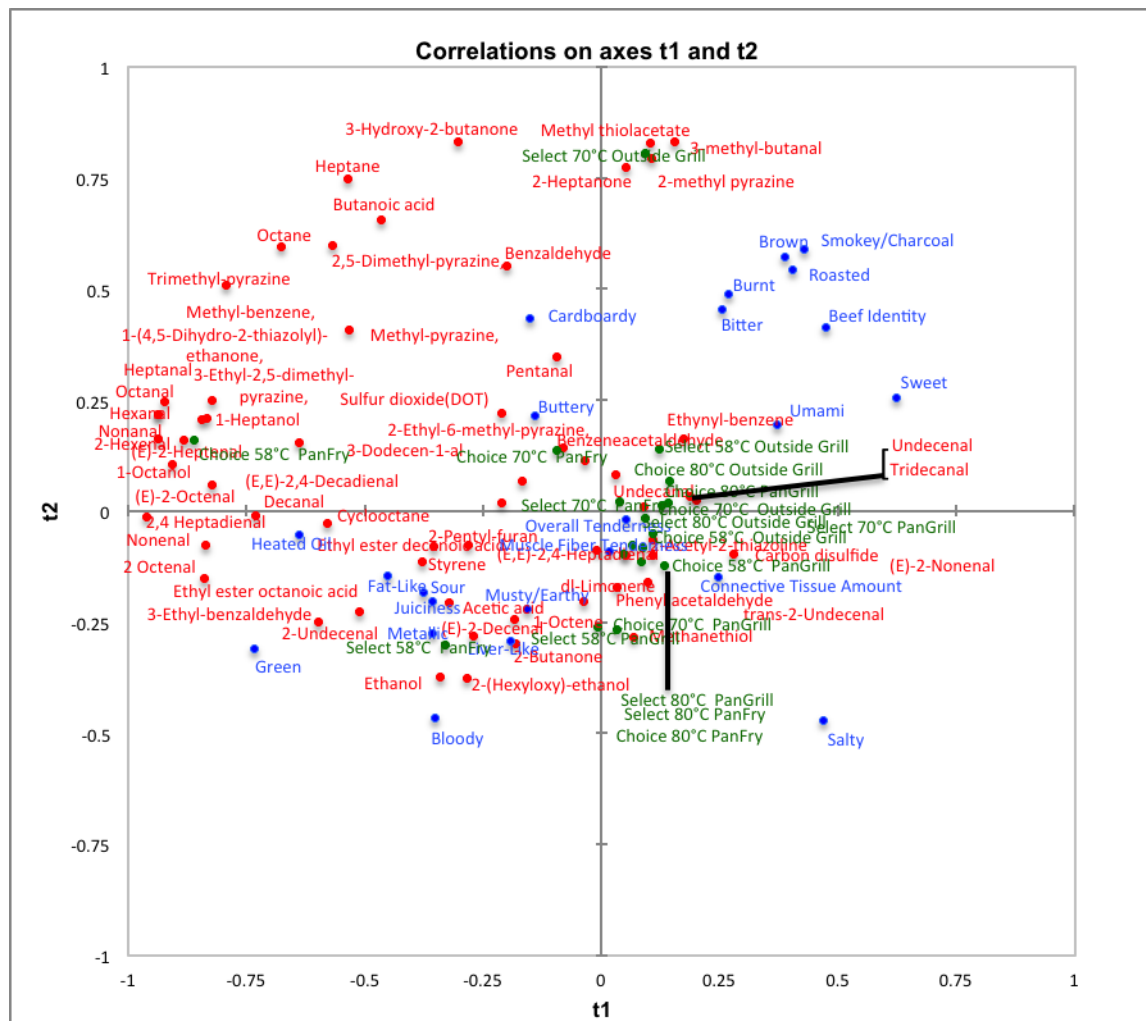


Figure 6. Outside skirt partial least squares regression biplot for trained descriptive flavor, volatile aromatic compounds, and steak treatments.



