

EFFECTS OF POSTMORTEM pH MODIFICATION
AND OXYGEN SATURATION ON LEAN
COLOR CHARACTERISTICS
OF DARK CUTTING
BEEF

By

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FORMAT OF THESIS

This thesis is presented in the Journal of Animal Science style format, as outlined by the Oklahoma State University Graduate College style manual. The use of this format allows for independent chapters to be prepared suitable for submission to scientific journals.

CHAPTER I

INTRODUCTION

Consumers tend to associate the “cherry-red” lean color of beef with freshness and wholesomeness. Any deviation from this is believed to be associated with “unsafe” or “spoiled” beef. With this in mind, consumers are reluctant to purchase cuts from “dark cutting” beef. Hood and Riordan (1973) demonstrated that bright red beef out-sells discolored beef by a ratio of 2:1. Muscle can be naturally dark for a variety of reasons. Darker lean is typically produced in forage fed cattle as opposed to grain fed cattle, in cattle older than 4 years, in retail beef that has been on display too long, or in animals with dark cutting carcasses (Smith et al., 1993). These “dark cutting” carcasses are discounted at packing plants and sold as “non-conforming” beef. According to the 2000 National Beef Quality Audit (McKenna et al., 2002), discounts as high as \$240 per head are associated with dark cutting beef carcasses. During that audit, 2.3% of all steer and heifer carcasses were dark cutters, which represented in excess of 700,000 carcasses for the year 2000. Therefore, the beef industry lost roughly \$165 - \$170 million dollars that year on dark cutting carcasses alone.

Dark cutting beef is believed to be associated with long-term stress prior to harvesting of animals (Ashmore et al., 1973; McVeigh and Tarrant, 1982).

Some examples are stress factors associated with shipping, cattle held overnight at the packing plant in a strange, new environment, severe weather changes and mixing of unfamiliar cattle. Other factors shown to have a negative effect on the production of dark cutting beef carcasses are gender, implants, disposition, breed type and muscle fiber type. Any one of these stresses or factors alone can cause a dark cutting carcass. However, it is usually a combination of these that cause dark cutting beef. Stressors deplete muscle glycogen immediately prior to harvest. Once the animal is exsanguinated, the cells no longer have access to oxygen, therefore metabolism turns anaerobic. The lack of glycogen alters normal postmortem rigor mortis onset. This abnormality results in lean that is very dark, has a higher water holding capacity than normal beef due to densely packed muscle fibers and a very short retail shelf life, due to more free water available to bacteria (Dikeman, 1998). Light is not absorbed by the lean surface giving it the characteristic “dark” appearance. The surface also appears dry and is sticky to the touch.

Limited research has been conducted to improve the consumer appeal of dark cutting beef. Most of the research devoted to dark cutting beef has been aimed to develop methods and practices to prevent dark cutting beef prior to harvest. Improved management, transportation and facilities have all shown to help prevent dark cutting beef, however environmental factors have proven to be difficult to manage and control (Scanga et al., 1997). Thus, dark cutting beef has never been diminished from the United States. Therefore, some research should be aimed to improve the consumer appeal of dark cutting beef.

Based on this information, the objectives of this study were to assess the impact of high pressure oxygen saturation and enhancement of normal and dark cutting strip loins using a combination of acidic phosphates rather than the alkaline phosphates typically used in today's meat industry. Several subjective tests were conducted including retail shelf life, cooked color, odor and sensory evaluation. Several objective tests were conducted as well that included pH, L*a*b* color values, drip loss, headspace of modified atmosphere packages, total aerobic plate counts, oxidation using the Thiobarbituric Acid analysis and slice shear force.

CHAPTER II

REVIEW OF LITERATURE

Lean Color in Today's Meat Industry

Consumers make a purchase of retail meat determined on visual appraisal of meat color (MacKinney et al., 1966, Hedrick et al., 1994 and Kropf, 1980. The state of myoglobin in the muscle largely determines meat color. The purplish color of deoxymyoglobin is the reduced form of myoglobin. The brown color of metmyoglobin, which results from prolonged display in a retail case, is the oxidized form of myoglobin. The bright cherry red color of oxymyoglobin is myoglobin in the oxygenated state and this bright cherry red color in meat is recognized and demanded by the consumer (Munns and Burrell, 1966). Any deviation from this could cause the consumer to believe the meat is old or spoiled. Dikeman (1998) stated that if consumers see dark colored lean, they will associate it with meat from older cattle, poor flavor, poor keeping quality and toughness. Hood and Riordan (1973) showed that bright red beef out-sells discolored beef by a ratio of 2:1. Muscle can be naturally dark for a variety of reasons. Forage fed cattle can produce darker lean than grain fed cattle, older cattle (> 4 years) produce darker lean, retail beef that has been on display too long or because the animal is a dark cutting carcass (Smith et al., 1993).

Definition of Dark Cutting Beef

The beef quality defect known as “dark-cutting” (also known as high pH beef or dark, firm and dry beef) refers to the color of lean after a cross-section has been cut and exposed to air for an adequate period of time. It is termed “dark-cutting” because normal beef muscle will “bloom” (turn from purple to bright cherry red as myoglobin becomes oxygenated on the lean cut surface), whereas, lean from a dark cutting carcass will remain dark after exposure to oxygen from a failure of the myoglobin on the surface to become oxygenated (Smith et al., 1993).

Causes of Dark Cutting Beef

Hedrick et al., (1959) discovered that dark cutting beef occurs when glycogen supplies in the lean tissue are abnormally low prior to harvest. However, in a summary by Hall et al. (1944), it was reported that in 1877, Claude Bernard was probably the first to report that exhausted animals frequently exhibited an alkaline rigor, and scientists concluded that the elevated pH associated with dark cutting meat coincided to the reduction of muscle glycogen content prior to harvest. In unstressed, well-fed cattle, the concentration of muscle glycogen will be 0.8% to 1.0% of the muscle weight (Dikeman, 1998). Runnion et al. (1939) and Munns and Burrell (1966) found that dark cutting was more prevalent in the lower grades. Better-finished cattle usually have higher tissue glycogen levels.

Normal muscle pH will be above 7.0 in a living animal (Dikeman, 1998). After the animal is exsanguinated, muscle metabolism shifts from aerobic to

anaerobic (Berg, 2001). According to Mayes (1993), when muscle contracts, lactic acid results as the principal end product of glycolysis in an anaerobic environment as opposed to pyruvate in an aerobic environment. Lactic acid does not accumulate in an aerobic environment, because either the circulatory system removes it for disposal by the liver or it is oxidized to CO₂ and water. During anaerobic glycolysis, one molecule of glucose will generate 3 moles of adenosine triphosphate (ATP). ATP may be maintained for several hours during anaerobic glycolysis, however creatine phosphate is rapidly depleted. As glycogen is depleted, energy for the regeneration of ATP must come from secondary energy sources by β -oxidation of adipose tissue or from gluconeogenic amino acids resulting in proteolysis and muscle atrophy. The conversion of glycogen to lactic acid will lower the muscle pH until the glycogen is used up or the contractile proteins stop functioning as a result of low intramuscular pH. Accumulation of lactic acid and the lowered pH converts muscle to meat (Berg, 2001).

Because of the low glycogen amounts prior to harvest, the lean tissue is higher in pH than normal beef post-rigor (Hedrick et al., 1959). Dikeman (1998) stated that glycogen levels under 0.6% of the body weight will likely result in incomplete acidification. Dikeman (1998) also stated that a post-rigor pH of 6.0% or greater will result in some degree of dark cutting. According to Page (2001), a pH value of 5.87 was found to be the cutoff between dark cutters and non-dark cutters. Munns and Burrell (1965) reported that 90% of dark cutters have an ultimate pH of 6.0 or higher in the longissimus dorsi muscle.

Several explanations for the dark appearance of the lean surface of dark-cutting beef have been given. According to Ashmore et al. (1973), when pH remains high in the muscle of carcasses, the mitochondrial respiration also remains high. Myoglobin is deoxygenated, and the dark red color associated with dark cutting beef occurs. According to Lawrie (1952), at higher pH values, oxygen utilization by the surviving enzyme systems is much greater than in normal beef. Therefore, myoglobin is only partially oxygenated or remains in the reduced state, resulting in dark cutting beef. In normal beef, mitochondria are inactivated with a low ultimate muscle pH (Ashmore et al., 1972). The lower pH value permits oxygenation of myoglobin to produce oxymyoglobin, which produces the appealing cherry-red color of fresh beef (Munns and Burrell, 1966).

According to Anon. (1944), the permeability of lean tissue to oxygen is reduced at higher pH values. Ledward et al. (1992) concluded that when muscle pH is higher than normal, proteins will bind more strongly with water, allowing less available free water. When the muscle proteins bind more water, the fibers become swollen, leaving less space between them. Thus, there will be less reflectance of light because there is less free water to reflect light, causing a dark appearance. Ultimately, however, dark-cutting beef will not bloom when exposed to air, and is therefore discounted at the retail level (Price and Schweigert, 1987).

As previously mentioned, the cause of dark cutting beef results from the depletion of muscle glycogen prior to harvest. This depletion of muscle glycogen results from a single stress or a combination of several stressors on the cattle before they are harvested. Fraser et al. (1975) proposed a definition of stress:

“An animal is said to be in a state of stress if it is required to make abnormal or extreme adjustments in its physiology or behavior in order to cope with adverse aspects of its environment and management. A husbandry system can be said to be stressful if it makes abnormal or extreme demands on the animals. Finally, an individual factor may be called a stressor if it contributes to the stressful nature of a system of husbandry.”

According to Scanga et al. (1997), the three main types of stress in beef animals are environmental, physiological and managerial. Dikeman (1998) included stress-induced adrenaline secretion (psychological). Grandin (1993) came up with an idea that she coined the “gas tank” theory. In this theory, the “gas tank” is the ability of a beef animal to cope with stress, the gas being the animal’s muscle glycogen. When a single stress occurs, some glycogen is burned. When numerous stresses occur, and the “gas tank” empties, it causes the animal to be at a greater risk to become a dark cutter at harvest if harvest does occur before the animal naturally restores the muscle glycogen to a normal level. According to Berg (2001), the physiological response of an animal to stress is controlled by the autonomic nervous system; the brain sends messages through the efferent nerves to produce a response. The nerves carry the messages to muscles, capillaries, and the heart, which are involuntary or ‘automatic’. The autonomic nervous system can be broken down into two parts: the sympathetic and parasympathetic. These two systems are responsible for the response and recovery from a stressor. The sympathetic nervous system controls alertness, arousal, activation and mobilization and is more commonly

known to control the physiological responses fight, fright and flight. In response to a stressor and the subsequent activation of the sympathetic nervous system, the adrenal medulla releases epinephrine and norepinephrine (catecholamines), which stimulates a series of reactions to rapidly generate energy while inhibiting processes such as energy storage, digestion, and immune function. Epinephrine stimulates glycogenolysis and lipolysis, therefore mobilizing glucose from glycogen stores in the liver and muscle. Fatty acids are also released from fat cells. According to Tarrant (1989), glycogen phosphorylase regulates glycogenolysis in skeletal muscle. Either or both increased catecholamine levels or muscle contraction can trigger the activation of glycogen phosphorylase. Epinephrine increases heart rate and blood pressure and dilates blood vessels to increase blood flow to the extremities for the better delivery of metabolites to the muscle to quicken the escape from the stressor. The increased blood flow also removes the metabolic waste product lactic acid, which causes fatigue, and also provides a method for heat dissipation produced by the increased muscle metabolism. The release of norepinephrine will also stimulate lipolysis, however it does not affect glycogenolysis. Opposite of epinephrine, norepinephrine will stimulate constriction of blood vessels preventing heat dissipation. The elevated muscle temperature increases the rate of shortening, contractile force, maximum tension and Ca^{2+} sensitivity of the contractile proteins, allowing the muscle to maximize muscle efficiency. The parasympathetic nervous system controls calm activities and promotes growth, energy storage, digestion, absorption and tissue repair. Therefore, the two systems work in balance (Berg 2001).

Breed type and genetics have been found to have an effect on an animal's ability to be a dark cutter as well. Shackelford et al. (1994) reported that genetic variation in the incidence of dark, firm and dry beef existed, but was small in comparison to environmental variation. The frequency and severity of dark cutting carcasses in the 1990 NBQA was dependent on breed type; 9.7 percent of dairy carcasses were dark cutters (Lorenzen et al., 1992). Tyler et al. (1982) reported that Brahman crosses were less susceptible to stress than Hereford and Shorthorn steers in Australian production systems. Lorenzen et al. (1992) also reported a slightly lower incidence of dark cutting beef in *Bos indicus* than in *Bos taurus* carcasses. According to Voisinet et al. (1997), the excitability of cattle has a direct effect on the incidence of borderline dark cutting beef, the more excitable an animal was, the greater the tendency was to exhibit dark cutting properties at slaughter. They also concluded that animals that were more excitable tended to exhibit tougher lean. According to Le Neindre et al. (1995); Hearnshaw et al. (1979); Fordyce et al. (1988); Stricklin et al. (1980); and Tulloh (1961), temperament in cattle is a heritable trait between and within cattle breeds that may affect the animal's reaction to handling. Genetics also affect an animal's response to stress. According to Zavy et al. (1992), Brahman-cross cattle had higher cortisol levels when restrained in a squeeze chute compared to English-cross cattle. Shackelford et al. (1994) concluded that genetic variations have an effect on cattle to produce dark cutting beef. Lean color and texture are lowly heritable traits; therefore, rapid improvement for those traits is difficult. Several researchers have shown that meat tenderness is correlated with ultimate muscle

pH (Purchas, 1990; Watanabe et al., 1996) and muscle color (Jeremiah et al., 1991; Wulf et al., 1997).

