

BEEF MUSCLE COLOR AS AFFECTED BY NUTRITIONAL REGIME AND VACUUM PACKAGING

by

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CHAPTER 1

GENERAL INTRODUCTION

Visual appearance is the first sensory perception of a prospective meat customer upon viewing the supermarket meat display counter. At purchase, freshness of product is of extreme importance to the customer. Juiciness, tenderness, flavor and overall acceptability are important as well, but these product traits are evaluated at consumption. Freshness, however, is evaluated before purchase and is therefore extremely important to beef sales. Bright color has long been associated with meat freshness. Because of this association, the consumer discriminates against dark colored beef and will buy it only at a discounted price.

The fresh meat retailer desires beef that not only brightens rapidly after cutting, but will retain a bright color during normal display time of 1 to 3 days. Retailers, like consumers, discriminate against suppliers whose beef does not brighten rapidly after cutting or maintain an adequate display color stability.

Considerable interest has developed in researching methods of minimizing production and processing costs of beef. Reduced periods of grain finishing and/or increased levels of forages in finishing rations have resulted from increased grain costs in recent years. The shift from long feeding periods on high concentrate rations has been encouraged by changes in the U.S.D.A. quality grading standards for beef and consumer preference for beef with lower fat levels.

Cost reductions for the processor are being sought through lower energy consumption and centralized processing. Aging beef at higher temperatures will reduce refrigeration costs and decrease energy use. Vacuum packaging of beef not only reduces transportation costs, but gives the seller greater inventory flexibility and lowers labor costs.

The primary purpose of this study was to evaluate muscle color differences that may result from changes in production and processing methods. Color and shelf life comparisons and interactions were studied among four nutritional regimes, two post-slaughter conditioning periods and two packaging methods using four muscles.

CHAPTER 2

REVIEW OF LITERATURE

Pigment chemistry

Color of beef muscle is due primarily to the amount and chemical state of two heme pigments, myoglobin and hemoglobin. Each myoglobin molecule is composed of a globin protein portion and a non-protein called heme which is composed of an iron atom and a porphyrin ring (Lemberg and Legge, 1949; Schweigert, 1954).

In the live animal, myoglobin accepts oxygen from blood hemoglobin for use in oxidative energy yielding reactions in the cell (Schweigert, 1954). This is essentially an oxygen storage mechanism in the cell (Fox, 1968). Shenk, Hall and King (1934) stated the storage role is reflected in the quantities of myoglobin found in various tissues. Giffie et al. (1960) define the quantities of heme pigments as functions of (1) the amount of use of the "principally muscular-activity tissue", (2) the blood supply, (3) the oxygen availability and (4) the age of the animal.

The iron atom has six bond orbitals in which it shares two electrons with another atom. Five electron pairs are often shared with nitrogen atoms and one with oxygen. The bond type formed with the oxygen bond orbital and other chemicals is the most important factor in determining the final color of the complex. Oxidative state of the iron and physical state of the globin portions of the myoglobin molecule also are important in determining the final color (Lawrie, 1966; Bodwell and McClain, 1971).

In the presence of oxygen, reduced myoglobin (Mb, causes the purplish-red color of unbloomed meat) is reversibly converted to oxymyoglobin (MbO_2 , causes the bright red color desired by consumers) or metmyoglobin (Mb^+ , causes the brown color of old meat). Iron exists as Fe^{++} while part of Mb and MbO_2 , but is converted to Fe^{+++} in Mb^+ (Lawrie, 1966).

Schweigert (1956), during a review of chemical reactions of Mb, concluded: (1) Oxygen is required for formation of MbO_2 from Mb; however, prolonged oxygen exposure results in formation of Mb^+ . (2) Reducing conditions keep Mb in the reduced state and can convert Mb^+ to Mb. (3) Once oxidation proceeds to formation of green compounds, the reactions are irreversible.

Since pigment color changes result from combination of Mb and oxygen, the partial pressure of oxygen ($p\text{O}_2$) determines relative proportions of MbO_2 and Mb^+ . Thus, in fresh meat, the bright red color of MbO_2 is dependent on a plentiful supply of oxygen and reducing substrates. Mb exists within the interior of fresh meat as long as the supply of oxidizable substrates last, but will be oxidized to Mb^+ once the reducing power of the muscle is lost (Brooks, 1929; 1938). At low $p\text{O}_2$ (4 mm Hg (Brooks, 1938); 1-11.4 mm Hg (George and Stratmann, 1952); 30% Mb^+ in 30 minutes, packaged in O_2 impermeable film (Zimmerman and Snyder, 1969); and 1% O_2 (Ledward and Macfarlane, 1971)), autoxidation of Mb to Mb^+ is reported to be maximal. Solberg (1968) stated $p\text{O}_2$ increases above these low levels slowed autoxidation and increased MbO_2 stability. George and

Stratmann (1952) reported that at 30°C, pO_2 greater than 30 mm Hg failed to influence formation rate of Mb^+ .

Mb^+ formation was accelerated as temperature increased above freezing and pH decreased from 7.0 to 5.0 (Brown and Mebine, 1969). In addition, Mb^+ formation is catalyzed by products of polyunsaturated fatty acid oxidation, particularly malonaldehyde (Ledward and Macfarlane, 1971; Greene, 1969; and Hutchins, Liu and Watts, 1967).

Archer and Bandfield (1950), using veal loaf, compared tungsten and fluorescent lights and observed time required for the least visually perceptible discoloration to appear. Under light, muscle pigments faded and absorbed light energy apparently decomposed the pigment. Shorter wavelengths (4000 Å^o or less) caused faster discoloration than longer wavelengths. The authors theorized blue and green lights would cause the greatest destruction of heme pigments since their strongest absorption bands in the visible spectrum lie in this region (Archer and Bandfield, 1950). Marriott et al. (1967) found fresh beef stored in complete darkness retained desirable color longer than beef under lighted display. Hansen and Sereika (1969) and Santamaria (1970) indicated shortening of display life with increased light intensity. Schweigert (1956) pointed out light was catalytic to irreversible oxidation reactions yielding "green compounds".

Muscle color measurement by absorbance and reflectance

Firko and Ayres (1957) plotted absorption maxima at 580 and 635 nm and a minimum at 555 nm and related these to changes

in MbO_2 , Mb^+ and Mb. The value at 555 nm was equated to Mb, 580 nm to MbO_2 and 635 nm to Mb^+ .

Broumand, Ball and Stier (1958) spectrophotometrically determined myoglobin derivatives in aqueous extracts of muscle. The ratio 507 nm/573 nm was used to estimate percent Mb^+ and 473 nm/597 nm to estimate percent Mb. Percent MbO_2 was calculated by subtracting percent Mb^+ and Mb from 100.

Dean and Ball (1960) suggested Broumand's method may give erroneous values since Mb may be converted to MbO_2 when muscle pigment is extracted in water. Broumand assumed MbO_2 and Mb^+ to have equal solubility in water, a questionable assumption. Naughton, Zeitlin and Frodyma (1958), using tuna muscle, found methemoglobin and oxyhemoglobin quite different in solubility, a possibility that may exist for myoglobin. Dean and Ball (1960) proposed a method based upon Broumand's, but depending upon reflectance instead of absorption. The authors felt reflectance would give more reliable results than absorbance or transmittance because the meat surface could be measured directly.

Snyder (1965) used suspensions of myoglobin chemical forms in non-fat dried milk to plot standard reflectance curves for Mb, MbO_2 and Mb^+ . The author found the major problems with absorbance to be: (1) obtaining clear solutions when extracting muscle pigments, (2) selecting surface volume (especially depth from the surface) to be analyzed, (3) changes in chemical form of muscle pigment during the extraction process and (4) sample destruction during extraction and handling.

Snyder (1965) used R_A values (absorbance values on a log scale considering reflectance equal to transmittance) from the reflectance spectra after R_A had been adjusted to 1.0 at 525 nm, an isobestic point for Mb, MbO₂ and Mb⁺. Pigment concentration was measured by plotting R_A at 473 nm for Mb⁺ and MbO₂ and at 571 nm for Mb and MbO₂. The properties of the myoglobin forms were calculated from the plots.

Stewart, Zipser and Matts (1965) observed K/S (K=absorption coefficient, S=scattering coefficient) ratios from reflectance data were quite different from ratios of absorbancy coefficients at the same wavelengths calculated from transmission data. Snyder and Armstrong (1967) found that with water suspensions of Mb⁺ or MbO₂ and non-fat dry milk K/S 571 nm accurately estimated Mb⁺; however, the authors stressed that for intact muscle, ratios of K/S values were preferable.

Dean and Ball (1960) found reflectance gave truer values than absorbance of percent of myoglobin chemical forms on the muscle surface. Measurement of unbloomed muscle (Mb assumed to be the predominant chemical form of myoglobin) by reflectance showed Mb constituted 50% and Mb⁺ was 10% of the total pigment, whereas absorbancy ratios suggested 20% Mb and 20% Mb⁺.

Reflectance curves obtained by Ginger, Wilson and Schweigert (1964) with a Coleman Junior spectrophotometer confirmed discoloration observed visually, but absorption curves from pigment extracts of similar samples failed to relate to pigment changes on the muscle surface.

Firko and Ayres (1957) discovered a rapid decrease in percent reflectance (R) at 635 nm between 0 and 4 hours post-cutting for biceps femoris and semitendinosus muscles. The typical scarlet-red color "associated with presence of free O_2 " was lost after 3 to 6 days and replaced by the brown color typical of old meat.

Kropf et al. (1974) reported at each day (comparing 0, 1, 3, 7, 14 and 42 days) of lighted display, %R 630, 635 and 650 nm had the highest correlations of single wavelength %R with subjective scores for longissimus and psoas major muscles. Of 7 ratios of %R at selected wavelengths (507 nm/572 nm, 474 nm/597 nm, 474 nm/525 nm, 572 nm/525 nm, 582 nm/525 nm, 630 nm/525 nm, 635 nm/525 nm), 474 nm/597 nm and 474 nm/525 nm exhibited the least value for predicting subjective color score in either muscle at any time. Correlations of ratios 582 nm/525 nm, 507 nm/572 nm and 572 nm/525 nm with subjective color scores were approximately equal in predictive value at all times after 3 days display, but 582 nm/525 nm was a better predictor for most time periods.