APPENDIX B
TRAINED PANEL BALLOT

	Beef Flavor ID	Brown	Roasted	Bloody Serumy	Fat-Like	Metallic	Liver-Like	Green	Umami	Sweet	Sour	Salty	Bitter	Cardboardy/oxidized	Burnt	Other Notes	Buttery	Heated Oil	Musty-Earthy/Humus	Smokey Charcoal	Tenderness	Juiciness	Muscle Fiber Tenderness	Connective Tissue	Overall Tenderness	
146																										
709																										
422																										
795																										
451																										
188																										
Break																										
702																										
203																										
821																										
362																										
043																										
033																										

APPENDIX C
TRAINING GUIDELINES

Day 1

- Introduce basic tastes, beef flavor ID, brown, roasted, and bloody/serummy.
- Sample evaluation for the introduced attributes
 - Select strip steak cooked to 65°C
 - Skirt steak cooked to 70°C
 - Beef brisket cooked to 70°C

Day 2

- Review previously introduced attributes
- Introduce metallic, fat-like, liver-like, and overall sweet
- Sample evaluation for all attributes
 - Choice strip steak cooked to 70°C
 - Prime beef ribeye cooked to 70°C
 - Select strip steak cooked to 51°C
 - Skirt steak cooked to 70°C

Day 3

- Review previously introduced attributes
- Introduce cocoa, burnt, green, and green-haylike
- Sample evaluation for all attributes
 - Grass fed strip steak cooked to 70°C
 - Skirt steak cooked to 70°C
 - Select strip steak cooked to 80°C
 - Choice beef ribeye cooked to 70°C

Day 4

- Review previously introduced attributes
- Introduce sour milk/sour dairy, sour aromatics, dairy, and cooked milk
- Sample evaluation for all attributes
 - Prime tenderloin cooked to 70°C
 - Organic strip steak cooked to 70°C
 - Select top sirloin cooked to 62°C
 - Skirt steak cooked to 58°C

Day 5

- Review previously introduced attributes
- Introduce cardboardy, leather, animal hair, and barnyard
- Sample evaluation for all attributes
 - Choice strip steak cooked to 58°C
 - Grass fed ribeye cooked to 70°C
 - Organic strip steak cooked to 70°C
 - Skirt steak cooked to 58°C
 - Prime tenderloin cooked to 70°C

Day 6

- Review previously introduced attributes
- Introduce chemical, rancid, spoiled putrid, and warmed over
- Sample evaluation for all attributes
 - Flap steak cooked to 58°C
 - Beef ribeye cooked to 70°C
 - Skirt steak cooked to 58°C
 - Select chuck eye steak cooked to 70°C
 - Prime tenderloin cooked to 58°C

Day 7

- Review previously introduced attributes
- Introduce musty-earthly/humus, oxidized, petroleum like, and fishy
- Sample evaluation for all attributes
 - Grass fed beef ribeye cooked to 70°C
 - Blade tenderized skirt steak cooked to 70°C
 - Skirt steak, soaked in fish oil for 15 minutes, cooked to 70°C
 - Grass fed top sirloin cooked to 70°C

Day 8

- Review previously introduced attributes
- Introduce refrigerator stale, heated oil, and buttery
- Sample evaluation for all attributes
 - Flap cooked on pan fry to 70°C
 - Skirt cooked on pan grill to 58°C
 - Skirt cooked on pan fry to 80°C
 - Ribeye cooked on pan grill to 70°C

Day 9

- Review previously introduced attributes
- Introduce painty, smokey wood, and smokey charcoal
- Sample evaluation for all attributes
 - Skirt cooked on pan fry to 80°C
 - Flap cooked on pan grill to 70°C
 - Ribeye cooked on pan fry to 58°C
 - Skirt cooked on pan grill to 70°C

Day 10

- Review previously introduced attributes
- Introduce juiciness, muscle fiber tenderness, connective tissue, and overall tenderness
- Sample evaluation for all attributes
 - Flap cooked on pan grill to 70°C
 - Choice inside skirt cooked on pan fry to 80°C
 - Ribeye cooked on pan grill to 58°C
 - Tenderloin cooked on pan fry to 70°C
 - Choice inside skirt cooked outside to 70°C
 - Flap cooked outside to 80°C
 - Select outside skirt cooked outside to 58°C