Several studies have shown that muscle fiber types have effects on the capability of cattle to become dark-cutting carcasses. According to Berg (2001), muscle fibers that have a slow *m*ATPase activity, are oxidative and make up slow, fatigue resistant motor units are referred to as Type I muscle fibers. These fibers have greater aerobic endurance, possess a greater amount of myoglobin and a higher concentration of mitochondria. Muscle fibers that possess a fast *m*ATPase activity, a low oxidative capacity and high glycolytic metabolism are Type II fibers. Type II fibers are more suited for short bursts of power, higher in glycogen content, lower in mitochondria and myoglobin. Type II fibers can be separated based on metabolic function. Type IIB fibers make up fast-fatigue motor units associated with the great bursts of strength. Type IIA fibers are distinguished from IIB because they have a higher aerobic capacity and make up fast, fatigue resistant motor units that are capable of greater endurance. Schiaffino et al. (1989) identified another fast twitch fiber type IIX (also IID) that possesses an aerobic oxidative enzymatic activity that is between types IIB and IIA. The physiology of these different fiber types will affect the length of postmortem biochemical activity (Berg 2001). According to Pette and Staron (1990), types IIB and IID are more stable under acid conditions. Because fast muscle fibers have a glycolytic metabolism that supports rapid and strong bursts of power, they will propel glycogenolysis to the formation of lactic acid longer in postmortem muscle. McVeigh and Tarrant (1983) suggested that muscle

contraction might be the primary mechanism responsible for antemortem glycogenolysis in cattle. The Type I slow twitch fibers are found in a high amount in motor units capable of sustained use, such as posture related muscles. These fibers are capable of maintaining a strong *m*ATPase activity at low pH levels. However, glycogen is depleted rapidly and postmortem activity cannot be maintained due to the lack of glucose for conversion to ATP. Hydrolysis of ATP by *m*ATPase is necessary for myosin to bind and to release from the myosin-binding site on actin. Without the presence of ATP, myosin can neither bind to actin to generate a 'power stroke', nor can myosin release from actin once bound. Therefore, the depletion of ATP stores within postmortem muscle is the first step in forming the permanent acto-myosin or 'rigor bond' and significantly influences meat tenderness and water holding capacity (Berg, 2001). Young & Foote (1984) reported that beef muscles that are low in glycolytic capacity are less prone to the dark-cutting condition, although all muscles have the potential to attain an abnormally high ultimate pH as the work with adrenaline by Tarrant & Sherington (1980) showed. The findings by Lacourt & Tarrant (1985) support the findings by showing that the response to stress by fiber types depends on the nature of the stressor. If the antemortem stress relates to physical, muscular work, those muscles will be depleted of glycogen. Glycogen depletion will tend to be greater in the fast-twitch fibers versus the slow-twitch fibers. Emotional or psychological stress prior to harvest will result in the sympathetic arousal and subsequent adrenaline release. According to Tarrant & Sherington (1980), there is a general glycogenolytic response throughout the skeletal musculature as

muscles are exposed to circulating hormones. However, the response may be the greatest in those muscles with the highest proportion of slow fibers. Lacourt & Tarrant (1985), therefore concluded that muscles that are high in fast-twitch (glycolytic) fibers are more apt to become dark-cutting carcasses when the stress is mostly physical in nature, whereas the muscles that are high in slow-twitch (oxidative) fibers are more likely to produce adrenaline-mediated dark-cutting carcasses. Zerouala and Strickland (1991) reported that there were more Type 1 muscle fibers in dark cutting bulls and steers than in normal bulls. Dark cutting bulls and steers contained less fast, glycolytic (a White) muscle fibers than normal bulls. The results showed that the combination of slow oxidative and fast, oxidative, glycolytic (a Red) fibers in dark cutting bulls and steers exhibited more oxidative metabolism in the longissimus dorsi muscle than in normal bulls. According to Young & Foote (1984), cattle of Continental European influence have a higher percentage of white muscle fibers than cattle of British influence. According to Smith et al. (1993), because of the higher amount of white muscle fibers in Continentally influenced cattle, they are more susceptible to become dark-cutting carcasses. The findings of Zerouala & Strickland (1991) support the previous statement. They concluded that bulls and steers that have muscles with high proportions of white fibers versus red fibers produce more dark-cutters. The usage of Continental breeds in the U.S. cattle population has increased greatly. According to Mies (1992), 10 to 15 percent of the U.S. steers/heifer population was purebreds or crossbreds of Continental European breeds in the mid-1980's. In 1992 that number had risen dramatically to over 50 percent Continental

influence. According to Smith et al. (1993), that increase could contribute to the perceived increase in the number of dark-cutters.

According to Grandin (1992), the national percentage of dark cutters in 1982 was approximately 0.5%. That number increased to 1.0% in 1992. She cites two major factors that account for a large increase in dark cutters. The use of trenbolone acetate implants and using more exotic cattle than in the past are directly correlated with the increase. They do not directly cause dark cutters, however they make the animals more susceptible to stress. Trenbolone acetate is a synthetic male hormone that increases muscle mass and makes cattle grow faster. Misusing trenbolone acetate will reduce the quality grade and increase the percentage of dark cutters.

Grandin (1992) stated that holding cattle over the weekend without feeding as well as mixing strange cattle at the abattoir would increase the number of dark cutting carcasses. Warriss (1990) reported that transporting cattle to the packing plants was very stressful on cattle in that they are being removed from their home environment, being loaded and unloaded onto trucks, and transported. During this time they may be exposed to various stressors. These stresses may include all or some such as noise, unfamiliar odors, vibration and changes of acceleration, deprivation of food and water, breakdown of social groups, as well as close confinement or overcrowding.

According to Tarrant (1981), the gender of an animal affects its ability to become a dark-cutting carcass. Estimates from a survey from 19 countries revealed that the occurrence was 1 to 5 percent in steers and heifers, 6 to 10

percent in cows and 11 to 15 percent in bulls. According to Warriss et al. (1984) and Kenny & Tarrant (1987), mixing of unfamiliar bulls prior to harvest results in increased numbers of dark-cutting carcasses due to the physical activity, particularly mounting of the bulls when social structure is broken. According to Stabenfeldt & Edqvist (1984), heifers experiencing oestrus immediately prior to harvest show behaviors such as restlessness, increased alertness, increased interest in other animals, mounting other animals, standing for others to mount and decreased appetite. Hedrick (1981) suggested that the physical activity and stress associated with oestrus contribute to the incidence of dark-cutting beef. However, this is prevented by the feeding of melengestrol acetate (MGA) to suppress estrus.

Occurrences of Dark Cutting Beef

Many independent studies have been conducted trying to find the incidence or occurrence of dark cutting beef. Munns and Burrell (1966) reported that the incidence of dark cutters in Canada during 1958-1961 was 8 percent. From 1986 to 1992 in Canada, Jones (1992) indicated that the incidence of dark cutting beef occurrence in steers and heifers ranged from 2.0 to 2.2 percent. He also reported that dark cutting occurrence was highest in bulls, intermediate in heifers and lowest in steers. He also reported that dark cutting carcasses were monitored precisely because they were given a unique quality grade (Canada B4). Canadian beef are mostly sold on a quality and yield basis, therefore affecting the producer directly by incidence of dark cutting carcasses (Smith, 1993). Jones and Tong (1989) reported that Canadian producers lose

approximately \$5 million annually from discounted dark cutting beef carcasses. Janloo et al. (1998) reported that from 1,129 steers observed, 2.8 percent were dark cutters with 0.7 percent of those classified as full dark cutters. Tarrant (1981) reported that a survey of scientists in 19 countries estimated that dark cutting beef in steers and heifers was 1 to 4 percent, 6 to 10 percent in cows and 11 to 15 percent in young bulls. Oltjen (1982) concluded from a European Symposium in 1980 that dark cutting beef among steers and heifers in developed countries was 1 to 5 percent.

In 1990, the program known as the National Beef Quality Audit (NBQA) was started by the National Cattlemen's Beef Association to find and address the problems, defects and inconsistencies of the beef industry. The first audit was conducted in 1990 and audits are completed every five years. The 1990 NBQA (Smith et al., 1992) concluded that 5.0 percent of all carcasses surveyed were dark cutters and of that 5.0 percent, 3.4 percent were discounted one-third of a quality grade, 1.1 percent were discounted two thirds of a quality grade and 0.5 percent were discounted one full quality grade. According to the 1995 NBQA, 2.7% of the carcasses audited were classified as dark cutters to some degree (Boleman et al., 1998). This resulted in a \$6.08 loss per steer/heifer harvested that year and ranked fifth in severe problems among packers (Smith et al., 1995). That percentage decreased to 2.3% of the carcasses audited during the 2000 NBQA, which represented in excess of 700,000 carcasses for 2000 (McKenna et al., 2002). According to the 2000 NBQA (McKenna et al., 2002), discounts as high as \$240 per carcass are associated with DC beef and it cost the industry an

estimated \$5.81 per head. The 1999 Canadian beef audit concluded that 1.2% of the carcasses examined were dark cutters; 1.0% of steers, 0.5% of heifers, 3.0% of cows and 14.0% of bulls were dark cutters (Donkersgoed).

Smith et al. (1993) concluded that the occurrence of dark cutting beef is most likely to occur during temperature extremes and especially during large fluctuations in temperature over very short time periods. Precipitation also has a major effect by increasing the rate of body-heat loss and therefore causes physiological stress by causing the animal to shiver. Grandin (1992) reported that during periods of temperature fluctuations, dark-cutting can occur up to 8% in feedlot cattle. This type of weather mostly occurs in the spring and fall. Tarrant and Sherington (1980) also found a relationship between the occurrence of dark cutting beef and season. Munns and Burrell (1966) also found the incidence of dark cutting beef to show a strong seasonal trend. They found the highest occurrence was encountered in the fall with a rise in the spring as well.

Effects on Dark Cutting Beef

Research has shown that beef derived from dark cutting carcasses not only has poor appearance, but also has less consistency and less desirable sensory characteristics (Tarrant, 1981). According to Lacourt & Tarrant (1985), water-holding capacity, tenderness, as well as keeping quality are all influenced a great deal by the rate and extent of pH decline after harvest. Dark cutting beef has a decreased shelf life (Vanderzant et al., 1983). According to Bem et al. (1976), dark cutting meat is more susceptible to bacterial spoilage than normal meat. Dark cutting beef allows an environment more favorable for spoilage

bacteria to grow due to the elevated pH associated with it (Gill and Newton, 1979). According to Gill and Newton (1981), glucose is utilized first by most bacteria found in meat. During aerobic storage, the spoilage of meat occurs when glucose is used up and amino acids are subsequently attacked. In dark cutting meat, the amino acids are attacked immediately, and spoilage odors can be detected at lower cell densities. The bacterial flora of vacuum-packaged meat is comprised of facultative anaerobes, mostly lactic acid bacteria and others such as *Brochothrix thermosphacta* and *Enterobacteriaceae*. The spoilage of normal vacuum-sealed meat results from the accumulation of the products of fermentation by these bacteria. The high pH of dark cutting meat allows the development of *Serratia liquefaciens* and *Alteromonas putrefaciens*. *S. liquefaciens* produces spoilage odors at low cell densities on dark cutting meat by degrading amino acids. *A. putrefaciens* can cause green discoloration on the surface of meat by producing hydrogen sulfide, which binds with myoglobin to produce sulfmyoglobin (Gill and Newton, 1979).

Dark-cutting beef results in reduced carcass value because of reduced quality grade (USDA, 1989). USDA graders evaluate muscle color for indications of carcass maturity and muscle pH when assigning overall quality grades (USDA 1997). USDA graders can discount the quality grades for dark cutters by 1/3 of a grade for a carcass classified as 1/3 dark, 2/3 of a grade for a carcass classified as 2/3 dark, and a full grade for a carcass classified as fully dark. However, many packers can usually merchandise the 1/3 dark cutting carcasses through normal marketing channels, and therefore only report those in which cannot be

channeled through the normal markets (Smith et al., 1993). Beef processors have to discount dark cutting carcasses 20% to 40% to market them (Dikeman, 1998). Most of this product goes to the hotel and restaurant trade rather than retail because the product has already been cooked when the consumer first sees it. Dark cutting beef cost the beef industry approximately \$132.5 million in 1991, or approximately \$5 for every steer and heifer slaughtered (Smith et al., 1992). According to Dikeman (1998), producers absorb the losses from dark cutting cattle if the cattle are sold on a grid system and packers absorb the losses when the cattle are sold on a live basis.

Prevention of Dark Cutting Beef

Much research has been conducted to prevent antemortem stress in beef animals; however, stress cannot be completely eliminated prior to harvest. Environmental factors can be measured, however, they are unmanageable and therefore, are uncontrollable (Scanga et al., 1997). Dark cutting beef has never been diminished from the U.S. from an economic standpoint. Improved management, transportation and packing plant facilities can be improved. However, the environmental effect cannot be controlled without the construction of enclosed feedlots, which is not economically feasible. Ashmore et al. (1973) showed that prevention of dark cutting meat could be achieved by injecting live animals with propranolol, a beta-adrenergic blockade agent as well as a competitive inhibitor of epinephrine, to prevent antemortem depletion of muscle glycogen.

Several treatments have been used to negate some of the physiological responses associated with transport and handling stress including the use of preconditioning regimens (Woods et al., 1973; Cole, 1988), vitamin treatments (Cole et al., 1979), vaccines (Johnson et al., 1988), and feeding of fats (Cole and Hutcheson, 1987). Hutcheson and Cole (1986) and Matter et al. (1986) attempted to remedy hypoglycemia through electrolyte nutritional treatment and high-energy diets either before or after transport. This has been shown to be effective for controlling meat quality defects (Wajda and Wichlacz, 1987; Eldridge, 1988; Lister, 1988; Tarrant, 1988). However, according to McVeigh and Tarrant (1982) and Lister (1988), it takes several days for animals to recover fully using these methods. Therefore, these are not feasible methods for animals being transported to abattoirs. According to Smith et al. (1993), sufficient rest and adequate feed intake is needed to replenish glycogen stores in muscle. McVeigh et al. (1979) reported that the recovery of glycogen in muscle is a slow process, requiring 7 days to return to normal concentration. In epinephrine-induced depletion of glycogen, the animal requires 11 to 14 days to fully restore initial glycogen level (McVeigh & Tarrant (1981).

Improving Lean Color of Dark Cutting Beef

Limited research has been done after an animal has become a dark-cutting carcass to increase consumer appeal. Cornforth and Egbert (1985) experimented successfully the capability of pre-rigor muscle to obtain bloom when blended with rotenone, a mitochondrial respiratory inhibitor, or when thin slices of pre-rigor muscle were chilled in an oxygen rich atmosphere (Cornforth et

al., 1985). However, rotenone is an insecticide and is therefore unsafe for human consumption. Dark cutting meat is similar to pre-rigor meat because not enough acid is produced during postmortem glycolysis to inactivate the mitochondria. As long as the mitochondria is active and respiration continues, myoglobin will be deoxygenated and the muscle will remain dark Egbert and Cornforth (1986).