Ockerman and Cahill (1969) evaluated 150 longissimus samples with as much color variation as possible visually (five member panel) and with a B & L 20 spectrophotometer at 415, 445, 475, 485, 505, 535, 565, 595, 625, 655 and 685 nm. Multiple regression analysis showed %R 685 nm to be the simplest method of estimating visual score. The correlation coefficient of %R at 685 nm to visual score was over .85.

Dean and Ball (1960) used K/S 507nm/K/S 573 nm to estimate proportion of Mb⁺ and K/S 473 nm/K/S 597 nm for Mb. Proportion of MbO₂ was calculated by difference.

Snyder and Armstrong (1967) compared reflectance measured on the absorbance scale with K/S values. K/S values were best suited for quantitative analysis of myoglobin derivatives since they were linear in relation to concentration whereas raw reflectance or R_A was nonlinear. Adjustment of spectra to eliminate scatter and compensate for various pigment levels as suggested by Snyder (1965) was unnecessary with K/S ratios.

Stewart et al. (1965) used K/S 525 nm to estimate total pigment concentration as a linear relationship vs. a nonlinear relationship using reflectance measured on the absorbance scale. K/S 572 nm/K/S 525 nm also produced a straight line when plotted against total pigment.

Zimmerman and Snyder (1969) used K/S ratios to estimate proportions of pigment forms. For Mb⁺, K/S 571 nm/K/S 525 nm was .59. For MbO₂, K/S 571 nm/K/S 525 nm was 1.36 and K/S 474 nm/K/S 525 nm was .88. For Mb, K/S 474 nm/K/S 525 nm was .53.

Kropf et al. (1974) showed uncorrected (raw) reflectance ratios or those adjusted for K/S using wavelengths 474 nm/525 nm or 474 nm/597 nm appear to distinguish Mb from MbO₂ and Mb⁺ as does K/S 474 nm. K/S 600 nm appeared to distinguish MbO₂. K/S ratios at 572, 630 or 650 nm seemed capable of distinguishing Mb⁺ as did the unadjusted reflectance ratio or K/S 572 nm/K/S 525 nm.

Satterlee and Hansmeyer (1974) used K/S 580 nm to estimate percent MbO_2 on the surface of beef muscle.

Allen et al. (1969) determined reflectance scores on rib steaks 2.54 cm thick at 26 intervals from 0 to 240 hours post-cutting. A significant decrease in reflectance between 0 and 5 minutes was observed at 474, 525, 538, 568 and 571 nm and $\%R$ 474 nm/ $\%R$ 525 nm. Reflectance at 525, 538, 568 and 571 nm were generally insensitive to color deterioration. A gradual decrease in reflectance at 600, 610, 620 and 630 nm occurred as color deteriorated. $\%R$ 474 nm/ $\%R$ 525 nm tended to decline as color brightened and increased as color deteriorated and seemed more useful in following muscle color changes than $\%R$ 571 nm/ $\%R$ 525 nm.

Santamaria (1970) stated that unadjusted reflectance values taken at less than 600 nm were insensitive to color deterioration in frozen beef muscle and reflectance values from 600 to 650 nm were useful in detecting color deterioration.

Hansen and Sereika (1969) determined $\%R$ 582 nm/ $\%R$ 525 nm indicated proportion of MbO_2 on muscle surface and $\%R$ 630 nm/ $\%R$ 525 nm indicated proportion of Mb^+ . The authors defined acceptable muscle color as a value of 1.12 or more for $\%R$ 582 nm/ $\%R$ 525 nm and/or less than .55 for $\%R$ 630 nm/ $\%R$ 525 nm.

Eagerman, Clydesdale and Francis (1974) used $\%R$ 632 nm minus $\%R$ 614 nm to report proportions of Mb , MbO_2 and Mb^+ . The difference was greatest for Mb , intermediate for MbO_2 and least or negative for Mb^+ .

Franke and Solberg (1971), using reflectance scans made with a Beckman DK-2 spectrophotometer, discovered peak height at 632 nm decreased as proportion of Mb^+ increased. An increase in peak height at 632 nm denoted an increase in proportion of Mb^+ and a decrease in proportion of MbO_2 on the muscle surface.

Snyder (1964), using a Gardner automatic color difference meter, reported the "a" value (a mathematical representation involving a weighted integration of reflectance primarily from the red region of the spectrum) alone made it difficult to tell whether myoglobin in a sample was oxidized or reduced. The author found it convenient to plot a/b (a measure of hue) ratios as well as "a" values since a/b decreased "considerably" with conversion of MbO_2 or Mb to Mb^+ .

Strange et al. (1974) measured reflectance of beef semitendinosus muscle with a Beckman D.U. Model 2400 spectrophotometer at 520, 540, 560, 580, 600, 620, 630, 640, 650 and 700 nm. Difference measurements were taken with a Gardner color difference meter. The "a" value was highly correlated ($r=.91$) with panel score, H (50 point scale with 50=extremely acceptable, 10 panelists). The authors correlated %R 580 nm (minimum for MbO_2), %R 630 nm (minimum for Mb^+) and %R 700 nm (highest reflectance found for muscle in visible region) with H. The low coefficient for H with %R 700 nm indicated reflectance at this point was independent of both Mb^+ and MbO_2 concentrations. Plots of H with %R 580 nm and %R 630 nm suggested a decrease in MbO_2 and as increase in Mb^+ , respectively, with display time. Linear correlation coefficients for any %R with H were "significantly lower" than for Gardner "a" with H. Higher linear

correlation coefficients for $\%R$ with Gardner "a" were achieved than for $\%R$ with H. The higher coefficients were "probably due to the objectivity of the instruments versus the subjectivity of the panel". The authors stated Gardner "a" value is an accurate replacement for hedonic scoring.

Jeremiah, Carpenter and Smith (1972a) compared several instruments used for objectively measuring muscle color. Using standard partial regression coefficients and coefficients of determination, the authors found visual score can be predicted with almost equal accuracy using the Macbeth-Munsell, Gardner or Bausch and Lomb 20 systems. The Photovolt 610 RM system accounted for 13% less variation in visual scores than other systems. Gardner "b" (measure of hue) was twice as important as "a" and six times more than R_d (measure of brightness). Of B and L reflectance measurements at single wavelengths, those at 625 and 655 nm were most closely related to visual scores. Red and black Munsell readings were relatively more important than white and yellow readings in accounting for differences.

Comparison of muscle color from beef fed various levels of forage

Longwell (1936) found spectrophotometric measurement of longissimus muscle from steers fed grass alone; grass alone for 56 days, then grain on grass for 84 days; grain on grass for 140 days; grass alone for 140 days, then grain on grass for 56 days; or grass alone for 140 days, then grain in drylot for 56 days had no effect on muscle color. The author concluded (1) grass as a feed does not produce dark beef and (2) muscle color of grass finished beef will be as bright as muscle from

grain finished cattle which have similar finish. This agrees with McCampbell and Mackintosh (1927), Mackintosh and Hall (1935), Bray and U.S.D.A. workers (1937), Mackintosh, Latshaw and Kramer (1937), Brown (1954), McCampbell et al. (1960) and Malphrus et al. (1962), all who reported no effect of feeding grain in drylot, grain on pasture or pasture only on muscle color.

Kropf, Allen and Thouvenelle (1975) visually evaluated muscle color of cattle fed bluestem pasture without supplement until late October, concentrate in drylot for 70 days or concentrate in drylot for 150 days. Steaks from cattle fed 150 days had the most desirable color after cutting and after 3 days of display. Cattle fed 70 days were intermediate and grass fed cattle were darkest, unacceptably so after 3 days of display. Craig, Blumer and Barrick (1959) fed six rations of varying proportions of grain and grass to 60 steers. Muscle color of cattle fed ground corn and supplement was brighter initially, 30 minutes after cutting and after aging 7 days than muscle from cattle fed pasture only or grain on pasture.

Nobles and U.S.D.A. workers (1937) compared muscle color from steers fed grass only for 142 days, corn and supplement on grass for 142 days or grass only for 142 days followed by grain and mixed hay in drylot for 60 days. Muscle color of grass only steers was darkest and muscle from drylot steers was lightest.

Other findings have shown only slight differences in muscle color of cattle fed grass only or varied levels of concentrate. Bray (1938) conducted a 4 year study to evaluate the economic advantages of feeding grain on pasture versus white clover-

Bermuda grass pasture only. Muscle color of grass fed cattle was only slightly darker than grain fed cattle. Craig et al. (1966) fed steers grain on pasture or cut forage and grain in drylot and found no difference in visual color of longissimus muscle although muscle from drylot cattle was scored slightly darker.

Some research has shown muscle color becomes lighter with increased length of concentrate feeding. Gramlich, Loeffel and U.S.D.A. workers (1937a) evaluated muscle color periodically over a 224 day feeding period and found color became slightly lighter with longer feeding. Gramlich and U.S.D.A. workers (1937b) found 3 year old Hereford steers fed in Montana had darker muscle color than steers fed two different concentrate rations for 64 days following the range period. Conversely, Matthews and Bennett (1962) fed rations designed to produce varying rates of gain to three groups totaling 36 cattle. Muscle color, as scored by standards derived from Munsell color paddles, did not differ among treatments, but was scored darker as concentrate increased.

Effect of vacuum packaging on beef color

Several researchers have shown a loss of brightness by bloomed meat packaged in vacuum and a reoxygenation or brightening of the meat when exposed to oxygen (Iandrock and Wallace, 1955; Rickert, Ball and Stier, 1957a; Rickert et al., 1957b; Rickert et al., 1957c; Firko and Ayres, 1957; Dean and Ball, 1960; Jaye, Kittaka and Ordal, 1962; Fellers et al., 1963; and Pierson, Collins-Thompson and Ordal, 1970). Greene (1969)

showed anaerobic packaging can prevent Mb⁺ formation if adequate Mb⁺ reducing activity is present. Vacuum packaged muscle stored at 0°C for 14 days or more did not always brighten when exposed to air and was often greyish-brown on the surface (Fredholm, 1963). Work by Pierson et al. (1970) showed a preponderance of Mb during a 15 day storage period in anaerobic packaging. In aerobic packaging, a decline in MbO₂ and an increase in Mb⁺ was noted. Mb⁺ was the predominant pigment on the muscle surface after 5 days in aerobic packaging.