CHAPTER III

EFFECTS OF POSTMORTEM pH MODIFICATION AND OXYGEN SATURATION ON LEAN COLOR CHARACTERISTICS OF DARK CUTTING BEEF

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ABSTRACT

Paired strip loins (n = 32) were obtained from four carcasses from each of the following groups: normal, 1/3, 2/3, and full dark cutting carcasses. 2 pairs from each group were designated for a 7 d postmortem aging period and the other 2 pairs were aged 14 d. After each of the respective aging periods, 1 strip from each pair was randomly selected and enhanced to 110% of original weight using water, an acidic phosphate blend, NaCl and Herbalox™. Upon enhancement, 2.54 cm steaks (n = 6) were fabricated from each of the enhanced and non-enhanced loins. These steaks were then subjected to a 12 hr, high-pressure (240 psi) oxygenation cycle. After the oxygenation was completed, an additional 6 steaks were cut from the enhanced and non-enhanced loins for a non-oxygenation treatment. The steaks were used for the following tests: lean color assessment, odor assessment, cooked internal lean color, thiobarbituric acid analysis (TBA), total aerobic plate counts, sensory panel, slice shear force, pH, drip loss and package headspace composition. Steaks were packaged in a

modified atmosphere consisting of 80% oxygen and 20% carbon dioxide. Steaks for lean color assessment were subjected to a 14 d retail display period, whereas steaks assessed for cooked lean color, sensory panel, slice shear force and odor panel were subjected to a 7 d retail display period. High-pressure oxygenation was found to improve lean color characteristics of strip steaks derived from dark cutting carcasses while not having any detrimental effects on oxidative rancidity. Enhancement was not found to improve lean color. Enhancement did improve juiciness, tenderness and connective tissue amounts when presented before a trained sensory panel, however, uncharacteristic flavor development was a downside of the enhancement.

INTRODUCTION

Consumers tend to associate the “cherry-red” color of beef with freshness and wholesomeness. Any deviation from this is believed to be associated with “unsafe” or “spoiled” beef. With this in mind, consumers are reluctant to purchase cuts from “dark cutting” (DC) beef. These DC carcasses are discounted at the packing plant and sold as “non-conforming” beef. According to the 2000 National Beef Quality Audit (McKenna et al., 2002), 2.3% of all steer and heifer carcasses were dark cutters, which represented in excess of 700,000 carcasses for the year 2000. Therefore, the beef industry lost roughly 165 – 170 million dollars that year on DC carcasses alone.

Dark cutting beef is believed to be associated with long-term stress prior to harvesting of the animal (Ashmore et al., 1973; McVeigh et al., 1982). Some

examples are stress factors associated with shipping, cattle held overnight at the packing plant in a strange, new environment, severe weather changes and mixing of unfamiliar cattle. Other factors shown to have a negative effect on the production of dark cutting beef carcasses are gender, implants, disposition, breed type and muscle fiber type. Any one of these stresses or factors alone can cause a dark cutting carcass. However, it is usually a combination of these that cause dark cutting beef. Stressors deplete muscle glycogen immediately prior to harvest. Once the animal is exsanguinated, the cells no longer have access to oxygen, therefore metabolism turns anaerobic. The lack of glycogen alters normal postmortem rigor mortis onset. This abnormality results in lean that is very dark, has a higher water holding capacity than normal beef due to densely packed muscle fibers and a very short retail shelf life, due to more free water available to bacteria (Dikeman, 1998). Light is not absorbed by the lean surface giving it the characteristic “dark” appearance. The surface also appears dry and is sticky to the touch.

The objectives of this study were to assess the impact of high pressure oxygen saturation and enhancement of normal and dark cutting strip loins using a combination of acidic phosphates rather than the alkaline phosphates typically used in today’s meat industry. Several subjective tests were conducted including retail shelf life, cooked color, odor and sensory. Several objective tests were conducted as well that included pH, L*a*b* color values, drip loss, headspace of modified atmosphere packages, total aerobic plate counts, oxidation using the Thiobarbituric Acid analysis and slice shear force.

MATERIALS AND METHODS

Meat Samples

At the National Beef processing facility in Liberal, KS, 16 pairs (n = 32) of strip loins were selected based on pH, objective color scores (L*a*b*) and USDA Grader's quality grade or lean color evaluation (non-DC, 1/3, 2/3 or full DC). The 16 pairs consisted of 4 pairs each of 1/3, 2/3, full DC and normal A-lean maturity beef. After selection, strip loins were shipped to the Oklahoma State University (OSU) Meat Laboratory. Upon arrival at OSU, 2 pairs from each lean quality group were designated into a 7d postmortem aging period and the remaining (n =2) pairs from each group were designated into a 14 d postmortem aging period. On d 6 of the 7 d postmortem aging period, one strip loin from each pair was randomly enhanced to 110% of its original weight with a brine solution containing 91.7% H₂O, 4.2% acidic phosphate blend, 3.0% NaCl and 1.2% Herbalox™ as an antioxidant. The loins were enhanced using a MetalQuimia™ high-pressure brine injector. The final pH of the brine was 4.92. Once the strip loins were allowed to equilibrate for 3 hr, 2.54 cm steaks (n=6) were cut from both the enhanced and non-enhanced strip loins from each pair. These steaks were then subjected to 240 psi of pure oxygen for a 12-hour cycle in a Vivotec_{GmbH} MD003. The remaining uncut portions of the strip loins were repackaged. On d 7, 2.54 cm steaks (n=6) were fabricated from the remaining portions of both the enhanced and non-enhanced loins. The oxygenated steaks were also removed from the Vivotec. This resulted in 4 treatments: Oxygenated/Enhanced (OE), Oxygenated/Non-Enhanced (ON), Non-

Oxygenated/Enhanced (NE) and Non-Oxygenated/Non-Enhanced (NN). Steak 1 from each treatment was used for day 1 oxidative rancidity testing using the thiobarbituric acid analysis (TBA), total aerobic plate count (TPC), objective color scores ($L^*a^*b^*$), pH, drip loss, and oxygen penetration (Appendix A). Steaks 2 through 6 from each treatment were packaged in a Modified Atmosphere Packaging (MAP) system utilizing Rock-Tenn DuraFresh™ rigid # 10 trays sealed with clear, multi-layer barrier film (LID 1050 film, Cryovac Sealed Air, Duncan SC) in a Mondini semi-automatic tray-sealing machine (Model CV/VG-5, G. Mondini S.P.A. Cologne, Italy). The gas used for the atmosphere consisted of 80% O₂ and 20% CO₂. Steaks 2 - 5 were then subjected to fluorescent lighting in a cold room (avg. 40 ° F) to simulate retail display for 7 d. Following the 7 d display period, steaks were removed from the display cooler. Steak 2 was utilized for day 7 TBA, TPC, $L^*a^*b^*$, pH and headspace composition determination. Steak 3 was used for a subjective cooked color panel, cooked $L^*a^*b^*$, cooked pH and a subjective odor panel. Steaks 4 and 5 were removed from the MAP trays, vacuum packaged and frozen for sensory panel and slice shear force, respectively. Steak 6 was left in the simulated retail storage for an additional 7 d. During the 14 d period, a subjective panel evaluated the steaks daily for lean color, fat color, percent discoloration and overall acceptability. Upon completion of the 14d period, the steaks were removed from the case and used for day 14 TBA, TPC, $L^*a^*b^*$, pH and headspace. On d 13 of the 14 d aging period, the above-mentioned procedures were repeated for the remaining 2 pairs of strip loins from each group (1/3, 2/3, full DC and normal beef).

However, the pH of the enhancement brine for this aging treatment should be noted as 5.03.

Drip Loss

Drip loss found by fabricating 2.54 cm cubes from steak 1 for each treatment. Drip loss assessment was accomplished by a method modified from Honikel (1987). The initial cubes were weighed and suspended from a rack for a 24 hr period in a 4° C, dark room. The rack was covered with a plastic film to prevent dehydration of the samples by airflow. After the 24 hr period, the cubes were reweighed and drip loss was calculated as a % by using the equation:

$$\frac{(\text{initial weight} - \text{end weight})}{\text{initial weight}} * 100$$

initial weight

Subjective Retail Color Assessment

Steaks were visually evaluated by a six member trained panel once daily for a 14 d period. The steaks were assessed for lean color, fat color, percent lean discoloration, and overall acceptability (Appendix B). Overall acceptability represented the combined effects of lean color, fat color, and percent discoloration and was utilized as an indicator of acceptability of the retail products.

Procedures for Cooked Lean Color Analysis

Steaks were cooked on an impingement oven (Lincoln Impinger, Model 1132-000-A, Ft. Wayne, IN) at 177° C to an internal temperature of 70° C. The internal temperature was monitored using a VersaTuff 386 thermocouple thermometer (Atkins Tempotec, Gainesville, FL). Following cooking, steaks were allowed to cool for approximately 3 min. Steaks were then cut through their horizontal center exposing the geometric center. A trained panel then evaluated the center of each cooked steak. Evaluators assigned a number according to a six point photographic scale (Appendix C) according to the Beef Steak Color Guide-Degree of Doneness chart (American Meat Science Association [AMSA], 1995).

Objective Color Assessment

Objective color values ($L^*a^*b^*$) were obtained using a Miniscan XE Plus (Hunter Lab, Reston, VA). $L^*a^*b^*$ s were obtained 5 times for each treatment: at the National Beef packing plant in Liberal, KS, d 1 post-treatment, d 7 post-treatment, d 7 post-treatment cooked and d 14 post-treatment.

pH

pH was obtained using a model IQ150 pH meter. pH was measured at the same times as objective color: at the National Beef packing plant in Liberal, KS, d 1 post-treatment, d 7 post-treatment, d 7 post-treatment cooked and d 14 post-treatment.

Headspace

Headspace of the MAP trays was measured for each treatment on d 7 and 14 post-treatment using a Mocon Oxygen headspace analyzer (model HS-750, MOCON Modern Controls Inc., Minneapolis, MN). Headspace was reported as percent oxygen.

Odor Panel

Steaks for the odor panel were left in simulated retail storage for 7 d post-treatment. After the steaks were removed from storage, each package was opened individually and presented before a panel immediately after breaking the seals on the MAP trays. The panel evaluated odor on a six-point scale (Appendix D).

Total Aerobic Plate Counts (TPC)

Samples for TPC were taken on d 1, 7, and 14 post-treatment. All samples were aseptically packaged in whirl-pak bags overnight mailed to Food Safety Net Services (San Antonio, TX) for standard total plate counts. Food Safety Net followed standard plating methodology outlined by FDA's Bacteriological Analytical Method (BAM). Samples were diluted with peptone in a sterile stomacher bag and pummeled for 1 min. The homogenate was then spiral plated (0.25 mL per plate in quadruplet) onto tryptic soy agar. Plates were incubated at 25° C for 48 hours, counted and reported in TPC per cm².

Thiobarbituric Acid Analysis

Estimates of lipid oxidation on the surface of samples are made using the thiobarbituric acid (TBA) test. Samples from d 1, d 7 and d 14 post-treatment were packaged in whirl-pak bags and frozen at -20° C until further analysis. TBA analysis was performed using the test procedures described by Buege and Aust (1978) with the following modifications: a 10 g sample was homogenized with 30 mL deionized water in a Waring Commercial Blender (Model 33BL79, Waring Products Division Dynamics Corporation of America, New Hartford, CT) and centrifuged at 1850 G for 10 min. at 4° C (Beckman Induction Drive Centrifuge, Model J-6M, Beckman Instruments, Inc., Houston, TX). Two mL of homogenate, in duplicate, was subjected to TBA reagent and cooked in a boiling water bath. After cooling, absorbencies of the supernatant at 531 nm were measured using a spectrophotometer (Model DU 7500, Beckman Instruments, Inc., Houston, TX). Results were reported as Thiobarbituric acid reactive substance (TBARS) representing mg malondialdehyde (MDA) equivalents per kg of fresh meat.

Slice Shear Force

Steaks were randomly assigned to a cooking order. They were then allowed to temper at 4° C 24 h prior to cooking on an impingement oven (Lincoln Impinger, Model 1022, Ft. Wayne, IN) to an internal temperature of 70° C. The internal temperature was monitored using a VersaTuff 386 thermocouple thermometer (Atkins Temptec, Gainesville, FL). Steaks were then chilled overnight to a temperature of 4° C. A 5 cm long sample was then cut from each

steak about 2 cm from the lateral end of the muscle. A two bladed knife (1 cm apart) was used to cut a 1 cm thick, 5 cm long sample at a 45° angle to the long axis of the longissimus and parallel to the muscle fibers. The slice was then sheared perpendicular to the muscle fibers using a Universal Instron Testing Machine (Model 4502, Instron, Canton, MS) equipped with a flat, blunt-end blade with a crosshead speed set at 500 mm/min. Shear force of each sample was reported in kg of peak load, which was logged by a Dell Optiplex GS400 computer.

Sensory Panel

Sensory panelists were trained for sensory analysis following AMSA (1995) guidelines. Steaks were removed from the MAP packages after 7 d of retail display and were placed in vacuum-sealed packages and stored at -2° C until further analysis. Meat cuts were tempered for 24 hours at 4° C, and then broiled on an impingement oven (Lincoln Impinger, Model 1022, Ft. Wayne, IN) at 177° C to an internal temperature of 70° C (medium degree of doneness). The internal temperature was monitored using a VersaTuff 386 thermocouple thermometer (Atkins Temptec, Gainesville, FL). Eight sessions, consisting of six-trained panelists were performed, 14 samples per panel. Two cubic portions (1.3 cm x 1.3 cm x cooked steak thickness) from each sample were served warm to panelists under red lighting to masque color differences. The panelists were instructed to record the average of their two portions. Samples were evaluated for tenderness, juiciness, connective tissue, uncharacteristic flavor, and overall

acceptability (Appendix E). Panelists were given unsalted crackers, distilled water, and an expectorant cup to cleanse their palate between samples.

Statistical Analyses

All results were analyzed using generalized least squares (PROC MIXED, SAS Inst., Inc., Cary, NC). Data was analyzed to measure the effect of post-mortem aging, high-pressure oxygenation and enhancement on sensory analysis, thiobarbituric acid analysis, cooked color analysis, total plate count, slice shear force, pH, L*a*b*, odor analysis, headspace of MAP packages and drip loss. Repeated measurement analysis was performed on all subjective retail display data. All tests were conducted at the nominal significance level of 0.05.