Jeremiah, Smith and Carpenter (1972b) reported cuts from vacuum packaged lamb racks and sirloins stored 14 days at 0°C discolored no faster than cuts from fresh lamb stored identically. During retail display in oxygen permeable film, however, vacuum packaged cuts discolored twice as fast as fresh cuts. Kennick et al. (1971) randomly assigned paired beef short loins representing five degrees of marbling (slight through slightly abundant). One short loin from each pair was aged in air or vacuum in a 35°F cooler before cutting and display. Color description and desirability were evaluated daily by a three member panel. No differences in caselife, color description or desirability scores were associated with type of aging.

Seideman et al. (1976) vacuum packaged 150 beef knuckles, 140 ribs and 60 arm chucks in bags manufactured from films differing in oxygen and moisture vapor transmission rates using two systems (nozzle and chamber vacuumizing machines). Fabricated steaks were evaluated by a three member panel using a 7 point scale (7=no discoloration, 1=total discoloration)

after 1 and 4 days of display. Steaks from cuts packaged by the chamber system utilizing film with low oxygen transmission rate had reduced surface discoloration ($P < .05$).

Reagan, Carpenter and Bauer (1976) scored muscle color and surface discoloration of longissimus steaks fabricated from beef ribs obtained from carcasses of known grass-grain feeding regimes and stored in vacuum bags at 1 to 3°C for 0, 21 and 28 days. Beef muscle obtained from grass-grain fed cattle was brighter and had less surface discoloration than grass fed cattle. The authors concluded beef from grass fed cattle was inferior to grain-grass fed cattle in stability traits during storage and in the retail case.

Effect of carcass conditioning temperature and time on beef color

Doty and Pierce (1961) showed color of longissimus steaks from Good and Prime grade carcasses, as determined by comparison with Munsell Color Plates, was definitely improved by a carcass aging period of 2 weeks at 34°F. McIntosh et al. (1942) reported muscle color of short loins aged at 34°F for 7 and 14 days was "distinctly lighter, fresher" than those aged at 50°F.

Kastner, Henrickson and Morrison (1973) randomly assigned six Hereford steer carcasses to each of three holding periods; namely 2, 5 or 8 hours at 16°C for one carcass side fabricated hot. The other side was identically fabricated after aging 48 hours at 2°C. Reflectance measurements on 16 steaks from each carcass taken 1 hour after cutting with a Photovolt Reflectance Meter were converted to Munsell color parameters.

Degrees of lightness and darkness were determined and used to express color reflectance values. A relatively dark colored product was produced by hot boning at 2 hours compared to conventional cutting as measured visually and objectively ($P < .01$). However, cold boned steaks were darker than hot boned steaks cut at 5 and 8 hours. Even though Munsell value for steaks cut at the 5 and 8 hour periods were different ($P < .10$ and $P < .05$, respectively) between hot and cold treatments, subjective evaluation by a seven member, untrained panel failed to verify the objective differences. The authors concluded color differences between treatments were not visually apparent and would not influence acceptability if carcasses are subjected to the higher temperature at least 5 hours before fabrication.

Kastner and Russell (1975) assigned 15 Good and Choice heifer carcasses to one of three postmortem holding periods. One half was chilled for 6, 8 or 10 hours at 16°C prior to muscle excision and the other half chilled 48 hours at 2°C . Photovolt reflectance readings were converted to Munsell values according to Kastner et al. (1973). Means for hot boned steaks were larger (darker) ($P < .05$ and $P < .10$, respectively) than cold boned steak means at the 8 and 10 hour periods; however, subjective scores reflected no difference. Apparently differences were too subtle to be visually detected.

Effect of maturity on beef color

Mackintosh and Hall (1935) reported observations of 54 mature and 53 yearling steers indicating that greater maturity

tends to produce darker beef. Readings of longissimus muscle by the spinning wheel and standard Munsell discs showed a lower brilliance and chroma in mature steers.

Doty's (1956) summary of 153 graded carcasses (Commercial, Good or Prime) reported carcass grade was related ($P < .01$) to longissimus color measured with Munsell color plates. Prime grade beef was lighter than beef from Commercial cows.

Tuma et al. (1962) used 24 Hereford females (18, 24 or 90 months old) fed a high concentrate ration to a level of finish estimated to produce a marbling level of slight or slightly abundant. Longissimus steaks were read 1 hour after cutting on a Photovolt Reflectance Meter and Munsell data were calculated. Decreases in hue ($P < .005$), value ($P < .01$) and chroma ($P < .05$) notations occurred as animal age increased. Decreased hue and value notations indicate color darkening. Decreasing chroma suggests a color darkening or less intense red.

Further work by Tuma et al. (1963), using loin steaks from 56 Hereford steers and heifers 6, 18, 42 and 90 months old evaluated by Photovolt readings converted to Munsell data, supported the results of Tuma et al. (1962). Muscle color darkened with advancing age as shown by decreases in value ($P < .01$) and chroma ($P < .05$).

Photovolt readings of steaks from 80 beef ribs of four maturity levels (A,B,C and D) converted to Munsell data by Romans, Tuma and Tucker (1965) showed only value to be affected by maturity ($P < .01$). A maturity cuts had a higher value (lighter color) than other maturity groups ($P < .01$).

Effect of marbling on muscle color

Mackintosh and Hall (1935) found muscle color brightened as marbling increased. The authors explained the correlation as (1) the effect of added white coloring matter on the surface and (2) the effect of fat on oxygen permeability.

Doty (1956) in a study of 153 graded carcasses, found longissimus color compared with Munsell color plates to be related to carcass grade. Prime beef was lighter in color than Good beef or Commercial Cows.

Tuma et al. (1962), using longissimus steaks representing two marbling levels (slight and slightly abundant), discovered no marbling influence ($P < .05$) on any of the three dimensions of color (hue, value or chroma). Romans, Tuma and Tucker (1965) compared steaks from 80 ribs with either moderate or slight marbling and found similar hue, value and chroma notations.

Kernick et al. (1971) reported that marbling had a positive linear effect ($P < .01$) on color description scores (6=bright, 1=greenish) at 24 and 48 hours post-cutting of steaks from short loins representing five degrees of marbling (slight through slightly abundant). In addition, marbling had a curvilinear effect ($P < .05$) on case life (terminated when a visible spot of Mb⁺ appeared), with slight and slightly abundant having the shortest case life.

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CHAPTER 3

EFFECT OF VACUUM PACKAGING AND LENGTH OF DISPLAY ON VISUAL COLOR SCORE, PERCENT METMYOGLOBIN AND PERCENT REFLECTANCE AT 630 nm MINUS PERCENT REFLECTANCE AT 580 nm OF FOUR BEEF MUSCLES.

INTRODUCTION

Distribution of vacuum packaged beef cuts is currently an industry-wide practice. Today's more efficient meat distribution patterns with emphasis on centralized cutting, storage and distribution as a means of reducing labor and transportation costs insure the continued and/or increased use of vacuum packaging.

Research on anaerobic storage of beef wholesale cuts has dealt with weight loss (Hodges et al., 1974), microbiological and sensory considerations (Pierson et al., 1970; Jeremiah et al., 1971; Kennick et al., 1971; Minks and Stringer, 1972; Jeremiah et al., 1972a; and Jeremiah et al., 1972b) and differences in color due to different nutritional regimes (Reagan et al., 1976).

However, a review of the literature shows no one has completed any extensive study of initial color and color stability of beef muscle from carcasses of varied nutritional background. This study compared beef muscle color and stability during simulated retail display of cuts vacuum packaged 21 days versus cuts fabricated 48 hours post-slaughter. Nutritional effects on color are reported in the preceding chapter.

MATERIALS AND METHODS

Thirty-two steers of known background, approximately 18 months old, from the USDA Meat Animal Research Center at Clay Center, Nebraska, were wintered alike on a ration composed of 48% corn silage (IRN 3-02-824), 50% alfalfa haylage (IRN 3-08-151) and 2% soybean meal (IRN 5-04-604) supplement containing calcium, phosphorus, vitamin A and chlorotetracycline.

All steers were grazed through the summer on brome and bluestem pasture with no additional concentrate. At the end of the grazing period eight randomly selected steers were slaughtered. The remaining 24 animals were assigned to either a short-fed, long-fed or silage-fed finishing program in drylot. The short-fed group was fed a 20% alfalfa haylage, 75.2% cracked corn (IRN 4-02-932) and 4.8% SBM supplement ration for 49 days. Long-fed cattle were fed 98 days on an identical ration. The silage-fed group received a ration consisting of 40% corn silage, 20% alfalfa haylage, 36% cracked corn and 4% SBM supplement for 98 days. All steers also had free choice access at all times to both block salt and a mixture of 1/3 loose salt, 1/3 limestone and 1/3 dicalcium phosphate.

As each feeding period ended the steers were trucked to the Kansas State University meat laboratory for slaughter.

One side of each carcass was randomly assigned to a conditioning-chill treatment of eight hours at 16 C, then 40 hours at 2 C; the other side was chilled for 48 hours at 2 C. At 48 hours post mortem carcasses were fabricated and the longissimus

(L), semitendinosus (ST), biceps femoris (BF) and semimembranosus (SM) muscles were removed from each side and transversely halved. The anterior (proximal) half of each muscle was fabricated immediately into steaks 2.54 cm thick. The posterior (distal) half was placed in a vacuum bag and stored 21 days at 2 C before fabrication.

Display steaks from each carcass side were from the same anatomical location and were placed in a styrofoam tray, overwrapped with PVC (polyvinylchloride) and displayed for five days at 2 C under continuous (24 hours/day) General Electric Delux Warm White lighting at an intensity of 1076 lumens/m² (100 foot candles) at meat surface level. Lighting consisted of two 40 watt tubes 126 cm from the muscle surface.

Subjective muscle color was scored individually by five panelists under display lighting at initial display (day 0) and at one, three and five days of display to the nearest .5 point using a scale of 1 = very bright red, 2 = bright red, 3 = slightly dark red or brown, 4 = dark red or brown and 5 = exceptionally dark red or brown (Kansas State University). A visual score of 3.5 was considered marginally unacceptable.

Objective color was measured at days 0, 1, 3 and 5 of display with a Bausch and Lomb 600 Spectrophotometer with reflectance attachment calibrated for 100% reflectance with MgCO₃ blocks. Reflectance spectra were scanned from 400 nm to 700 nm at a speed of 50 nm/min. Reflectance at 10 nm intervals was measured and printed out with an Autolab Minigrator connected to the spectrophotometer and set for a peak height mode.

Reflectance at intermediate points was determined by interpolation.

Percent metmyoglobin (Mb^+) was calculated according to Stewart et al. (1965).

Stepwise multiple regression was used to determine the contribution of a number of objective measurements to R^2 (Snedecor and Cochran, 1967). Analysis of regression was performed on the percent reflectance (%R) at every 10 nm from 410 nm to 700 nm and for %R 630 nm - %R 580 nm, %R 630 nm/%R 580 nm, %R 632 nm - %R 614 nm, %R 582 nm/%R 525 nm, %R 572 nm/%R 525 nm, %R 507 nm/%R 572 nm and %R 474 nm/%R 597 nm, as well as the K/S ratios of these measurements for each muscle group. Because %R 630 nm - %R 580 nm had the greatest contribution to R^2 in three of four muscles, it was used as the best indicator of visual score.

Analysis of variance by least squares method was calculated for day and packaging effects and for the packaging x day interaction on each variable. The main effects and interaction were tested by the least significant difference method to find which means were different (Steel and Torrie, 1960).

Preliminary analysis showed no conditioning-chill treatment effect on any of the three variables; therefore, both were pooled.

RESULTS AND DISCUSSION

Day effect. As expected, visual scores for all muscles were darker ($P < .05$) with increasing display time (table 1). L and ST steaks remained visually acceptable through the fifth display day; however, BF and SM steaks approached the marginally unacceptable score of 3.5 at day 3 and were definitely unacceptable by day 5.

Percent Mb⁺ and 630 nm - 580 nm difference for all muscles followed the visual score pattern. Percent Mb⁺ was higher ($P < .05$) and 630 nm - 580 nm difference was lower ($P < .05$) at successive evaluation periods.

Packaging effect. Visual scores of L, BF and SM steaks from cuts held 21 days in vacuum prior to cutting were lower (brighter) ($P < .05$). The greatest improvement was realized for the L muscle. ST muscle was adversely affected ($P < .05$) by vacuum packaging.

In anaerobic packaging, reduced myoglobin (Mb) becomes the predominant chemical form of myoglobin (Pierson et al., 1970). Upon exposure to oxygen, Mb is converted to oxymyoglobin (MbO₂) which results in a brightening of the muscle surface (Landrock and Wallace, 1955; Rickert, Ball and Stier, 1957a; Rickert et al., 1957b; Rickert et al., 1957c; Pirko and Ayres, 1957; Dean and Ball, 1960; Jaye, Kittaka and Ordal, 1962; Fellers et al., 1963; and Pierson, Collins-Thompson and Ordal, 1970). Rickansrud and Henrickson (1967) reported mean myoglobin values of 3.18, 2.40, 3.64 and 1.99 mg/g wet tissue for beef longissimus, psoas major,

biceps femoris and semitendinosus muscles, respectively. Muscles with low myoglobin concentrations would be expected to show less response to vacuum packaging and subsequent exposure to oxygen. This may explain the behavior of vacuum packaged ST muscle.

Percent Mb^+ values as a result of packaging did not coincide well with visual scores. L and BF percent Mb^+ were not different and SM values were opposite ($P < .05$) the visual scores.

It may also be that a complete vacuum was not pulled in some of the packages. Low oxygen partial pressures (pO_2) (4 mm Hg) (Brooks, 1938); 1 - 11.4 mm Hg (George and Stratmann, 1952); muscled packaged in O_2 impermeable film (Zimmerman and Snyder, 1969); and 1% O_2 (Ledward and Macfarlane, 1971) strongly encourage autoxidation of Mb to Mb^+ .

Percent Mb^+ values as a result of packaging did not coincide well with visual scores. L and BF percent Mb^+ were not different and SM values were opposite ($P < .05$) the visual scores. Several factors could have caused the lack of agreement between visual scores and percent Mb^+ . The calculation of percent Mb^+ developed by Stewart, Zipser and Watts (1965) may be an inaccurate measure of Mb^+ . Perhaps pigment analysis would have given the answer to this. In addition, the very small area (.3 cm x .5 cm) scanned by the spectrophotometer may very well have not been representative of the entire steak surface. Although great care was taken to scan a representative area of each steak, a larger scanning area would have been much more desirable. A different visual scoring method may have given greater correlation

between visual score and percent Mb^+ . As it was, each panelist was asked to score color of each steak based on an average of the entire surface. Quite possibly the average color score was not related to the small area of scan. A visual system that took into account percent of steak surface discolored may have been more helpful.

In each instance %R 630 nm - %R 580 nm values agreed with visual score patterns. This agrees with Strange et al. (1974) who found %R 630 nm - %R 580 nm values were apparently closely related to MbO_2 and therefore to visual score.

Packaging x day interaction.

L steaks from cuts stored in vacuum 21 days at 2 C before cutting (vac L steaks) showed brighter ($P < .05$) visual scores through the display period compared to L steaks from cuts fabricated immediately after a carcass chill of 48 hours at 2°C (non-vac L steaks) (table 2). Non-vac L steaks showed no difference in visual score between days 3 and 5, indicating non-vac L steaks discolor faster up to 3 days of display, but then have little additional color deterioration.

Packaging method had no effect on visual score of ST steaks through day 1; however, non-vac ST steaks had a brighter ($P < .05$) visual color than vac ST at days 3 and 5 of display.

The greatest benefit of vacuum packaging was realized by BF and SM muscles. In terms of visual score, display life of the two muscles was lengthened by one to two days. Vac BF showed improved ($P < .05$) visual scores through day 3. At day 3 non-vac

BF steaks were visually unacceptable (3.64), but vac BF muscle visual scores (3.33) were still below 3.5, the marginally unacceptable point. However, at day 5 vac BF muscle had a more undesirable ($P < .05$) visual color than non-vac. It may be that aerobic microorganisms deprived nutritionally by being encased in the anaerobic atmosphere discolor muscle at a higher rate after adjusting to aerobic conditions.

SM steaks showed an identical pattern as BF steaks.

Percent Mb^+ of L steaks was not affected by packaging method through day 3, but at day 5 vac L muscle had more Mb^+ ($P < .05$) than non-vac. This might be explained by a low pO_2 in vacuum which would cause a more rapid conversion of Mb to Mb^+ or it might be concluded that Stewart's method is not a good measure of Mb^+ .

Mb^+ percent was lower ($P < .05$) at each evaluation period for ST steaks fabricated after a 48 hour carcass chill than for ST steaks cut after a 21 day holding period in vacuum bags. Apparently the rate of Mb^+ formation in ST muscle is increased by vacuum packaging, perhaps because of the existence of low pO_2 during vacuum storage.

No established pattern for Mb^+ formation in BF muscle was seen. Initially, Mb^+ percentage for BF steaks was similar between packaging treatments. Less Mb^+ was evident on the surface of vac BF muscle at day 1, but the proportion was similar at day 3. On day 5 the percentage of Mb^+ was higher in the vac BF steaks.

SM muscle displayed a more predictable pattern of Mb^+ formation than BF. Percent Mb^+ was initially less ($P < .05$) in the

vac steaks, was similar at day 1 between packaging methods and was greater ($P < .05$) in vac steaks at the last two days of display.

%R 630 nm - %R 580 nm values for L steaks generally followed the pattern of visual scores; however, the non-vac steaks were not different from vac between days 0 and 1 or at day 5.

ST %R 630 nm - %R 580 nm results were quite unusual and, in one respect, different from the pattern of visual scores. The value of vac muscle at day 0 was less than the same muscle (vac ST) at days 1 and 3. Display temperature may account for this phenomenon; ST steaks were held at 1-2 C less than the other steaks which might have slowed oxidation of Mb. Perhaps, too, because of the low myoglobin content of ST muscle, a longer period is required for oxygenation.

%R 630 nm - %R 580 nm values reacted similarly for BF and SM, but neither followed the pattern established by the visual scores. The values were larger ($P < .05$) at days 0 and 1 for vac BF and SM, but there was no difference due to packaging on days 3 and 5.

Table 1. Packaging and day of display effects on mean visual score, percent Mb⁺ and %R 630 nm - %R 580 nm of four beef muscles.

	Visual Score ^e	Percent Mb ⁺ ^f	%R 630 nm - %R 580 nm
<u>Longissimus:</u>			
Packaging:			
Non-vac ^g	3.03 ^b	18.27 ^a	20.88 ^b
Vac ^h	2.54 ^a	18.80 ^a	23.48 ^a
Day: 0	2.43 ^a	9.74 ^a	24.45 ^a
1	2.62 ^b	16.72 ^b	23.84 ^b
3	2.99 ^c	21.39 ^c	21.42 ^c
5	3.20 ^d	26.31 ^d	19.01 ^d
<u>Semiterdinosus:</u>			
Packaging:			
Non-vac	2.15 ^a	11.56 ^a	29.82 ^a
Vac	2.23 ^b	17.37 ^b	29.24 ^b
Day: 0	1.77 ^a	4.19 ^a	33.00 ^a
1	1.98 ^b	12.03 ^b	31.80 ^b
3	2.36 ^c	18.79 ^c	28.02 ^c
5	2.66 ^d	22.86 ^d	25.30 ^d
<u>Biceps femoris:</u>			
Packaging:			
Non-vac	3.35 ^b	34.40 ^a	14.12 ^b
Vac	3.07 ^a	34.65 ^a	16.58 ^a
Day: 0	2.59 ^a	13.85 ^a	22.70 ^a
1	3.05 ^b	30.76 ^b	16.13 ^b
3	3.48 ^c	43.40 ^c	12.26 ^c
5	3.73 ^d	50.10 ^d	10.29 ^d
<u>Seminembranosus:</u>			
Packaging:			
Non-vac	3.33 ^b	28.16 ^a	16.73 ^b
Vac	3.08 ^a	30.38 ^b	20.26 ^a
Day: 0	2.63 ^a	13.34 ^a	24.75 ^a
1	2.96 ^b	25.20 ^b	19.77 ^b
3	3.35 ^c	36.10 ^c	15.70 ^c
5	3.87 ^d	42.50 ^d	13.77 ^d

a-b Observations within the same muscle-day-color parameter or muscle-packaging-color parameter block bearing similar superscript letters are not different (P<.05).

e 1=very bright red, 2=bright red, 3=slightly dark red or brown, 4=dark red or brown, 5=exceptionally dark red or brown

f Calculated according to Stewart et al., (1965)

g Fabricated after 48 hour carcass chill.

h Fabricated after 21 day storage in vacuum bag at 2°C.