RESULTS AND DISCUSSION

pH

pH measurements were taken at five different points in this experiment: at the plant, on the first day post-treatment, on the seventh day post-treatment, on the fourteenth day post-treatment, and after cooking steaks to a medium degree of doneness (70 ° C) on the seventh day post-treatment. Means for pH values ($P < 0.05$) at the packing plant are given in Table 1. Dark Cutter Score was determined by a USDA Grader combined with Hunter Color Values (L*a*b*). Mean pH of longissimus dorsi from non-dark cutting carcasses was 5.42, 5.92 in 1/3 dark cutters, 6.25 in 2/3 dark cutters and 6.85 in full dark cutters. This agrees

with the findings of Munns and Burrell (1965). They concluded that approximately 90% of dark cutters have an ultimate longissimus dorsi pH of at least 6.0.

Main effects of the four treatments for day one post-treatment are given in Table 2. There was a dark cutter score effect on pH values ($P < 0.05$), however there was also an oxygenation effect on pH values ($P < 0.05$) in that non-oxygenated steaks had a lower mean pH value than oxygenated steaks. It should be mentioned that there was no effect on pH for neither enhancement treatment nor aging period ($P > 0.05$). Perhaps the fact that there was no enhancement effect could be attributed to the solution not being completely equilibrated throughout the steaks.

Day 7, following oxygenation and enhancement, means of the main effects on pH values are given in Table 3. The oxygenation effect present in d 1 was no longer observed ($P > 0.05$). However, both enhancement and aging period displayed an influence on pH values ($P < 0.05$). A pH lowering effect resulted from enhancement treatment, and this was expected after using the acidic phosphate blend in the enhancement brine solution.

An interaction was found to exist after taking measurements fourteen days post-treatment (Table 4); dark cutter score combined with enhancement treatment incorporation had an effect on pH values. The non-enhanced steaks exhibited significantly higher ($P < 0.05$) pH values for both 2/3 and full dark cutters when compared to their enhanced counterparts. The low pH of the brine solution obviously had more of an effect on the higher pH levels of the steaks

from 2/3 and full dark cutters than it had on the steaks from non-dark cutters and 1/3 dark cutters, as the non-enhanced steaks from the non-dark cutters and 1/3 dark cutters were not significantly different ($P > 0.05$) than the enhanced steaks from the same dark cutter scores.

Main effects of the treatments on pH values of cooked steaks are given in Table 5. Once again, there is both an enhancement effect as well as aging effect in addition to the dark cutter effect. As expected, enhanced steaks, once again, displayed lower pH values ($P < 0.05$) than their non-enhanced counterparts. Steaks that were aged 7 d prior to treatment were lower in pH than steaks aged 14 d prior to treatment ($P < 0.05$). Full dark cutters were found to be higher in pH ($P < 0.05$) than 2/3 dark cutters. 2/3 dark cutters were in turn higher in pH ($P < 0.05$) than 1/3 dark cutters. However, no difference in pH ($P > 0.05$) was found between 1/3 dark cutters and non-dark cutters.

Objective Lean Color

Results for carcass $L^*a^*b^*$'s from the packing plant are given in Table 6. As expected, L^* readings decrease as dark cutter score increases, a^* readings decrease as dark cutter score increases, and b^* readings decrease as dark cutter score increases. These results agree with Page et al. (2001) and Wulf and Wise (1999). Page et al. concluded that $L^*a^*b^*$ values are negatively correlated to muscle pH. They described that the higher the pH of the beef was, the more green and blue it was rather than the more red or yellow colors associated with the lower pH of normal beef.

Main effect results for L* values for d 1 of retail display of strip loin steaks are given in Table 7. Non-oxygenated steaks were found to be darker ($P < 0.05$) than oxygenated steaks. L* values once again decreased as dark cutter score increased. There were no effects found to occur for enhancement treatment or aging period.

An interaction for L* values occurred on d 7 between dark cutter score and enhancement treatment (Table 8). The only significant difference ($P < 0.05$) found for enhancement treatment occurred for full dark cutters. However, all non-enhanced steaks were darker in color than the enhanced steaks. As expected, steaks became darker for both enhanced and non-enhanced steaks as dark cutter score increased.

A four-way interaction for a* values occurred on d 1 of retail display (Table 9). Oxygenated steaks for all combinations were more red ($P < 0.05$) than non-oxygenated steaks. The non-enhanced steaks were more consistent in having lower values as dark cutter score increased than enhanced steaks did.

An interaction between dark cutter score and enhancement treatment occurred on d 14 of retail display for a* values (Table 10). Non-enhanced steaks were shown to be more red ($P < 0.05$) than enhanced steaks for all dark cutter score groups. No difference was shown to exist between dark cutter scores 1/3, 2/3 and full dark cutters. However, the non-dark cutters were found to not be as red ($P < 0.05$) than steaks from the dark cutter categories.

Main effects of oxygenation treatment and aging period for b* values on d 1 of retail display are given in Table 11. Oxygenated steaks had higher b*

values ($P < 0.05$) than non-oxygenated steaks and steaks aged 7 d prior to treatment had higher b^* values ($P < 0.05$) than steaks aged 14 d prior to treatment.

An interaction between dark cutter score and enhancement treatment occurred on d 7 of retail display for b^* values (Table 12). b^* values for steaks from non-enhanced full dark cutters were significantly lower ($P < 0.05$) than steaks from enhanced full dark cutters and both non-enhanced and enhanced steaks from non-dark cutters, 1/3 and 2/3 dark cutters.

L^* values for cooked steaks are shown in a three-way interaction between oxygenation treatment, enhancement treatment and dark cutter score (Table 13). L^* values decreased as dark cutter score increased in all oxygenation-enhancement combinations other than non-oxygenated/enhanced. No differences were found to exist between oxygenation/enhancement combinations.

An interaction between dark cutter score and oxygenation treatment occurred for a^* values of cooked steaks (Table 14). Non-oxygenated steaks from the 1/3 and 2/3 dark cutter classifications had higher a^* values ($P < 0.05$) than oxygenated steaks did. a^* values also increased as dark cutter score increased.

Main effects of enhancement treatment and aging period for a^* values of cooked steaks are given in Table 15. Non-enhanced steaks were had higher a^* values ($P < 0.05$) than enhanced steaks and steaks aged 7 days prior to treatment had higher a^* values ($P < 0.05$) than steaks aged 14 days prior to treatment.

Cooked Color Panel

No significant interactions among the treatments were found for subjective cooked color. However, each treatment independently had an effect on cooked color (Table 16). As previously mentioned, steaks were cooked to a medium degree of doneness (70° C). Non-dark cutters were viewed as being more brown or done than 1/3, 2/3 and full dark cutters ($P < 0.05$). In addition, 1/3 dark cutters were seen as being more done than 2/3 and full dark cutters as well ($P < 0.05$). There was no difference in 2/3 versus full dark cutters ($P > 0.05$).

Oxygenated steaks were viewed as being more done ($P < 0.05$) than non-oxygenated steaks. Perhaps this could be attributed to more metmyoglobin formation from the increased amount of oxygen presented within the steak because of the high-pressure system.

Panelists viewed enhanced steaks being more done ($P < 0.05$) than non-enhanced steaks. a^* values were also higher for non-enhanced steaks than their enhanced counterparts. Salt and phosphates have both been found to cause color problems in cooked meat by altering the denaturation of myoglobin (von Hippel and Schleich, 1969).

Steaks aged 14 d prior to treatment were found to be more done appearing ($P < 0.05$) than steaks aged only 7 d prior to treatment. This could be attributed to the denaturation of myoglobin during the extra seven days of age.

Retail Shelf Life

A significant interaction involving enhancement treatment, dark cutter score, aging period and retail display period effects on overall acceptability are given in Figure 1. Several general observations can be made in that it appeared that throughout the initial 10 d of retail simulation, full dark cutting steaks, regardless of oxygenation treatment, that were aged for 14 d had low overall desirability rating than remaining treatment combinations. However, the last four days of display (d 11-14), other treatment combinations received similar ratings to the extended aged, full dark cutting steaks. Additionally, results suggested that non-enhanced steaks received the highest overall ratings throughout retail simulation. This appeared to be especially true during the initial along with the completion of retail display. Additionally, a great deal of variation in overall desirability was in the 2/3 and full dark cutting steak population aged 14 d prior to any treatments. This could possibly be a result of the denaturation of myoglobin directly resulting from the additional age of the steaks. A gradual decline in overall acceptability was found to exist from d 1 through 11. On d 12 through 14 however, the most variation in the treatments was found to occur. Most of the enhanced treatments were found be at least slightly undesirable in overall acceptability – especially the steaks aged 14 d prior to treatment in the 2/3 and full dark cutting categories.

A second interaction involving oxygenation treatment, enhancement treatment, aging period and dark cutter score effects on overall acceptability are

given in Figure 2. Steaks from 2/3 and full dark cutting strip loins aged 14 d prior to oxygenation and enhancement treatments were found to not be as acceptable as steaks aged 7 d. Steaks from 1/3 dark cutting strip loins were found that they can be aged 14 d as long as they are not enhanced.

Total Plate Count

Three significant interactions were noticed on day 1 post-treatment. An enhancement treatment and aging period effected total aerobic plate counts (Table 17) in that non-enhanced and enhanced steaks aged 14 d prior to enhancement displayed higher bacterial growth ($P < 0.05$) than steaks aged for only 7 d prior to enhancement.

A second interaction was observed in that oxygenation treatment and aging period influenced total aerobic plate counts from samples taken on day 1 of retail display (Table 18). Both oxygenated steaks and non-oxygenated steaks aged 14 prior to treatment displayed a higher number bacteria ($P < 0.05$) than oxygenated and non-oxygenated steaks aged only 7 d prior to oxygenation. A contrasting finding was noticed in that non-oxygenated steaks aged 14 d displayed higher total plate counts ($P < 0.05$) when compared to oxygenated steaks of the same postmortem aging period. No difference ($P > 0.05$) existed between oxygenated and non-oxygenated steaks aged 7 d prior to oxygenation treatment.

The concluding interaction that occurred on day 1 was the influence of dark cutter score and aging period on total plate counts (Table 19). Yet again,

steaks aged 14 d prior to oxygenation treatment yielded higher total plate counts ($P < 0.05$) than those aged 7 d prior to treatment. Steaks from 1/3, 2/3 and full dark cutting strip loins aged 14 d prior to treatment contained higher total plate counts ($P < 0.05$) than steaks from non-dark cutters within the same postmortem aging period.

Odor Panel.

A four-way interaction that involved oxygenation treatment, enhancement treatment, dark cutter score and aging period's influence on odor score are summarized in Table 20. Most steaks aged 14 d prior to treatment had a stronger odor than steaks aged only 7 d for all oxygenation/enhancement treatments, which suggests a possibility of more bacterial growth, which is to be expected. Enhanced steaks that were either oxygenated or non-oxygenated had higher odor scores than non-enhanced steaks for both oxygenated and non-oxygenated steaks for all dark cutter scores with a couple of exceptions. Oxygenated/non-enhanced steaks had a stronger odor in the 1/3 and 2/3 dark cutter scores than their enhanced counterparts. This suggests the possibility that enhancement solution worked well enough in the enhanced steaks to deter microbial growth.

Lipid Oxidation.

A three-way interaction occurred involving oxygenation treatment, enhancement treatment and dark cutter score on lipid oxidation concentration (Table 21). No difference was found between enhanced steaks for neither

oxygenated nor non-oxygenated steaks among dark cutter scores. However, in the dark cutter scores 1/3, 2/3 and full, a very noticeable difference ($P < 0.05$) was found between oxygenated and non-oxygenated steaks that were not enhanced. The non-oxygenated steaks had more lipid oxidation than the oxygenated steaks. As dark cutter score got higher, the amount of lipid oxidation went down for all oxygenation, enhancement treatment combinations.

A second three-way interaction occurred involving aging period, oxygenation treatment and retail display period effects on lipid oxidation levels (Table 22). As expected, oxidation of the steaks increased for all aging period/oxygenation treatment treatments as retail display time increased with a few exceptions. No difference was found between d 1 and 7 retail display for both 7 d aged/oxygenated steaks and 14 d aged/oxygenated steaks. There was also no difference between d 7 and 14 retail display for 14 d aged/non-oxygenated steaks. Oxygenated steaks contained less oxidation for d 7 and 14 than non-oxygenated steaks did. However, on day 1 of retail display the trend was the opposite. Steaks aged 14 d prior to treatment had more oxidation than steaks aged 7 days prior to treatment.

A third interaction occurred involving dark cutter score and day of retail display period effects on lipid oxidation (Table 23). As retail display period decreased and dark cutter score increased, oxidation levels were lower. However, in the full dark cutter category, even though lipid oxidation increased as retail display time increased, the changes were not significant ($P > 0.05$).

A fourth interaction occurred involving enhancement treatment and retail display period effects on lipid oxidation (Table 24). Like previously mentioned, as retail display time increased, lipid oxidation also increased. Initial levels on d 1 were the same ($P > 0.05$) for enhanced and non-enhanced steaks. However, at 7 and 14 d, non-enhanced steaks had more oxidation ($P < 0.05$) than enhanced steaks did. The fact that the enhancement decreased lipid oxidation values agrees with findings by Paterson and Parrish (1988).

Sensory Panel

Panelists evaluated the strip steaks for tenderness, juiciness, connective tissue, uncharacteristic flavor and overall acceptability (Appendix E). There were no significant interactions among any treatments or sensory attributes. The investigation did however, find several main effects (Table 25). Panelists found the non-oxygenated steaks to be more tender than the oxygenated steaks ($P < 0.05$). Perhaps this could be attributed to a halo effect of the attributes juiciness and uncharacteristic flavor although they were not significant. Connective tissue was also insignificantly lower in non-oxygenated steaks than oxygenated steaks. As expected, the panelists found the enhanced steaks to be more tender, juicier and have less connective tissue than the non-enhanced steaks ($P < 0.05$). This agrees with findings by Vote et al. (2000), Sheard et al. (1999) and Robbins et al. (2003). There was no effect for aging period on any of the five sensory attributes measured. There was a negative effect of enhancement on uncharacteristic flavor as compared to non-enhancement ($P < 0.05$). The panelists found the

enhanced steaks to have a moderately uncharacteristic flavor, whereas they found the non-enhanced steaks to have a slight to no uncharacteristic flavor. common responses for the enhanced steaks included salty, livery, rancid, soapy and metallic off-flavors. Non-oxygenated steaks were found to be more acceptable overall than steaks that were oxygenated ($P < 0.05$). This possibly could be attributed to the notion that the panelists found them to be more acceptable in the other four attributes, although not significantly. Non-enhanced steaks were also found to be more acceptable overall than steaks that were enhanced ($P < 0.05$) and can probably to attributed to the differences in uncharacteristic flavors. Steaks from full dark cutters were also found to be more acceptable overall than steaks from non-dark cutters, 1/3 dark cutters and 2/3 dark cutters. Perhaps this can be attributed to the panelists finding them juicier and with the least uncharacteristic flavor, although not significantly.

Slice Shear Force

No interactions were found among the four treatments for shear force values, however there were main effects. Enhanced strip steaks, as expected, were significantly more tender (Table 26) than non-enhanced strip steaks ($P < 0.05$). There was also a dark cutter effect on tenderness ($P < 0.05$); non dark cutters and full dark cutters were significantly more tender than 1/3 and 2/3 dark cutters. This coincides with results found by Purchas (1990), Watanabe et al. (1995), and Wulf et al. (1997). They found that toughness was maximized at pH levels between 5.8 and 6.0. The initial pH levels of strip loins classified as 1/3

and 2/3 dark cutters in this study were 5.92 and 6.25, respectively. Another possible explanation for this could be that Wulf et al. (2002), described a greater variation in shear force values from cooked longissimus dorsi from dark, firm and dry carcasses as compared to normal carcasses.

Headspace

An interaction involving dark cutter score and retail display period effected % oxygen headspace (Table 27). There were no differences between the four dark cutter categories in percent Oxygen on day 7 of retail display. However, on d 14 of retail display, non-dark cutters and 1/3 dark cutters contained more oxygen in the headspace than 2/3 and full dark cutters. The non-dark cutters and 1/3 dark cutters also contained more oxygen on d 14 than on d 7. For 2/3 and full dark cutters, the opposite occurred.

A second interaction that occurred involved aging period in combination with retail display period (Table 28). Steaks that were aged 7 d prior to treatment and on retail display for 14 d after treatment and steaks that were aged 14 d prior to treatment and on retail display for 7 days post-treatment contained more oxygen in their headspace ($P < 0.05$) than steaks aged 7 days and on retail display 7 days and also steaks aged 14 days and on retail display 14 days. The two combinations of steaks with the most Oxygen in their headspace were each a total of 21 days old. The other two groups were a total of 14 days old and 28 days old.

Drip Loss

A three-way interaction involving oxygenation treatment, enhancement treatment and aging period effects on drip loss are shown in Table 29. The largest difference in drip loss is found to occur between steaks aged 7 days and steaks aged 14 days prior to treatment. Steaks aged 14 days lost much more weight ($P < 0.05$) than steaks aged only 7 days. Perhaps this can be attributed to the breaking down of muscle proteins during the extra 7 days of age, therefore decreasing the water holding capacity of the samples. Oxygenated/enhanced steaks had the lowest drip loss for all steaks aged 7 d, however, for steaks aged 14 d drip loss for the same treatment was higher ($P < 0.05$) than all other treatment combinations. One possible explanation for this could be that the combination of the high-pressure oxygenation, aging and enhancement disrupted enough muscle proteins to make them incapable of holding the extra water weight that was injected into them.

IMPLICATIONS

The inclusion of high-pressure oxygenation in strip steaks from dark cutting beef carcasses improves lean color characteristics. However, further research should be conducted to lower the oxygenation cycle from 12 hours to make it more industry applicable. Enhancement using an acidic phosphate had no effect on lean color characteristics and was detrimental on uncharacteristic flavors using a trained sensory panel. Continued research is also needed in lowering pH in dark cutting muscle to improve shelf life without affecting sensory attributes.

Table 1. The effect of dark cutter score on pH of longissimus dorsi muscle from carcasses measured at packing plant.

	<i>Dark Cutter Score</i> ¹			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>pH</i>	5.42 ^d	5.92 ^c	6.25 ^b	6.85 ^a
<i>Standard Error</i>	0.02	0.04	0.12	0.03

¹ 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

^{a,b,c,d} Means with differing superscripts are significantly different (P < 0.05).

Table 2. The effects of dark cutter score, oxygenation treatment, enhancement treatment and aging period on pH of strip loin steaks on day 1 of retail display¹.

	Dark Cutter Score ²			Oxygenation Treatment ³			Enhancement Treatment ⁴			Aging Period ⁵		
	0	1/3	2/3	3/3	O	N	E	N	7	14		
pH	5.60 ^d	5.89 ^c	6.27 ^b	6.46 ^a	6.12 ^a	5.99 ^b	6.11 ^a	6.11 ^a	6.04 ^a	6.07 ^a		
Standard Error	0.05	0.05	0.05	0.06	0.04	0.04	0.04	0.03	0.04	0.04		

¹ These variables were not involved in any significant interactions, therefore results of main effect separation are shown.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ O, N equals oxygenated and non-oxygenated, respectively.

⁴ E, N equals enhanced and non-enhanced, respectively.

⁵ 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a,b,c,d} Means with differing superscripts within a treatment are significantly different (P < 0.05).

Table 3. The effects of dark cutter score, oxygenation treatment, enhancement treatment and aging period on pH of strip loin steaks on day 7 of retail display¹.

	Dark Cutter Score ²			Oxygenation Treatment ³		Enhancement Treatment ⁴		Aging Period ⁵		
	0	1/3	2/3	3/3	O	N	E	N	7	14
pH	5.58 ^d	5.88 ^c	6.25 ^b	6.44 ^a	6.03 ^a	6.04 ^a	5.97 ^b	6.14 ^a	6.10 ^a	5.97 ^b
Standard Error	0.05	0.05	0.05	0.05	0.03	0.03	0.05	0.03	0.03	0.03

¹ These variables were not involved in any significant interactions, therefore results of main effect separation are shown.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ O, N equals oxygenated and non-oxygenated, respectively.

⁴ E, N equals enhanced and non-enhanced, respectively.

⁵ 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a,b,c,d} Means with differing superscripts within a treatment are significantly different (P < 0.05).

Table 4. The effect of dark cutter score and enhancement treatment on pH of strip loin steaks on day 14 of retail display.

<i>Enhancement Treatment</i> ²	<i>Dark Cutter Score</i> ¹			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>E</i>	5.79 ^{cd}	5.89 ^c	5.99 ^c	6.26 ^b
<i>N</i>	5.59 ^d	5.93 ^c	6.33 ^{ab}	6.48 ^a

¹ 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

² E, N equals enhanced and non-enhanced, respectively.

^{a,b,c,d} Means with differing superscripts are significantly different (P < 0.05).

Table 5. The effects of dark cutter score, oxygenation treatment, enhancement treatment and aging period on cooked pH of strip loin steaks cooked to an internal temperature of 70° C (medium degree of doneness) after 7 days of retail display¹.

	Dark Cutter Score ²			Oxygenation Treatment ³			Enhancement Treatment ⁴			Aging Period ⁵
	0	1/3	2/3	3/3	O	N	E	N	7	14
Cooked pH	6.02 ^c	6.13 ^c	6.37 ^b	6.64 ^a	6.28 ^a	6.30 ^a	6.18 ^b	6.40 ^a	6.36 ^a	6.23 ^b
Standard Error	0.07	0.07	0.07	0.07	0.04	0.04	0.05	0.05	0.05	0.05

¹ These variables were not involved in any significant interactions, therefore results of main effect separation are shown.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ O, N equals oxygenated and non-oxygenated, respectively.

⁴ E, N equals enhanced and non-enhanced, respectively.

⁵ 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a,b,c} Means with differing superscripts within a treatment are significantly different (P < 0.05).

Table 6. The effect of dark cutter score on L*a*b* values of longissimus dorsi muscle from carcasses measured at packing plant.

	<i>Dark Cutter Score</i> ¹			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>L</i> ²	40.23 ^a	34.39 ^b	30.16 ^c	30.00 ^c
<i>a</i> ³	22.59 ^a	18.17 ^b	17.16 ^b	14.10 ^c
<i>b</i> ⁴	19.56 ^a	14.21 ^b	13.03 ^c	9.85 ^d

¹ 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

² 0 = black, 100 = white.

³ Negative values = green, positive values = red.

⁴ Negative values = blue, positive values = yellow.

^{a,b,c} Means within a row with differing superscripts are significantly different (P < 0.05).

Table 7. The effects of dark cutter score, oxygenation treatment, enhancement treatment and aging period on L^{*1} values of strip loin steaks after 1 day of retail display².

	Dark Cutter Score ³			Oxygenation Treatment ⁴			Enhancement Treatment ⁵			Aging Period ⁶	
	0	1/3	2/3	3/3	O	N	E	N	7	14	
L^{*1}	39.44 ^a	34.35 ^b	32.98 ^c	31.79 ^c	36.71 ^a	32.57 ^b	35.43 ^a	34.83 ^a	34.25 ^a	35.02 ^a	
Standard Error	0.63	0.63	0.61	0.67	0.45	0.45	0.58	0.58	0.45	0.45	

¹ 0 = black, 100 = white.

² These variables were not involved in any significant interactions, therefore results of main effect separation are shown.

³ 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

⁴ O, N equals oxygenated and non-oxygenated, respectively.

⁵ E, N equals enhanced and non-enhanced, respectively.

⁶ 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a,b,c} Means with differing superscripts within a treatment are significantly different ($P < 0.05$).

Table 8. The effect of dark cutter score and enhancement treatment on L*¹ values of strip loin steaks on day 7 of retail display.

<i>Enhancement Treatment</i> ³	<i>Dark Cutter Score</i> ²			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>E</i>	40.08 ^a	37.29 ^b	31.45 ^c	30.62 ^c
<i>NE</i>	39.46 ^a	35.71 ^b	31.00 ^c	26.54 ^d

¹ 0 = black, 100 = white.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ E, N equals enhanced and non-enhanced, respectively.

^{a,b,c,d} Means with differing superscripts are significantly different (P < 0.05).

Table 9. The effect of oxygenation treatment, enhancement treatment, dark cutter score and aging period on a¹ values of strip loin steaks after 1 day of retail display.

<i>Oxygenation Treatment : Enhancement Treatment²</i>				
	<i>NO : E</i>	<i>NO : NE</i>	<i>O : E</i>	<i>O : NE</i>
<i>0 : 7</i>	16.27 ^{kl}	17.14 ^k	27.16 ^{efgh}	28.67 ^{cdefg}
<i>0 : 14</i>	14.84 ^{lm}	16.16 ^{kl}	25.07 ^{hij}	26.77 ^{gh}
<i>1/3 : 7</i>	15.00 ^{klm}	16.02 ^{kl}	29.52 ^{bcd}	30.78 ^{abc}
<i>1/3 : 14</i>	15.85 ^{kl}	16.77 ^{kl}	23.74 ^j	29.14 ^{cdef}
<i>2/3 : 7</i>	16.26 ^{kl}	16.57 ^{kl}	31.48 ^{ab}	30.44 ^{abc}
<i>2/3 : 14</i>	16.01 ^{kl}	14.64 ^{lm}	26.99 ^{fgh}	29.39 ^{bcde}
<i>3/3 : 7</i>	16.50 ^{kl}	14.64 ^{lm}	27.48 ^{defg}	26.58 ^{ghi}
<i>3/3 : 14</i>	15.33 ^{kl}	12.92 ^m	31.93 ^a	24.45 ^{ij}

Dark Cutter Score : Aging Period³

¹ Negative values = green, positive values = red.

² Oxygenation Treatment : Enhancement Treatment; NO = Non-Oxygenated, O = Oxygenated, NE = Non-Enhanced and E = Enhanced.

³ Dark Cutter Score : Aging Period; 0 = Non-Dark Cutter, 1/3 = 1/3 Dark Cutter, 2/3 = 2/3 Dark Cutter, 3/3 = Full Dark Cutter, 7 = 7 days of age prior to treatment, 14 = 14 days of age prior to treatment.

^{a,b,c,d,e,f,g,h,i,j,k,l,m} Means with differing superscripts are significantly different (P < 0.05).

Table 10. The effect of dark cutter score and enhancement treatment on a*¹ values of strip loin steaks on day 14 of retail display.

<i>Enhancement Treatment</i> ³	<i>Dark Cutter Score</i> ²			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>E</i>	15.83 ^c	18.49 ^b	17.47 ^{bc}	16.96 ^{bc}
<i>NE</i>	18.26 ^b	22.85 ^a	21.80 ^a	20.87 ^a

¹ Negative values = green, positive values = red.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ E, N equals enhanced and non-enhanced, respectively.

^{a,b,c} Means with differing superscripts are significantly different (P < 0.05).

Table 11. The effects of oxygenation treatment and aging period on b^{*1} values of strip loin steaks after 1 day of retail display.

	<i>Oxygenation Treatment</i> ²		<i>Aging Period</i> ³	
	<i>O</i>	<i>NO</i>	<i>7</i>	<i>14</i>
b^{*1}	22.10 ^a	13.96 ^b	18.56 ^a	17.50 ^b
<i>Standard Error</i>	0.27	0.27	0.22	0.22

¹ Negative values = blue, positive values = yellow.

² O, N equals oxygenated and non-oxygenated, respectively.

³ 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a,b} Means with differing superscripts within a treatment are significantly different ($P < 0.05$).

Table 12. The effect of dark cutter score and enhancement treatment on b^{*1} values of strip loin steaks on day 7 of retail display.

<i>Enhancement Treatment</i> ³	<i>Dark Cutter Score</i> ²			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>E</i>	21.25 ^{ab}	20.99 ^{ab}	20.81 ^{ab}	20.88 ^{ab}
<i>NE</i>	21.58 ^a	20.78 ^{ab}	20.05 ^b	15.73 ^c

¹ Negative values = blue, positive values = yellow.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ E, N equals enhanced and non-enhanced, respectively.

^{a,b,c} Means with differing superscripts are significantly different ($P < 0.05$).

Table 13. The effect of oxygenation treatment, enhancement treatment and dark cutter score on L*¹ values of cooked (70° C) strip loin steaks after 7 days of retail display.

Dark Cutter Score ³	Oxygenation Treatment : Enhancement Treatment ²			
	NO : E	NO : NE	O : E	O : NE
0	52.47 ^{abcde}	53.89 ^{abcd}	54.03 ^{abc}	51.90 ^{bcdef}
1/3	54.56 ^a	52.19 ^{abcde}	54.29 ^{ab}	53.35 ^{abcde}
2/3	49.33 ^{fg}	50.88 ^{efg}	51.31 ^{defg}	51.42 ^{cdefg}
3/3	53.14 ^{abcde}	48.84 ^g	51.29 ^{defg}	50.75 ^{efg}

¹ 0 = black, 100 = white.