Table 2. Packaging x day of display interaction on mean visual score, percent Mb+ and %R 630 nm - %R 580 nm of four beef muscles.

	Visual Score ^h , day					Percent Mb+ ⁱ , day					%R 630 nm - %R 580 nm, day				
	0	1	2	3	5	0	1	2	3	5	0	1	2	3	5
Longissimus:															
Non-vac ^j	2.72 ^c	2.94 ^d	3.34 ^e	3.33 ^e		10.02 ^a	16.49 ^b	21.37 ^c	25.20 ^d		21.34 ^{cd}	22.14 ^c	20.89 ^d	19.15 ^e	
Vac ^k	2.14 ^a	2.30 ^b	2.65 ^c	3.08 ^d		9.47 ^a	16.94 ^b	21.41 ^c	27.41 ^e		27.57 ^a	25.53 ^b	21.96 ^c	18.87 ^e	
Semitendinosus:															
Non-vac	1.80 ^{ab}	2.01 ^c	2.26 ^d	2.51 ^e		2.33 ^a	10.21 ^c	15.56 ^d	18.15 ^e		29.17 ^d	32.01 ^b	30.07 ^c	28.03 ^e	
Vac	1.75 ^a	1.94 ^{bc}	2.45 ^e	2.80 ^f		6.05 ^b	18.36 ^d	21.99 ^f	27.58 ^g		36.83 ^a	31.59 ^b	25.97 ^f	22.57 ^g	
Biceps femoris:															
Non-vac	2.76 ^b	3.36 ^c	3.64 ^d	3.64 ^d		13.42 ^a	32.11 ^c	43.74 ^d	48.34 ^e		19.05 ^b	14.79 ^d	12.08 ^e	10.55 ^f	
Vac	2.42 ^a	2.73 ^b	3.33 ^c	3.81 ^e		14.28 ^a	29.40 ^b	43.05 ^d	51.86 ^f		26.35 ^a	17.47 ^c	12.44 ^e	10.04 ^f	
Semimembranosus:															
Non-vac	2.87 ^c	3.20 ^d	3.53 ^e	3.72 ^f		15.34 ^b	24.56 ^c	34.09 ^d	38.77 ^e		19.43 ^c	18.11 ^d	15.32 ^e	14.07 ^f	
Vac	2.39 ^a	2.72 ^b	3.17 ^d	4.03 ^e		11.34 ^a	25.84 ^c	38.12 ^e	46.22 ^f		30.07 ^a	21.43 ^b	16.07 ^e	13.46 ^f	

^{a-g}Observations within the same muscle-color parameter block bearing similar superscript letters are not different (P<.05).

^h1=very bright red, 2=bright red, 3=slightly dark red or brown, 4=dark red or brown, 5=exceptionally dark red or brown

ⁱCalculated according to Stewart et al. (1965).

^jFabricated after 48 hour carcass chill.

^kFabricated after 21 day storage in vacuum bag at 2°C.

SUMMARY

Effects of two packaging methods (steaks fabricated immediately after a carcass chill of 48 hours at 2 C versus steaks from cuts stored 21 days in vacuum bags before fabrication) and length of display on visual color score, percent metmyoglobin (Mb^+) and percent reflectance at 630 nm minus percent reflectance at 580 nm ($\%R\ 630\ \text{nm} - \%R\ 580\ \text{nm}$) of beef longissimus (L), semitendinosus (ST), biceps femoris (BF) and semimembranosus (SM) muscles were compared.

For all muscles, visual color became darker, percent Mb^+ increased and $\%R\ 630\ \text{nm} - \%R\ 580\ \text{nm}$ values decreased ($P < .05$, respectively) as display time lengthened. Vacuum packaged L, BF and SM muscles had brighter ($P < .05$) visual color and higher ($P < .05$) $\%R\ 630\ \text{nm} - \%R\ 580\ \text{nm}$ values following cutting, blooming and display when compared to the same muscles cut at 48 hours post-slaughter; ST muscle had darker ($P < .05$) visual color and lower ($P < .05$) $\%R\ 630\ \text{nm} - \%R\ 580\ \text{nm}$ values when vacuum packaged.

Vacuum packaged L muscle was visually brighter ($P < .05$) than muscle excised and cut into steaks immediately after a 48 hour carcass chill. The $\%R\ 630\ \text{nm} - \%R\ 580\ \text{nm}$ value was higher ($P < .05$) for vacuum packaged L muscle at all evaluation periods except day 5. No difference in percent Mb^+ existed in the first three days of display.

Vacuum packaging improved ($P < .05$) visual scores of BF and SM through the first three days of display under continuous, 24 hour, 100 foot candles of lighting at 2 C packaged in PVC film.

%R 630 nm - %R 580 nm values for BF and SM were increased ($P < .05$) by vacuum storage at the first two display days. An inversion ($P < .05$) in visual score and %R 630 nm - %R 580 nm value between packaging treatments occurred at the last evaluation period.

ST visual score was not different between packaging treatments through two days of display, but was lower ($P < .05$) for vacuum packaged through the last two display days. %R 630 nm - %R 580 nm was higher ($P < .05$) initially for the vacuum stored ST and was lower ($P < .05$) in the last half of display. Percent Mb⁺ was higher ($P < .05$) for vacuum packaged ST at all evaluation periods.

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CHAPTER 4

EFFECT OF NUTRITIONAL REGIME, VACUUM PACKAGING AND LENGTH OF DISPLAY ON VISUAL COLOR SCORE, PERCENT METMYOGLOBIN AND PERCENT REFLECTANCE AT 630NM MINUS PERCENT REFLECTANCE AT 580NM OF FOUR BEEF MUSCLES.

INTRODUCTION

Costs of feed, fuel, equipment, processing, storage, labor, packaging and distribution of beef will most likely continue to increase in the future. Research directed toward evaluating and improving current production, processing and marketing methodology is necessary to increase efficiency and reduce the overall expense involved in producing and marketing beef. Assuming our country to have a freely competitive economy, benefits realized in improved efficiency and lowered production costs may be passed to the beef consumer. We must be concerned, however, that improvements which lower costs or increase efficiency are accomplished without reducing product quality thus enabling beef to maintain its position as the favored meat in the consumers' diet.

Severe cost pressure due to fluctuating feed ingredient costs and fed cattle prices have affected profitability of cattle feeding causing producers to accept alternate finishing systems for cattle. In addition, pressures have been brought to bear against the beef industry to reduce the volume of grains fed to cattle so that more is available for human consumption.

A number of alternate systems have been adopted by feeders in recent years to lower feeding costs. These methods include:

(1) finishing on grass alone, (2) growing on grass and finishing with predominantly roughage feeds and (3) growing to heavier weights on grass and then finishing in drylot on grain for shorter lengths of time. These alternate systems are designed for maximum utilization of roughages while limiting the amounts of grain fed.

Increased slaughter of cattle finished with no concentrate feeding or concentrate for shorter periods or finished on high roughage rations has raised questions by producers, packers, purveyors and retailers about carcass characteristics of beef produced under these systems. Some of the questions have been answered in the past. However, little recent data is available which relates effects of forage feeding or shorter periods of grain feeding on initial muscle color or on color stability during retail display. In addition, work relating the effect of vacuum packaging on beef muscle color from forage fed cattle is definitely lacking.

This study is designed to define changes in beef muscle color that may result from grazing grass alone, feeding high roughage rations in drylot or from shorter feeding periods on grain in drylot. Also, any color changes caused by interactions of nutritional regime and vacuum packaging were analyzed.

MATERIALS AND METHODS

Thirty-two steers of known background, approximately 18 months old, from the USDA Meat Animal Research Center at Clay Center, Nebraska, were used. The steers were wintered alike on a ration composed of 48% corn silage (IRN 3-02-824), 50% alfalfa haylage (IRN 3-08-151) and 2% soybean meal (IRN 5-04-604) supplement containing calcium, phosphorus, vitamin A and chlorotetracycline.

All steers were grazed through the summer on brome and bluestem pasture with no additional concentrate. At the end of the grazing period eight steers were randomly selected for slaughter. The remaining 24 animals were assigned to either a short-fed, long-fed or silage-fed finishing program in drylot. The short-fed group was fed a 20% alfalfa haylage, 75.2% cracked corn (IRN 4-02-932) and 4.8% supplement ration for 49 days. Long-fed cattle were fed 98 days on an identical ration. The silage-fed group received a ration consisting of 40% corn silage, 20% alfalfa haylage, 36% cracked corn and 4% supplement for 98 days. All steers also had free choice access at all times to both block salt and a mixture of 1/3 loose salt, 1/3 limestone and 1/3 dicalcium phosphate.

As each feeding period ended the steers were trucked to the Kansas State University meat laboratory for slaughter.

One side of each carcass was randomly assigned to a conditioning-chill treatment of eight hours at 16°C, then 40 hours at 2°C; the other side was chilled for 48 hours at 2°C. At 48

hours post mortem carcasses were fabricated and the longissimus (L), semitendinosus (ST), biceps femoris (BF) and semimembranosus (SM) muscles were removed from each side and transversely halved. The anterior or proximal half of each muscle was fabricated immediately into steaks. The posterior or distal half was placed in a vacuum bag and stored 21 days at 2°C before fabrication.

Display steaks 2.54 cm thick from each carcass side from the same anatomical location were placed in a styrofoam tray, overwrapped with PVC (polyvinylchloride) and displayed for five days at 2°C under continuous (24 hour day) General Electric Delux Warm White lighting at an intensity of 1076 lumens/m² (100 foot candles) at meat surface level. Lighting consisted of two 40 watt tubes 126 cm from the muscle surface.