² Oxygenation Treatment : Enhancement Treatment; NO = Non-Oxygenated, O = Oxygenated, NE = Non-Enhanced and E = Enhanced.

³ Dark Cutter Score: 0, 1/3, 2/3, 3/3 = Non-Dark Cutter, 1/3 Dark Cutter, 2/3 Dark Cutter and Full Dark Cutter, respectively.
a,b,c,d,e,f,g Means with differing superscripts are significantly different (P < 0.05).

Table 14. The effect of dark cutter score and oxygenation treatment on a^{*1} values of cooked (70° C) strip loin steaks after 7 days of retail display.

<i>Oxygenation Treatment</i> ³	<i>Dark Cutter Score</i> ²			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>O</i>	6.29 ^c	7.52 ^c	12.81 ^b	16.55 ^a
<i>NO</i>	6.54 ^c	13.10 ^b	18.53 ^a	16.21 ^a

¹ Negative values = green, positive values = red.

² Dark Cutter Score: 0, 1/3, 2/3, 3/3 = Non-Dark Cutter, 1/3 Dark Cutter, 2/3 Dark Cutter and Full Dark Cutter, respectively.

³ Oxygenation Treatment: O = Oxygenated, NO = Non-Oxygenated.

^{a,b,c} Means with differing superscripts are significantly different (P < 0.05).

Table 15. The effects of enhancement treatment and aging period on a^{*1} values of cooked (70° C) strip loin steaks after 7 days of retail display.

	<i>Enhancement Treatment</i> ¹		<i>Aging Period</i> ²	
	<i>E</i>	<i>NE</i>	<i>7</i>	<i>14</i>
a^{*1}	9.08 ^b	15.31 ^a	14.13 ^a	10.26 ^b
<i>Standard Error</i>	0.91	0.91	0.91	0.91

¹ Negative values = green, positive values = red.

² Enhancement Treatment: E = Enhanced, NE = Non-Enhanced.

³ Aging Period: 7, 14 = 7 and 14 days of age prior to treatment, respectively.

^{a,b} Means within a treatment with differing superscripts are significantly different (P < 0.05).

Table 16. The effects of dark cutter score, oxygenation treatment, enhancement treatment and aging period on subjective cooked lean color values¹ of strip loin steaks cooked to an internal temperature of 70° C (medium degree of doneness) after 7 days of retail display.

	Dark Cutter Score ²			Oxygenation Treatment ³			Enhancement Treatment ⁴			Aging Period ⁵	
	0	1/3	2/3	3/3	O	N	E	N	7	14	
Cooked Color	5.59 ^a	4.81 ^b	4.02 ^c	4.03 ^c	4.81 ^a	4.42 ^b	5.02 ^a	4.20 ^b	4.43 ^b	4.80 ^a	
Standard Error	0.16	0.16	0.16	0.16	0.11	0.11	0.11	0.11	0.11	0.11	

¹ Cooked Lean Color values were scored by a trained panel using the scale: 1 = very rare, 2 = rare, 3 = medium rare, 4 = medium, 5 = well done, 6 = very well done.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ O, N equals oxygenated and non-oxygenated, respectively.

⁴ E, N equals enhanced and non-enhanced, respectively.

⁵ 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a,b,c,d} Means with differing superscripts within a treatment are significantly different (P < 0.05).

Table 17. The effect of enhancement treatment and aging period on total plate counts of strip loin steaks on day 1 of retail display.

<i>Aging Period</i> ²	<i>Enhancement Treatment</i> ¹	
	<i>E</i>	<i>N</i>
7	2.88 ^c	2.47 ^d
14	3.82 ^b	4.32 ^a

¹ E, N equals enhanced and non-enhanced, respectively.

² 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a, b, c} Means with differing superscripts are significantly different (P < 0.05).

Table 18. The effect of oxygenation treatment and aging period on total plate counts of strip loin steaks on day 1 of retail display.

<i>Aging Period</i> ²	<i>Oxygenation Treatment</i> ¹	
	<i>O</i>	<i>N</i>
7	2.86 ^c	2.49 ^c
14	3.78 ^b	4.36 ^a

¹ O, N equals oxygenated and non-oxygenated, respectively.

² 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a, b, c} Means with differing superscripts are significantly different (P < 0.05).

Table 19. The effect of dark cutter score and aging period on total plate counts of strip loin steaks on day 1 of retail display.

<i>Aging Period</i> ²	<i>Dark Cutter Score</i> ¹			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
7	2.58 ^c	2.59 ^c	2.80 ^c	2.72 ^c
14	3.39 ^b	4.09 ^a	4.37 ^a	4.42 ^a

¹ 0, 1/3, 2/3 and 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

² 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a, b, c} Means with differing superscripts are significantly different (P < 0.05).

Table 20. The effect of oxygenation treatment, enhancement treatment, dark cutter score and aging period on subjective odor of strip loin steaks after 7 days of retail display.

		<i>Oxygenation Treatment : Enhancement Treatment²</i>			
		<i>NO : E</i>	<i>NO : NE</i>	<i>O : E</i>	<i>O : NE</i>
<i>Dark Cutter Score : Aging Period³</i>	<i>0 : 7</i>	1.21 ^{hij}	1.00 ^j	1.14 ^{ij}	1.00 ^j
	<i>0 : 14</i>	2.42 ^a	1.83 ^{cde}	1.75 ^{cdef}	1.50 ^{efghi}
	<i>1/3 : 7</i>	1.29 ^{ghij}	1.29 ^{ghij}	1.62 ^{defg}	1.21 ^{hij}
	<i>1/3 : 14</i>	2.08 ^{abc}	2.00 ^{bc}	1.00 ^j	1.58 ^{defgh}
	<i>2/3 : 7</i>	1.36 ^{ghij}	1.36 ^{ghij}	1.00 ^j	1.50 ^{efghi}
	<i>2/3 : 14</i>	2.00 ^{bc}	1.58 ^{defgh}	1.58 ^{defgh}	2.33 ^{ab}
	<i>3/3 : 7</i>	1.43 ^{fghi}	1.29 ^{ghij}	1.21 ^{hij}	1.21 ^{hij}
	<i>3/3 : 14</i>	1.50 ^{efghi}	1.92 ^{cd}	1.83 ^{cde}	1.42 ^{fghi}

¹ Odor Scores: 1 = not detectable, 2 = odor activates smell but is not distinguishable, 3 = odor activates smell, is distinguishable, but not necessarily objectionable in short periods.

² Oxygenation Treatment : Enhancement Treatment; NO = Non-Oxygenated, O = Oxygenated, NE = Non-Enhanced and E = Enhanced.

³ Dark Cutter Score : Aging Period; 0 = Non-Dark Cutter, 1/3 = 1/3 Dark Cutter, 2/3 = 2/3 Dark Cutter, 3/3 = Full Dark Cutter, 7 = 7 days of age prior to treatment, 14 = 14 days of age prior to treatment.

^{a,b,c,d,e,f,g,h,i,j} Means with differing superscripts are significantly different (P < 0.05).

Table 21. The effect of oxygenation treatment, enhancement treatment and dark cutter score on oxidative properties (TBA, mg MDA/kg sample) of strip loin steaks.

Dark Cutter Score ²	Oxygenation Treatment : Enhancement Treatment ¹			
	NO : E	NO : NE	O : E	O : NE
0	0.41 ^{cd}	0.48 ^c	0.39 ^{cde}	0.65 ^b
1/3	0.35 ^{cdef}	0.84 ^a	0.23 ^{efg}	0.41 ^{cd}
2/3	0.23 ^{efg}	0.65 ^b	0.22 ^{fg}	0.28 ^{defg}
3/3	0.21 ^{fg}	0.20 ^{fg}	0.16 ^g	0.26 ^{defg}

¹ Oxygenation Treatment : Enhancement Treatment; NO = Non-Oxygenated, O = Oxygenated, NE = Non-Enhanced and E = Enhanced.

² Dark Cutter Score: 0, 1/3, 2/3, 3/3 = Non-Dark Cutter, 1/3 Dark Cutter, 2/3 Dark Cutter and Full Dark Cutter, respectively. a,b,c,d,e,f,g Means with differing superscripts are significantly different (P < 0.05).

Table 22. The effect of aging period, oxygenation treatment and retail storage on oxidative properties (TBA, mg MDA/kg sample) of strip loin steaks.

Retail Display ²	Aging Period : Oxygenation Treatment ¹		
	7 : NO	14 : NO	7 : O
1	0.16 ^g	0.15 ^g	0.18 ^{fg}
7	0.29 ^{ef}	0.64 ^{ab}	0.26 ^{efg}
14	0.57 ^b	0.70 ^a	0.43 ^{cd}
			14 : O
			0.31 ^{de}
			0.23 ^{efg}
			0.54 ^{bc}

¹ Aging Period : Oxygenation Treatment; 7, 14 = 7 and 14 days of age prior to treatment, respectively : NO = Non-Oxygenated, O = Oxygenated.

² Retail Display 1, 7, 14 = days on which measurements were taken.

a,b,c,d,e,f,g Means with differing superscripts are significantly different (P < 0.05).

Table 23. The effect of dark cutter score and retail display period on oxidative properties (TBA, mg MDA/kg sample) of strip loin steaks.

Retail Display, day	Dark Cutter Score ¹			
	0	1/3	2/3	3/3
1	0.33 ^{ef}	0.15 ^g	0.17 ^g	0.15 ^g
7	0.47 ^{cd}	0.43 ^{cde}	0.34 ^{def}	0.19 ^g
14	0.65 ^b	0.79 ^a	0.53 ^{bc}	0.27 ^{fg}

¹ 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter, and full dark cutter, respectively. a,b,c,d,e,f,g means with differing letters are significantly different (P < 0.05).

Table 24. The effect of enhancement treatment and retail display period on oxidative properties (TBA, mg MDA/kg sample) of strip loin steaks.

<i>Retail Display, days</i>	<i>Enhancement Treatment</i> ¹	
	<i>E</i>	<i>N</i>
1	0.20 ^d	0.20 ^d
7	0.26 ^d	0.46 ^b
14	0.36 ^c	0.76 ^a

¹ E and N equals Enhanced and Non-Enhanced, respectively.

^{a,b,c,d} Means within a column with differing letters are significantly different (P < 0.05).

Table 25. The effects of dark cutter score, oxygenation treatment, enhancement treatment and aging period on sensory attributes of strip loin steaks.

Response	Dark Cutter Score			Order of Means ^c	Treatment			Order of Means ^c	Enhancement			Order of Means ^c	Aging Period			Order of Means ^c		
	0 ^b	1/3 ^b	2/3 ^b		3/3 ^b	N ^e	O ^d		E ^e	N ^e	7		14	Order of Means ^c	7		14	Order of Means ^c
	(1)	(2)	(3)		(4)	(5)	(6)		(7)	(8)	(9)		(10)	(8)	(9)		(10)	(10)
Tenderness ^g	5.73 (3.21)	3.01 (0.86)	3.96 (1.56)	5.12 (0.69)	1.4.3.2	4.19 (0.94)	5.06 (0.94)	3.85 (0.94)	4.96 (1.25)	3.95 (1.39)	7.8	4.96 (1.25)	3.95 (1.39)	9.10				
Juiciness ^h	5.15 (0.20)	5.39 (0.20)	5.26 (0.20)	5.44 (0.20)	4.2.3.1	5.13 (0.14)	5.69 (0.14)	4.93 (0.14)	5.40 (0.14)	5.22 (0.14)	7.8	5.40 (0.14)	5.22 (0.14)	9.10				
Connective Tissue ⁱ	6.43 (1.27)	4.25 (0.41)	5.32 (0.64)	5.95 (0.62)	1.4.3.2	5.27 (0.57)	5.98 (0.41)	4.99 (0.41)	5.32 (0.53)	5.65 (0.61)	7.8	5.32 (0.53)	5.65 (0.61)	10.9				
Uncharacteristic Flavor ^j	2.54 (0.12)	2.65 (0.12)	2.72 (0.12)	2.88 (0.12)	4.3.2.1	2.67 (0.82)	1.96 (0.08)	3.43 (0.08)	2.78 (0.08)	2.61 (0.08)	8.7	2.78 (0.08)	2.61 (0.08)	9.10				
Overall Acceptability ^k	3.82 (0.25)	3.46 (0.25)	3.99 (0.25)	4.64 (0.25)	4.3.1.2	3.70 (0.18)	3.14 (0.18)	4.81 (0.18)	4.06 (0.18)	3.89 (0.18)	8.7	4.06 (0.18)	3.89 (0.18)	9.10				

^a These variables were not involved in significant four-way, three-way or two-way interactions between dark cutter score, treatment, enhancement, and aging period; thus, results of main effect separation are shown.

^b 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, and full dark cutter, respectively. Mean/std error of each dark cutter score for all treatment/enhancement/aging period combinations.

^c Response mean values within the same row, within dark cutter score, treatment, enhancement, and aging period, underscored by a common line do not differ significantly (P > 0.05).

^d N, O equals non-oxygenated and oxygenated, respectively. Mean/std error of each treatment for all dark cutter score/enhancement/aging period combinations.

^e E, N equals enhanced and non-enhanced, respectively. Mean/std error of each enhancement for all dark cutter score/treatment/aging period combinations.

^f 7, 14 equals 7 days and 14 days of age prior to treatment, respectively. Mean/std error of each aging period for all dark cutter score/treatment/enhancement combinations.

^g Intensity values: 1, extremely tough; 8, extremely tender.

^h Intensity values: 1, extremely dry; 8, extremely juicy.

ⁱ Intensity values: 1, abundant; 8, none.

^j Intensity values: 1, extremely uncharacteristic; 4, none.

^k Intensity values: 1, extremely undesirable; 8, extremely desirable.

Table 26. The effects of dark cutter score, oxygenation treatment, enhancement treatment and aging period on slice shear force values (kg) of strip loin steaks¹.

	Dark Cutter Score ²			Oxygenation Treatment ³			Enhancement Treatment ⁴		Aging Period ⁵	
	0	1/3	2/3	3/3	O	N	E	N	7	14
<i>Slice Shear Force (kg)</i>	17.3 ^{bc}	23.9 ^a	21.5 ^a	15.6 ^c	20.2 ^a	18.9 ^a	17.5 ^b	21.7 ^a	19.5 ^a	19.6 ^a
<i>Standard Error</i>	1.37	1.37	1.34	1.42	0.98	0.98	1.16	0.74	0.98	0.98

¹ These variables were not involved in any significant interactions, therefore results of main effect separation are shown.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ O, N equals oxygenated and non-oxygenated, respectively.

⁴ E, N equals enhanced and non-enhanced, respectively.

⁵ 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

a, b, c Means with differing superscripts within a treatment are significantly different (P < 0.05).

Table 27. The effect of dark cutter score and retail display period on headspace (% Oxygen) of strip loin steaks packaged in a modified atmosphere of 80% Oxygen / 20 % Carbon Dioxide.

<i>Retail Display Period²</i>	<i>Dark Cutter Score¹</i>			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>7</i>	73.01 ^{bc}	73.99 ^{bc}	73.01 ^{bc}	72.90 ^c
<i>14</i>	75.38 ^a	74.21 ^{ab}	71.53 ^d	70.70 ^d

¹ Dark Cutter Scores 0, 1/3, 2/3, 3/3 equals Non-Dark Cutter, 1/3 Dark Cutter, 2/3 Dark Cutter and Full Dark Cutter, respectively.

² Retail Display Period: 7, 14 equals 7 days and 14 days of retail display, respectively.

^{a,b,c,d} Means within a row with differing superscripts are significantly different (P < 0.05.)

Table 28. The effect of aging period and retail display period on headspace (% Oxygen) of strip loin steaks packaged in a modified atmosphere of 80% Oxygen / 20 % Carbon Dioxide.

<i>Retail Display Period</i> ²	<i>Aging Period</i> ¹	
	7	14
7	72.61 ^b	73.84 ^a
14	74.15 ^a	71.76 ^b

¹ Aging Period: 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

² Retail Display Period: 7, 14 equals 7 days and 14 days of retail display, respectively.

^{a,b} Means within a row with differing superscripts are significantly different (P < 0.05).

Table 29. The effect of oxygenation treatment, enhancement treatment and aging period on drip loss (% of original weight) of strip loin steaks.

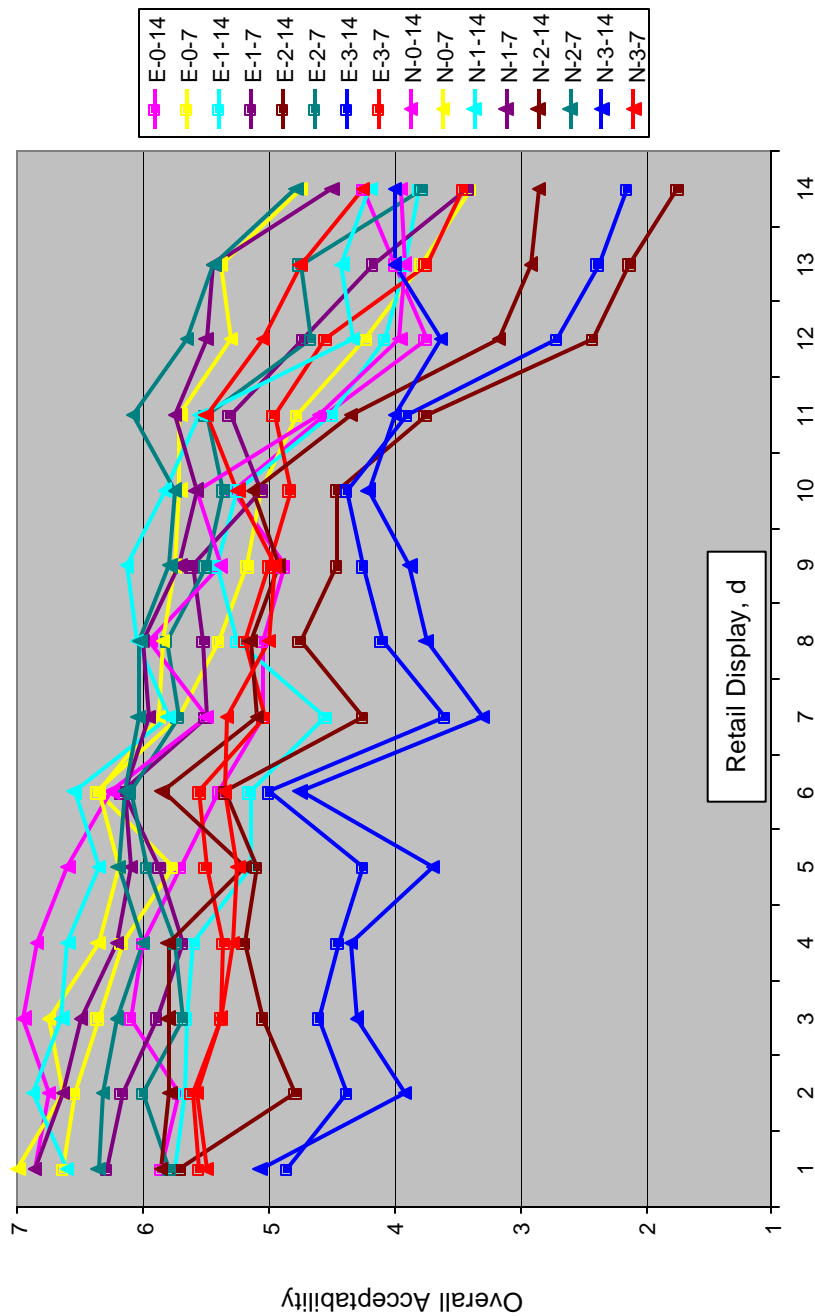
<i>Aging Period</i> ²	<i>Oxygenation Treatment : Enhancement Treatment</i> ¹			
	<i>NO : NE</i>	<i>NO : E</i>	<i>O : E</i>	<i>O : NE</i>
7	2.52 ^d	3.03 ^c	2.44 ^d	2.81 ^{cd}
14	9.89 ^b	9.70 ^b	10.41 ^a	9.62 ^b

¹ Oxygenation Treatment : Enhancement Treatment; NO = Non-Oxygenated, O = Oxygenated, NE = Non-Enhanced and E = Enhanced.

² Aging Period: 7, 14 equals 7 days and 14 days, respectively.

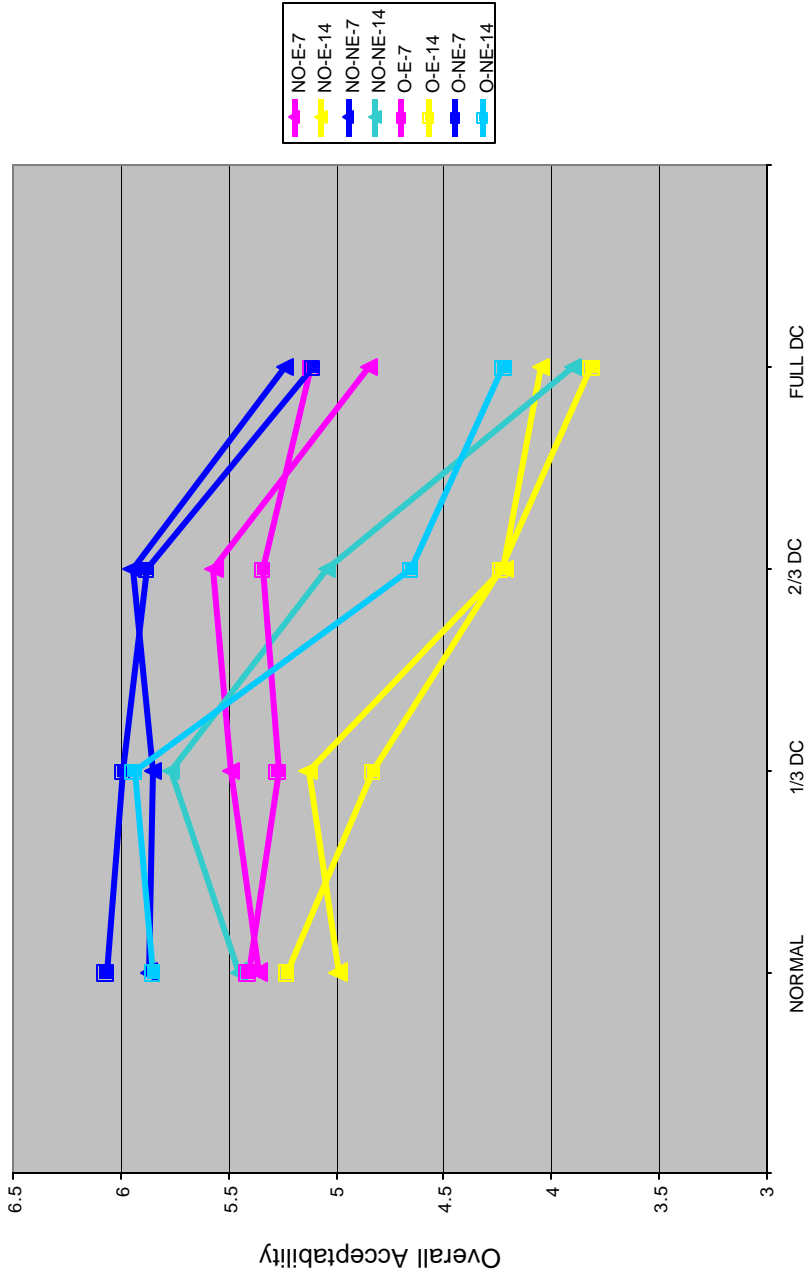
^{a,b,c,d} Means with differing superscripts are significantly different (P < 0.05).

Figure 1. The effect of enhancement treatment¹, dark cutter score², aging period³ and retail display day⁴ on overall acceptability⁵ of strip loin steaks.



¹ Enhancement Treatment: E=enhanced, N=non-enhanced.
² Dark Cutter Score: 0=normal, 1=1/3 dark cutter, 2=2/3 dark cutter, 3=full dark cutter.
³ Aging Period: 7=7 d, 14=14 d.
⁴ Retail Display: Steaks were assessed daily for a 14 day period.
⁵ Overall Acceptability: 7=extremely desirable, 6=desirable, 5=slightly desirable, 4=acceptable, 3=slightly undesirable, 2=undesirable, 1= extremely undesirable.

Figure 2. The effect of oxygenation treatment¹, enhancement treatment², aging period³ and dark cutter score⁴ on overall acceptability⁵ of strip loin steaks.



¹ Oxygenation Treatment: O=oxygenated, NO=non-oxygenated.

² Enhancement Treatment: E=enhanced, NE=non-enhanced.

³ Aging Period: 7=7 d, 14=14 d.

⁴ Dark Cutter Score: 0=normal, 1=1/3 dark cutter, 2=2/3 dark cutter, 3=full dark cutter.

⁵ Overall Acceptability: 7=extremely desirable, 6=desirable, 5=slightly desirable, 4=acceptable, 3=slightly undesirable, 2=undesirable, 1=extremely undesirable.

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APPENDIX

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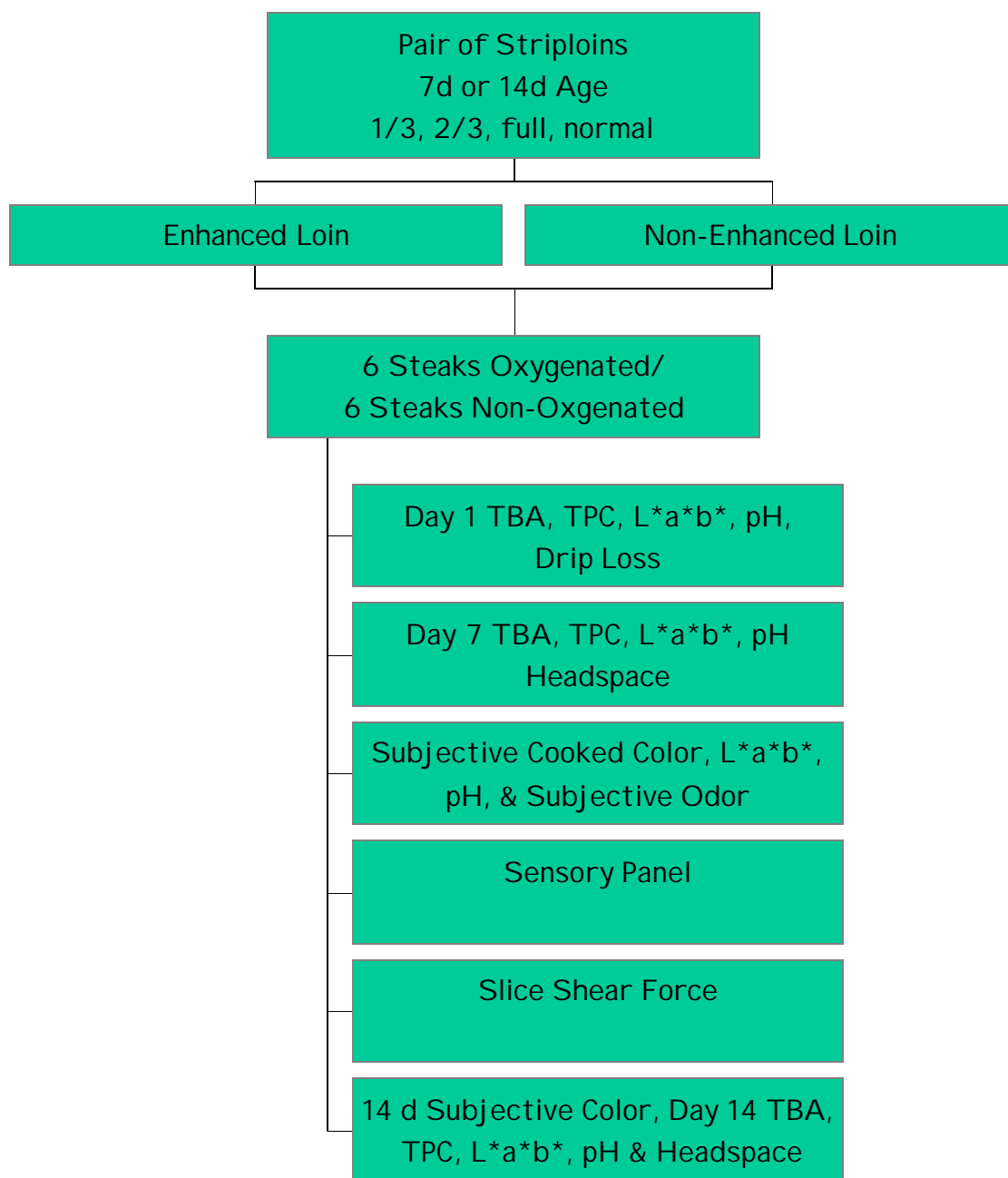
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Appendix A. Project Design.