Subjective muscle color was scored individually by five panelists under display lighting at initial display (day 0) and after one, three and five days of display to the nearest .5 point using a scale of 1 = very bright red, 2 = bright red, 3 = slightly dark red or brown, 4 = dark red or brown and 5 = exceptionally dark red or brown (Kansas State University). A visual score of 3.5 was considered marginally unacceptable.

Objective color was measured at days 0, 1, 3 and 5 of display with a Bausch and Lomb 600 Spectrophotometer with reflectance attachment calibrated for 100% reflectance with MgCO₃ blocks. Reflectance spectra were scanned from 400 nm to 700 nm at a speed of 50 nm/min. Reflectance at 10 nm intervals was measured and printed out with an Autolab Minigrator connected

to the spectrophotometer and set for a peak height mode. Reflectance at intermediate points was determined by interpolation.

Percent Mb⁺ was calculated according to Stewart et al. (1965).

Stepwise multiple regression was used to determine the contribution of a number of objective measurements to R² (Snedecor and Cochran, 1967). Analysis of regression was performed on the percent reflectance (%R) at every 10 nm from 410 nm to 700 nm and for %R 630 nm - %R 580 nm, %R 630 nm/%R 580 nm, %R 632 nm - %R 614 nm, %R 582 nm/%R 525 nm, %R 572 nm/%R 525 nm, %R 507 nm/%R 572 nm, %R 474 nm/%R 597 nm, as well as the K/S adjustments of these measurements for each muscle group. Because %R 630 nm - %R 580 nm had the greatest contribution to R² in three of four muscles, it was used as the best objective indicator of visual score. Strange et al. (1974) and Leising (1975) reported simple correlation coefficients to visual color score of -.86 and -.82, respectively, for %R 630 nm - %R 580 nm and suggested its use as a predictor of visual score.

Analysis of variance by least squares method was calculated for nutritional effect and for the nutrition x packaging, nutrition x day of display and nutrition x packaging x day of display interactions on each variable. The main effect and interactions were tested by the least significant difference method to find which means were different (Steel and Torrie, 1960).

Analysis showed no chill treatment effect on any of the three variables; therefore, both chill treatments were pooled.

RESULTS AND DISCUSSION

Nutrition effect. Type of feed had no effect ($P < .05$) on visual score for any of the four muscles (table 3). This agrees with the results of Bray (1938) and Matthews and Bennett (1962) who found no difference in muscle color from cattle fed rations differing in forage level. The results also agree with those of McCampbell and Mackintosh (1927), Mackintosh and Hall (1935), Longwell (1936), Bray and U.S.D.A. workers (1937), Mackintosh et al. (1937), Brown (1954), McCampbell et al. (1960), Malphrus et al. (1962) and Craig et al. (1966), all who reported no effect of feeding grain in drylot, grain on pasture or pasture only on muscle color. Conversely, Gramlich et al. (1937), Nobles and U.S.D.A. workers (1937), Craig et al. (1959) and Kropf et al. (1975) reported a brighter muscle color in cattle fed high grain rations compared to those on high forage or grass rations.

Differences observed in percent Mb^+ and %R 630 nm - %R 580 nm among nutritional regimes were apparently not great enough to influence visual score. Percent Mb^+ was greatest ($P < .05$) in steaks from long-fed and silage-fed animals and least in grass-fed and short-fed steaks. %R 630 nm - %R 580 nm differences tended to follow the same pattern, but with high values (brighter color) for means low in Mb^+ . This possibly reflects slightly higher maturity scores for the longer fed groups. Myoglobin levels tend to increase with maturity (Romans et al., 1965) and increased amounts may cause the steak to be more susceptible to oxidation.

Nutrition x day of display interaction.

Longissimus: L steaks from all nutritional regimes remained acceptable through day 3 of display according to visual scores (table 4). At day 5, however, those from grass-fed and short-fed cattle approached unacceptability. Apparently longer feeding periods improved color stability over longer display periods. L steaks from silage-fed cattle showed especially greater stability in the last half of display, being scored brightest ($P < .05$) at days 3 and 5.

Greatest differences in percent Mb^+ were observed at day 5. Short-fed L steaks had the least Mb^+ ($P < .05$) and grass-fed had the most ($P < .05$). Grass-fed L steaks showed a large increase in Mb^+ from day 3 to day 5, perhaps partly due to bacterial oxidation that occurred on the surface of vacuum packaged L steaks from grass-fed cattle at that time. Thomas (1977) evaluated vacuum packaged inside chucks from the same carcasses in this study stored 21 days at $2^{\circ}C$ and reported higher ($P < .05$) mean aerobic bacterial counts for inside chucks from grass-fed cattle compared with inside chucks from the other regimes. Possibly the muscles from grass-fed cattle we studied also had higher aerobic counts. This would explain the apparent occurrence of bacterial oxidation. He stated inside chucks from animals of different nutritional backgrounds can be stored under vacuum for 21 days at 0 to $1^{\circ}C$ and remain within acceptable microbial limits. This may be true initially, but the counts may be too high to insure color stability over an extended display.

%R 630 nm - %R 580 nm differences showed no established pattern and agreed poorly with visual scores, particularly in later display. Inconsistent agreement between subjective and objective evaluations resulted from a number of factors. Many steaks demonstrated uneven surface discoloration which presented the panelists with the problem of averaging visual score. This was further compounded by the small light beam area (.3 cm x .5 cm) of the spectrophotometer which likely prohibited scanning a representative area. Perhaps the use of an instrument with a larger light beam or one of the recently introduced photoacoustic spectrophotometers would give better agreement between objective and subjective scores.

The amount of dried juices on the covering film or degree of film glossiness could affect both objective and subjective evaluations. In addition, consistent visual scoring requires that the type and intensity of lighting be the same at all time periods. Even with structured visual scales with reference pictures, scorer variability may have been a problem.

Snyder (1965) discussed problems of reflectance measurements. Percent reflectance will depend on pigment concentration, amount of intramuscular fat and/or moisture. Effects of sample presentation were discussed by Clydesdale and Francis (1975). Uneven, concave or convex surface and "pillowing" are possible with muscle samples. Fiber orientation will also affect reflectance.

Semintendinosus: ST muscle from all nutritional regimes received very low visual scores (brighter color) through day 3.

Scores were higher by the end of the display period, but never approached unacceptability. The low scores can probably be attributed to low myoglobin levels in ST muscle.

Likewise, percent Mb^+ was extremely low initially. Percent Mb^+ in ST muscle from grass-fed cattle was consistently lowest ($P < .05$) from day 1 through day 5. Grass-fed steaks had a coarser lean texture score ($P < .05$) than steaks from the other regimes. The effects of lean texture on oxidation rate of muscle should be questioned.

The low Mb^+ percentages for ST muscle from grass-fed cattle were supported by higher (more desirable) ($P < .05$) %R 630 nm - %R 580 nm values. Lower %R 630 nm - %R 580 nm values were recorded for silage-fed and long-fed cattle. Evidently this observation is reduced by longer feeding.

Biceps femoris: Differences in BF visual score among nutritional regimes were small initially, but at day 1, BF steaks from grass-fed cattle were brighter (lower) ($P < .05$) in visual score than those from cattle fed grain (short-fed and long-fed). BF muscle from all nutritional regimes was visually acceptable through day 1. At day 3 BF steaks from cattle fed grain had passed the point of marginal unacceptability and those from silage-fed and grass-fed cattle closely approached the point. BF muscle from silage-fed cattle had the lowest (brightest) visual scores at days 3 and 5 which indicates greater stability over an extended display.

Percent Mb⁺ was greatest in BF from silage-fed cattle at day 1. Highest (P<.05) Mb⁺ levels were recorded for BF steaks from longer fed animals (long-fed and silage-fed) at day 3. A drastic increase occurred in percent Mb⁺ from day 3 to day 5 for BF steaks from grass-fed cattle. The increase can again be partly attributed to bacterial growth. For this muscle myoglobin oxidation was enhanced by longer feeding periods.

%R 630 nm - %R 580 nm values well supported the observed differences in Mb⁺ percentage as a function of the length of feeding. From day 1 through day 5 the values observed for the two longer fed groups were lower (P<.05) than BF steaks from silage and grass-fed cattle.

Semimembranosus: The only visual differences observed in SM muscle as a result of nutrition through day 1 occurred initially. SM steaks from short-fed cattle were scored lower (P<.05) than those from silage-fed cattle. By day 3 those steaks from grass-fed cattle were visually unacceptable and those from short-fed and long-fed cattle approached unacceptability. SM steaks from silage-fed cattle showed the greatest color stability. They tended to have the lowest visual score at day 3 and had the lowest (P<.05) at day 5. Again, steaks from those cattle fed grain (short- and long-fed) tended to have the darkest visual scores at day 5, which may indicate grain feeding tends to decrease color stability over an extended display.

Small differences were observed in percent Mb⁺ at day 0 although SM steaks from the silage-fed cattle tended to have the most through day 1. By day 3 the longer fed groups (silage-

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and long-fed showed the greatest amounts of Mb⁺. Once again, steaks from the grass-fed cattle showed a large increase in percent Mb⁺ from day 3 to day 5, probably as a result of bacterial contamination that occurred on the surface of the vacuum packaged SM muscle at this time. It is uncertain whether the contamination is attributable to poor sanitation or to nutrition. Percent Mb⁺ was similar for all nutritional regimes at day 5, but short-fed steaks tended to have the least.

Among nutritional regimes, SM steaks from silage-fed cattle had lowest ($P < .05$) %R 630 nm - %R 580 nm values at days 0 and 1. The longer fed groups (silage and long-fed) had lower ($P < .05$) scores at day 3 which supports the Mb⁺ observation. Steaks from short-fed cattle tended to have the highest value. Those from grass-fed cattle showed the greatest decrease from day 3 to 5, probably due to bacterial contamination.

Nutrition x packaging interaction.