Appendix B. Subjective Color Scale.

LEAN COLOR

8. Bright Cherry-Red
7. Moderately Bright Cherry-Red
6. Cherry-Red
5. Slightly Dark Red
4. Moderately Dark Red or Brown
3. Dark Red or Brown
2. Very Dark Brown
1. Extremely Dark Brown

% DISCOLORATION

7. None
6. 1-10
5. 11-25
4. 26-50
3. 51-75
2. 76-99
1. Complete

FAT COLOR

8. Creamy White
7. Mostly Creamy White
6. Slightly Tan
5. Tan
4. Slightly Brown
3. Moderately Brown
2. Brown or Slightly Green
1. Dark Brown or Green

OVERALL APPEARANCE

7. Extremely Desirable
6. Desirable
5. Slightly Desirable
4. Acceptable
3. Slightly Undesirable
2. Undesirable
1. Extremely Undesirable

Appendix C. Subjective Cooked Color Scale.



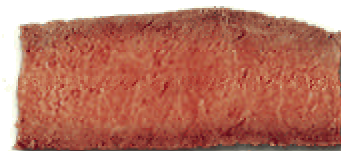
Very Rare
Approx. 130°F, 55°C



Rare
Approx. 140°F, 60°C
(Center is bright red, pinkish toward the exterior portion.)



Medium Rare
Approx. 145°F, 63°C
(Center is very pink, slightly brown toward the exterior portion.)



Medium
Approx. 160°F, 71°C
(Center is light pink, outer portion is brown.)



Well Done
Approx. 170°F, 77°C
(Steak is uniformly brown throughout.)



Very Well Done
Approx. 180°F, 82°C

Appendix D. Subjective Odor Scale.

OBJECTIONABLE CATEGORIES 1. Odor not detectable

2. Odor present, which activates smell, but is not distinguishable

3. Odor present, which activates smell, is distinguishable, not necessarily objectionable in short period

4. Odor present, which easily activates smell, is very distinct and may be objectionable

5. Odor present, is objectionable and may cause a person to avoid completely, could cause physiological effects.

6. Odor present, which is strong, overpowering, intolerable and easily produces physiological effects.

Appendix E. Sensory Ballet.

**DARK CUTTER PROJECT
SENSORY BALLOT**

NAME _____ BOOTH # _____ DATE: _____

TIME: _____

Sample	Tenderness	Juiciness	Connective Tissue Amount	Uncharacteristic Flavor	Overall Acceptability	Comments
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						
13						
14						

Tenderness

- 8 Extremely tender
- 7 Very tender
- 6 Moderately tender
- 5 Slightly tender
- 4 Slightly tough
- 3 Moderately tough
- 2 Very tough
- 1 Extremely tough

Juiciness

- 8 Extremely juicy
- 7 Very juicy
- 6 Moderately juicy
- 5 Slightly juicy
- 4 Slightly dry
- 3 Moderately dry
- 2 Very dry
- 1 Extremely dry

Overall Acceptability

- 8 Extremely desirable
- 7 Moderately desirable
- 6 Desirable
- 5 Slightly desirable
- 4 Slightly undesirable
- 3 Undesirable
- 2 Moderately undesirable
- 1 Extremely undesirable

Connective Tissue Amount

- 8 None
- 7 Practically none
- 6 Traces
- 5 Slight
- 4 Moderate
- 3 Slightly Abundant
- 2 Moderately Abundant
- 1 Abundant

Uncharacteristic Flavor

- 4 None
- 3 Slight
- 2 Moderate
- 1 Extremely uncharacteristic

Table A. The effects of oxygenation treatment and aging period on L^{*1} values of strip loin steaks after 7 days of retail display.

	<i>Oxygenation Treatment</i> ²		<i>Aging Period</i> ³	
	<i>O</i>	<i>NO</i>	<i>7</i>	<i>14</i>
L^{*1}	34.31 ^a	33.74 ^a	33.28 ^b	34.78 ^a
<i>Standard Error</i>	0.42	0.42	0.42	0.42

¹ 0 = black, 100 = white.

² Oxygenation Treatment: O = Oxygenated, NO = Non-Oxygenated.

³ Aging Period: 7, 14 = 7 and 14 days of age prior to treatment, respectively.

^{a,b} Means within a treatment with differing superscripts are significantly different ($P < 0.05$).

Table B. The effect of enhancement treatment, aging period and dark cutter score on L*¹ values of strip loin steaks on day 14 of retail display.

Dark Cutter Score ³	Enhancement Treatment : Aging Period ²			
	E : 7	E : 14	NE : 7	NE : 14
0	43.11 ^a	37.28 ^{bc}	42.30 ^a	42.37 ^a
1/3	35.27 ^{bcd}	37.80 ^b	34.39 ^{cd}	36.89 ^{bc}
2/3	35.34 ^{bcd}	33.67 ^{de}	32.94 ^{de}	33.12 ^{de}
3/3	33.53 ^{de}	35.42 ^{bcd}	31.07 ^e	24.74 ^f

¹ 0 = black, 100 = white.

² Enhancement Treatment : Aging Period; E = Enhanced, NE = Non-Enhanced, 7 = 7 days of age prior to treatment, 14 = 14 days of age prior to treatment.

³ 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

^{a,b,c,d,e,f} Means with differing superscripts are significantly different (P < 0.05).

Table C. The effect of dark cutter score and enhancement treatment on a*¹ values of strip loin steaks on day 7 of retail storage.

<i>Enhancement Treatment</i> ³	<i>Dark Cutter Score</i> ²			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>E</i>	21.92 ^b	23.71 ^a	24.39 ^a	23.53 ^a
<i>NE</i>	24.37 ^a	23.77 ^a	24.59 ^a	19.76 ^c

¹ Negative values = green, positive values = red.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ E, N equals enhanced and non-enhanced, respectively.

^{a,b,c} Means with differing superscripts are significantly different (P < 0.05).

Table D. The effect of dark cutter score and aging period on a*¹ values of strip loin steaks on day 7 of retail display.

<i>Aging Period</i> ³	<i>Dark Cutter Score</i> ²			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
7	24.47 ^a	24.09 ^{ab}	23.30 ^b	21.91 ^c
14	20.89 ^c	24.55 ^a	24.54 ^a	22.04 ^c

¹ Negative values = green, positive values = red.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ Aging Period: 7, 14 = 7 and 14 days of age prior to treatment, respectively.

^{a,b,c} Means with differing superscripts are significantly different (P < 0.05).

Table E. The effect of oxygenation treatment on a^{*1} values of strip loin steaks after 7 days of retail display.

	<i>Oxygenation Treatment</i> ²	
	<i>O</i>	<i>NO</i>
a^{*1}	22.57 ^b	23.88 ^a
<i>Standard Error</i>	0.29	0.29

¹ Negative values = green, positive values = red.

² Oxygenation Treatment: O = Oxygenated, NO = Non-Oxygenated.

^{a,b} Means with differing superscripts are significantly different ($P < 0.05$).

Table F. The effects of oxygenation treatment and aging period on a^{*1} values of strip loin steaks after 14 days of retail display.

	<i>Oxygenation Treatment</i> ²		<i>Aging Period</i> ³	
	<i>O</i>	<i>NO</i>	<i>7</i>	<i>14</i>
a^{*1}	18.57 ^a	19.30 ^a	20.78 ^a	17.09 ^b
<i>Standard Error</i>	0.46	0.46	0.46	0.46

¹ Negative values = green, positive values = red.

² O, N equals oxygenated and non-oxygenated, respectively.

³ 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a,b} Means with differing superscripts within a treatment are significantly different ($P < 0.05$).

Table G. The effect of dark cutter score and enhancement treatment on b^* ¹ values of strip loin steaks on day 1 of retail display.

<i>Enhancement Treatment</i> ³	<i>Dark Cutter Score</i> ²			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>E</i>	19.33 ^{ab}	17.91 ^{cd}	18.21 ^{bcd}	18.34 ^{bcd}
<i>NE</i>	19.73 ^a	18.97 ^{abc}	17.46 ^d	14.38 ^e

¹ Negative values = blue, positive values = yellow.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ E, N equals enhanced and non-enhanced, respectively.

^{a,b,c,d,e} Means with differing superscripts are significantly different ($P < 0.05$).

Table H. The effect of aging period and enhancement treatment on b^{*1} values of strip loin steaks on day 7 of retail display.

<i>Enhancement Treatment</i> ³	<i>Aging Period</i> ²	
	<i>7</i>	<i>14</i>
<i>E</i>	22.29 ^a	19.68 ^c
<i>NE</i>	20.81 ^b	18.26 ^d

¹ Negative values = blue, positive values = yellow.

² 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

³ E, N equals enhanced and non-enhanced, respectively.

^{a,b,c,d} Means with differing superscripts are significantly different ($P < 0.05$).

Table I. The effect of dark cutter score and enhancement treatment on b^* ¹ values of strip loin steaks on day 14 of retail display.

<i>Enhancement Treatment</i> ³	<i>Dark Cutter Score</i> ²			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
<i>E</i>	17.31 ^{bcd}	17.38 ^{bcd}	16.41 ^d	16.35 ^d
<i>NE</i>	18.64 ^{ab}	19.23 ^a	17.95 ^{abc}	16.72 ^{cd}

¹ Negative values = blue, positive values = yellow.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ E, N equals enhanced and non-enhanced, respectively.

^{a,b,c,d} Means with differing superscripts are significantly different ($P < 0.05$).

Table J. The effect of dark cutter score and aging period on b^* ¹ values of strip loin steaks on day 14 of retail display.

<i>Aging Period</i> ³	<i>Dark Cutter Score</i> ²			
	<i>0</i>	<i>1/3</i>	<i>2/3</i>	<i>3/3</i>
7	17.74 ^{bc}	18.81 ^{ab}	19.11 ^a	17.99 ^{abc}
14	16.89 ^{cd}	19.11 ^a	16.01 ^d	16.46 ^d

¹ Negative values = blue, positive values = yellow.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ Aging Period: 7, 14 = 7 and 14 days of age prior to treatment, respectively.
^{a,b,c,d} Means with differing superscripts are significantly different ($P < 0.05$).

Table K. The effect of oxygenation treatment, enhancement treatment, dark cutter score and aging period on b*¹ values of cooked (70° C) strip loin steaks after 7 days of retail display.

<i>Oxygenation Treatment : Enhancement Treatment²</i>				
	<i>NO : E</i>	<i>NO : NE</i>	<i>O : E</i>	<i>O : NE</i>
<i>0 : 7</i>	15.71 ^{ijkl}	18.32 ^{ghij}	14.71 ^{kl}	20.14 ^{efg}
<i>0 : 14</i>	15.46 ^{kl}	15.31 ^{kl}	14.77 ^{kl}	14.98 ^{kl}
<i>1/3 : 7</i>	21.07 ^{def}	25.02 ^{ab}	16.78 ^{hijkl}	22.97 ^{bcde}
<i>1/3 : 14</i>	17.44 ^{ghijk}	20.21 ^{efg}	14.78 ^{kl}	15.77 ^{ijkl}
<i>2/3 : 7</i>	25.88 ^{ab}	24.24 ^{bc}	13.94 ^l	24.22 ^{bc}
<i>2/3 : 14</i>	18.62 ^{ghi}	25.81 ^{ab}	17.19 ^{ghijk}	20.79 ^{def}
<i>3/3 : 7</i>	15.96 ^{ijkl}	27.54 ^a	21.26 ^{cdef}	24.92 ^{ab}
<i>3/3 : 14</i>	20.74 ^{def}	23.60 ^{bcd}	19.26 ^{fgh}	24.62 ^{ab}

Dark Cutter Score : Aging Period³

¹ Negative values = blue, positive values = yellow.

² Oxygenation Treatment : Enhancement Treatment; NO = Non-Oxygenated, O = Oxygenated, NE = Non-Enhanced and E = Enhanced.

³ Dark Cutter Score : Aging Period; 0 = Non-Dark Cutter, 1/3 = 1/3 Dark Cutter, 2/3 = 2/3 Dark Cutter, 3/3 = Full Dark Cutter, 7 = 7 days of age prior to treatment, 14 = 14 days of age prior to treatment.

^{a,b,c,d,e,f,g,h,i,j,k,l} Means with differing superscripts are significantly different (P < 0.05).

Table L. The effects of dark cutter score, oxygenation treatment, enhancement treatment and aging period on total plate counts of strip loin steaks on day 7 of retail display¹.

	Dark Cutter Score ²			Oxygenation Treatment ³			Enhancement Treatment ⁴			Aging Period ⁵	
	0	1/3	2/3	3/3	O	N	E	N	7	14	
Total Plate Count	3.93 ^c	5.26 ^{ab}	5.13 ^b	5.63 ^a	5.01 ^a	4.97 ^a	5.07 ^a	4.91 ^a	4.18 ^b	5.79 ^a	
Standard Error	0.22	0.22	0.21	0.23	0.16	0.16	0.18	0.12	0.16	0.16	

¹ These variables were not involved in significant interactions, therefore results of main effect separation are shown.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ O, N equals oxygenated and non-oxygenated, respectively.

⁴ E, N equals enhanced and non-enhanced, respectively.

⁵ 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a,b,c} Means with differing superscripts are significantly different ($P < 0.05$).

Table M. The effects of dark cutter score, oxygenation treatment, enhancement treatment and aging period on total plate counts of strip loin steaks on day 14 of retail display¹.

	Dark Cutter Score ²			Oxygenation Treatment ³			Enhancement Treatment ⁴			Aging Period ⁵	
	0	1/3	2/3	3/3	O	N	E	N	7	14	
Total Plate Count	4.87 ^b	5.90 ^a	6.11 ^a	6.17 ^a	5.64 ^a	5.89 ^a	5.93 ^a	5.60 ^b	5.83 ^a	5.70 ^a	
Standard Error	0.14	0.14	0.14	0.14	0.10	0.10	0.12	0.08	0.10	0.10	

¹ These variables were not involved in significant interactions, therefore results of main effect separation are shown.

² 0, 1/3, 2/3, 3/3 equals non-dark cutter, 1/3 dark cutter, 2/3 dark cutter and full dark cutter, respectively.

³ O, N equals oxygenated and non-oxygenated, respectively.

⁴ E, N equals enhanced and non-enhanced, respectively.

⁵ 7, 14 equals 7 days and 14 days of age prior to treatment, respectively.

^{a,b} Means with differing superscripts are significantly different (P < 0.05).

VITA

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