Longissimus: Overall, vacuum packaging lowered ($P < .05$) visual scores (brighter color) of the L muscle from all nutritional treatments (table 5). Among the non-vacuum packaged muscles those from the short-fed cattle tended to be scored darker. When vacuum packaged, the L muscle from grass-fed cattle had the highest (darkest) ($P < .05$) visual score and steaks from silage-fed cattle were scored lowest ($P < .05$).

Within nutritional regimes vacuum packaging had no effect on percent Mb⁺, however, within the non-vacuum packaged treatment L steaks from short-fed cattle had the lowest ($P < .05$) Mb⁺ level. No difference existed within the vacuum packaged treatment.

%R 630 nm - %R 580 nm values strongly supported the visual scores, being higher in each case for the vacuum packaged L muscle. Within the non-vacuum treatment, steaks from silage-fed animals had the lowest ($P<.05$) value. When vacuum packaged, muscle from silage-fed cattle tended to have the lowest value and that from grass-fed the highest.

Semitendinosus: Only ST steaks from grass-fed cattle had lower ($P<.05$) visual scores as a result of vacuum packaging. ST steaks from long-fed and silage-fed cattle were adversely ($P<.05$) affected by vacuum packaging. Within the non-vacuum packaged treatment ST steaks from short-fed cattle were scored highest ($P<.05$). Grass-fed ST muscle received the lowest ($P<.05$) score within the vacuum packaged treatment.

Except for grass-fed ST muscle, all nutritional regimes showed an increase ($P<.05$) in percent Mb⁺ when vacuum packaged. Between both packaging treatments, ST muscle from grass-fed cattle tended to have the lowest Mb⁺ percentages. Vacuum packaging evidently causes Mb in ST muscle to be more susceptible to oxidation in cattle that are fed on a high energy level.

%R 630 nm - %R 580 nm value of grass-fed ST muscle was increased ($P<.05$) by vacuum packaging and short-fed was decreased ($P<.05$). ST muscle from grass-fed cattle had the highest ($P<.05$) %R 630 nm - %R 580 nm values in both packaging treatments, supporting brighter visual color scores.

Biceps femoris: Vacuum packaging tended to lower BF visual scores for each nutritional regime, but the only differences ($P<.05$) existed in the short-fed and long-fed groups. Within

the non-vacuum treatment, short-fed and long-fed cattle had the highest ($P < .05$) BF visual scores, but no differences were found among nutritional regimes in the vacuum treatment.

Vacuum packaging did not alter Mb⁺ percentages in BF muscle for grass-fed, long-fed and silage-fed cattle, but did cause a lower Mb⁺ percentage in short-fed BF muscle, perhaps attributable to chance. The longer fed groups (long-fed and silage-fed) tended to have the greatest Mb⁺ levels within both packaging treatments.

%R 630 nm - %R 580 nm values for each nutritional regime were increased ($P < .05$) by vacuum packaging which tended to support visual observations. Within the non-vacuum grouping the values tended to decrease as feeding length increased. When vacuum packaged, the longer fed groups had lower ($P < .05$) values than the grass-fed and short-fed groups.

Semimembranosus: SM steaks from long-fed and silage-fed cattle had improved ($P < .05$) visual scores as a result of vacuum packaging. Silage-fed SM steaks showed the most improvement when vacuum packaged and had the lowest ($P < .05$) visual score. SM muscle from grass-fed cattle tended to have the highest visual scores when vacuum packaged. Grass-fed SM increased slightly when vacuum packaged, probably partially due to higher muscle pigment concentrations that may exist in grass-fed cattle compared to cattle from other regimes.

All nutritional treatments except silage-fed demonstrated an increase ($P < .05$) in Mb⁺ percentages when vacuum packaged. SM muscle from grass-fed cattle had the lowest ($P < .05$) proportion of myoglobin within the non-vacuum group. Short-fed had

the least ($P < .05$) within the vacuum packaged group. The large increase in Mb^+ noted for grass-fed supported the visual score and can likely be accounted for by bacterial contamination.

Increases ($P < .05$) in %R 630 nm - %R 580 nm occurred for all nutritional groups with vacuum packaging. Within the non-vacuum group, the values tended to decrease with length of feeding. Short-fed SM steaks had the highest ($P < .05$) value within the vacuum packaged group. Silage-fed tended to have the lowest.

Nutrition x packaging x day of display interaction.

This interaction had no effect ($P < .05$) on visual scores for any muscle so they are not listed with the other interaction means.

Longissimus: Mb^+ percentages were not different ($P < .05$) between packaging treatments for each nutritional regime through the display period for L steaks from short-fed, long-fed and silage-fed cattle (table 6). Steaks from grass-fed cattle showed similar behavior through day 3, but at day 5 the vacuum packaged L steaks from grass-fed cattle had more ($P < .05$) Mb^+ than the non-vacuum packaged steaks. Perhaps the inversion in Mb^+ existed because of bacterial growth that occurred on the surface of the vacuum packaged L steaks from grass-fed cattle at that time. This indicates vacuum packaged grass-fed beef is unstable over a long display. This may be related to possible higher pigment levels in grass-fed muscle and possible limited oxygen in packages.

Through day 1 vacuum packaged L steaks from all nutritional regimes had higher ($P < .05$) %R 630 nm - %R 580 nm values than the non-vacuum steaks. However, at day 3 only vacuum packaged L steaks from grass-fed and silage-fed cattle still had higher ($P < .05$) values. There was no difference for short-fed at day 3, but non-vacuum packaged L steaks from long-fed cattle showed a reversal to a higher ($P < .05$) value (brighter color) over vacuum packaged. By day 5 there was no difference in %R 630 nm - %R 580 nm values due to packaging within nutritional regime.

Semitendinosus: There was no difference ($P < .05$) in percent Mb⁺ for ST steaks from grass-fed cattle at any point in the display (table 7), although the vacuum treatment tended higher. ST steaks from short-fed cattle showed higher ($P < .05$) Mb⁺ levels for the vacuum groups at each evaluation period which means that short-fed ST was most adversely affected by vacuum packaging. ST from long-fed cattle had no difference in Mb⁺ as a result of packaging through day 1; silage-fed demonstrated no difference at day 0. However, greater ($P < .05$) Mb⁺ percentages in the vacuum packaged steaks at day 3 through day 5 and day 1 through day 5 occurred for long-fed and silage-fed, respectively. After day 1, ST muscle from grass-fed cattle definitely had the least ($P < .05$) Mb⁺ for all nutritional regimes.

%R 630 nm - %R 580 nm values for vacuum packaged steaks were greater ($P < .05$) at day 0 for all nutritional regimes and at day 1 for ST from grass-fed cattle ($P < .05$) than non-vacuum. ST steaks from grass-fed cattle were not affected by vacuum packaging at days

3 and 5, but the other nutritional regimes showed higher ($P < .05$) values (darker color) non-vacuum for as display progressed. For short-fed this occurred at day 1 and on day 3 for long-fed and silage-fed cattle. Overall, grass-fed ST muscle had higher ($P < .05$) values than that from other nutritional treatments, indicating ST steaks from grass-fed cattle had the most stable display color.

Biceps femoris: Table 8 shows no difference in percent Mb^+ at day 0 among any nutrition-packaging combinations. Among nutritional regimes at days 1 and 3 only vacuum packaged BF from short-fed cattle had less ($P < .05$) Mb^+ than non-vacuum when packaging was compared. All nutritional regimes except short-fed showed higher ($P < .05$) percentages of Mb^+ in the vacuum packaged treatment at day 5. Although vacuum packaging retarded Mb^+ formation early in the display, vacuum packaged BF muscle was more susceptible to oxidation over a long display.

Within each nutritional regime the vacuum packaged BF had a higher ($P < .05$) $\%R$ 630 nm - $\%R$ 580 nm value than non-vacuum at days 0 and 1. Vacuum packaged short-fed BF retained this advantage through day 3, thereby supporting the Mb^+ observation. The grass-fed and short-fed groups tended to have higher values at days 3 and 5 than the longer fed groups.

Semimembranosus: Vacuum packaged SM from groups fed a high grain ration (short-fed and long-fed) showed lower ($P < .05$) Mb^+ levels at day 0 than non-vacuum (table 9). SM steaks from grass-fed cattle showed a difference due to packaging at day 1, with the vacuum packaged SM having a higher ($P < .05$) Mb^+ percentage

than non-vacuum. This continued throughout the display for the grass-fed treatment. Again, the large increase in Mb⁺ in the vacuum packaged SM of grass-fed cattle may be attributed to the presence of bacteria on the surface of the steaks. Beginning at day 3, vacuum packaged long-fed SM also had more (P<.05) Mb⁺ than non-vacuum. By day 5, the vacuum packaged SM from all nutritional regimes except short-fed had a higher (P<.05) Mb⁺ level than non-vacuum. Generally, SM muscle from short-fed cattle seemed to be less affected by vacuum packaging than the other nutritional treatments.

%R 630 nm - %R 580 nm values were higher (P<.05) for vacuum packaged SM within each nutritional treatment at day 0. This was true at day 1 as well for all but the grass-fed group. The long-fed group showed a higher (P<.05) value at day 3 for non-vacuum. Both the silage-fed and short-fed groups continued to show higher (P<.05) values for the vacuum packaged SM steaks at day 3. Day 5 results illustrate a lower (P<.05) value for vacuum packaged SM compared with non-vacuum in the grass-fed treatment and a continued advantage in the short-fed group for vacuum packaged SM muscle. Overall, SM muscle from silage-fed cattle tended to have the lowest %R 630 nm - %R 580 nm values at each day of display when similar packaging treatments were compared.

Table 3. Effect of nutritional regime on mean visual score, percent Mb⁺ and %R 630 nm - %R 580 nm of four beef muscles.

	Visual Score ^d	Percent Mb ⁺ e	%R 630 nm - %R 580 nm
Longissimus			
Grass-fed ^f	2.90	18.67 ^b	23.15 ^a
Short-fed ^g	2.88	16.26 ^a	22.63 ^a
Long-fed ^h	2.80	19.31 ^b	22.01 ^{ab}
Silage-fed ⁱ	2.66	19.90 ^b	20.92 ^b
Semitendinosus			
Grass-fed	2.04	8.90 ^a	33.29 ^a
Short-fed	2.34	14.80 ^b	29.54 ^b
Long-fed	2.20	17.55 ^c	28.12 ^{bc}
Silage-fed	2.18	16.60 ^c	27.17 ^c
Biceps femoris			
Grass-fed	3.08	33.06 ^a	16.30 ^{ab}
Short-fed	3.32	32.09 ^a	16.43 ^a
Long-fed	3.32	36.06 ^b	14.92 ^{bc}
Silage-fed	3.12	36.90 ^b	13.73 ^c
Semimembranosus			
Grass-fed	3.26	27.89 ^a	19.23 ^a
Short-fed	3.26	27.96 ^a	19.62 ^a
Long-fed	3.23	30.40 ^b	18.54 ^b
Silage-fed	3.06	30.89 ^b	16.59 ^b

a-c Observations within the same muscle-color-parameter block bearing similar or no superscript letters are not different (P<.05).

d 1=very bright red, 2=bright red, 3=slightly dark red or brown, 4=dark red or brown, 5=exceptionally dark red or brown.

e Calculated according to Stewart *et al.* (1965)

f Grazed through the summer on brome-bluestem pasture with no additional concentrate.

g 20% alfalfa haylage (IRN 3-08-151), 75.2% cracked corn (IRN 4-02-932), 4.8% SBM supplement (IRN 5-04-604) ration in drylot for 49 days.

h Fed same ration as short-fed in drylot for 98 days.

i 40% corn silage (IRN 3-02-824), 20% alfalfa haylage (IRN 3-08-151), 36% cracked corn (IRN 4-02-932), 4% SBM supplement (IRN 5-04-604) ration in drylot for 98 days.

Table 4. Nutritional regime x day of display interaction on mean visual score, percent Mb+ and %R 630 nm - %R 580 nm of four beef muscles.

	Visual ^k , day					Percent Mb+ ^l , day					%R 630 nm - %R 580 nm, day				
	0	1	2	3	5	0	1	2	3	5	0	1	2	3	5
Longissimus:															
Grass-fed ^m	2.50 ^b	2.44 ^{ab}	3.25 ^{hi}	3.41 ⁱ	3.41 ⁱ	9.12 ^{ab}	15.27 ^c	20.08 ^{de}	30.21 ^h	25.50 ^a	24.91 ^{ab}	23.23 ^c	18.98 ^{gh}	20.69 ^f	18.45 ^h
Short-fed ^o	2.28 ^b	2.81 ^{bc}	2.97 ^{fg}	3.47 ^{gh}	3.09 ^{gh}	7.95 ^a	15.15 ^c	19.71 ^f	22.25 ^g	23.06 ^{cd}	24.76 ^{ab}	22.00 ^f	20.69 ^f	18.45 ^h	17.91 ⁱ
Long-fed ^p	2.50 ^b	2.59 ^{bcd}	3.00 ^{fg}	3.09 ^{gh}	2.84 ^{ef}	10.52 ^{ab}	17.98 ^d	22.99 ^f	25.76 ^g	25.27 ^a	23.91 ^{bc}	20.43 ^{fe}	18.45 ^h	17.91 ⁱ	17.91 ⁱ
Silage-fed ^p	2.44 ^{ab}	2.62 ^{bcd}	2.75 ^{cde}	2.84 ^{ef}	2.84 ^{ef}	11.39 ^b	18.46 ^d	22.76 ^f	27.00 ^g	23.97 ^{bc}	21.77 ^e	20.05 ^{fe}	17.91 ⁱ	17.91 ⁱ	17.91 ⁱ
Semitendinosus:															
Grass-fed	1.56 ^a	1.87 ^{bc}	2.25 ^{do}	2.47 ^{fg}	2.47 ^{fg}	2.46 ^a	7.61 ^c	11.47 ^d	14.07 ^{ef}	35.24 ^a	34.40 ^{ab}	32.79 ^{cd}	30.72 ^e	25.10 ^f	22.63 ^j
Short-fed	1.94 ^{bc}	1.97 ^{bc}	2.53 ^g	2.94 ^h	2.94 ^h	3.32 ^a	11.52 ^d	20.22 ^e	24.15 ⁱ	33.33 ^{bc}	32.25 ^d	27.47 ^e	25.10 ^f	22.63 ^j	22.63 ^j
Long-fed	1.78 ^b	1.94 ^{bc}	2.34 ^{efg}	2.75 ^{fg}	2.75 ^{fg}	4.67 ^{ab}	14.72 ^f	22.89 ^{hi}	27.94 ^j	32.51 ^{cd}	31.13 ^f	26.20 ^h	22.63 ^j	22.63 ^j	22.63 ^j
Silage-fed	1.81	2.12	2.31	2.47 ^{fg}	2.47 ^{fg}	6.31 ^{bc}	14.27 ^f	20.54 ^{gh}	25.27 ^g	30.92	29.41 ^f	25.60 ^h	22.74 ^j	22.74 ^j	22.74 ^j
Biceps femoris:															
Grass-fed	2.44 ^a	2.81 ^{bc}	3.44 ^{fg}	3.66 ^{hi}	3.66 ^{hi}	14.18 ^a	28.25 ^b	39.30 ^e	50.51 ^f	23.27 ^a	17.18 ^c	13.94 ^e	10.80 ^{fe}	11.75 ^{hi}	9.61 ⁱ
Short-fed	2.62 ^{ab}	3.22 ^{de}	3.62 ^{gh}	3.81 ^{ij}	3.81 ^{ij}	13.62 ^a	29.14 ^{bc}	40.35 ^f	45.24 ^f	22.67 ^{ab}	17.48 ^d	13.84 ^e	11.75 ^{hi}	9.61 ⁱ	9.61 ⁱ
Long-fed	2.66 ^b	3.16 ^{cd}	3.53 ^{ef}	3.94 ^{gh}	3.94 ^{gh}	13.33 ^a	31.39 ^d	47.15 ^f	52.36 ^g	23.22 ^b	15.97 ^e	10.88 ^{fe}	9.61 ⁱ	9.61 ⁱ	9.61 ⁱ
Silage-fed	2.66	3.00	3.34	3.50 ^{gh}	3.50 ^{gh}	14.28 ^a	34.25 ^d	46.78 ^f	52.30 ^g	21.64 ^b	13.88 ^e	10.38 ^{gh}	9.02 ⁱ	9.02 ⁱ	9.02 ⁱ
Semimembranosus:															
Grass-fed	2.66 ^{ab}	2.91 ^{cd}	3.56 ^h	3.94 ⁱ	3.94 ⁱ	12.99 ^{ab}	22.41 ^c	31.51 ^f	44.65 ^j	25.16 ^a	21.04 ^c	17.26 ^d	13.46 ^{gh}	15.04 ^e	12.50 ^h
Short-fed	2.50 ^a	3.09 ^{cd}	3.41 ^{gh}	4.06 ⁱ	4.06 ⁱ	11.35 ^a	24.81 ^{cd}	35.40 ^h	40.28 ^{hi}	26.03 ^a	20.72 ^c	16.71 ^{cf}	14.06 ^h	15.04 ^e	12.50 ^h
Long-fed	2.62 ^{ab}	2.91 ^{cd}	3.31 ^{ef}	4.06 ⁱ	4.06 ⁱ	13.90 ^b	25.64 ^d	39.50 ^h	42.55 ^{ij}	25.09 ^b	20.15 ^c	14.86 ^{fe}	14.06 ^h	15.04 ^e	12.50 ^h
Silage-fed	2.75	2.94 ^{cd}	3.12 ^{ef}	3.44 ^{gh}	3.44 ^{gh}	15.12 ^b	27.95 ^e	38.00 ⁱ	42.50 ^{ij}	22.72 ^b	17.18 ^d	13.95 ^{fe}	12.50 ^h	15.04 ^e	12.50 ^h

a-j Observations within the same muscle-color parameter block bearing similar superscript letters are not different (P<.05).

k1=very bright red, 2=bright red, 3=slightly dark red or brown, 4=dark red or brown, 5=exceptionally dark red or brown

l Calculated according to Stewart *et al.* (1965).

m-P See footnotes f-1, table 3.

Table 5. Nutritional regime x packaging interaction on mean visual score, percent Mb+ and $\%R$ 630 nm - $\%R$ 580 nm of four beef muscles.

	Visual		Percent Mb+ ^h		$\%R$ 630 nm - $\%R$ 580 nm	
	Non-vac ⁱ	Vac	Non-Vac	Vac	Non-Vac	Vac
Longissimus						
Grass-fed ^r	3.01 ^d	2.78 ^c	18.11 ^{bc}	19.23 ^{bc}	21.89 ^{cde}	24.42 ^a
Short-fed ^l	3.23 ^e	2.53 ^b	15.15 ^a	17.38 ^{ab}	21.57 ^{de}	23.69 ^{ab}
Long-fed ^m	3.01 ^d	2.58 ^b	19.17 ^{bc}	19.45 ^{bc}	20.92 ^e	23.11 ^{abc}
Silage-fed ⁿ	3.06 ^{de}	2.26 ^a	20.66 ^c	19.15 ^{bc}	19.14 ^f	22.71 ^{bcd}
Semitendinosus						
Grass-fed	2.14 ^{bc}	1.94 ^a	8.42 ^a	9.39 ^a	32.45 ^b	34.13 ^a
Short-fed	2.34 ^d	2.34 ^d	9.72 ^a	19.88 ^c	30.63 ^c	28.45 ^d
Long-fed	2.06 ^{ab}	2.34 ^d	14.49 ^b	20.62 ^c	28.46 ^d	27.77 ^{de}
Silage-fed	2.05 ^{ab}	2.31 ^{cd}	13.61 ^b	19.59 ^c	27.73 ^{de}	26.60 ^e
Biceps femoris						
Grass-fed	3.17 ^{ab}	3.06 ^a	32.55 ^{ab}	33.57 ^{bc}	15.20 ^{bc}	17.40 ^a
Short-fed	3.55 ^c	3.09 ^{ab}	33.55 ^{bc}	30.63 ^a	14.45 ^c	18.41 ^a
Long-fed	3.47 ^c	3.17 ^{ab}	35.26 ^{bcd}	36.85 ^d	13.91 ^{cd}	15.93 ^b
Silage-fed	3.22 ^b	3.03 ^{ab}	36.25 ^{cd}	37.55 ^d	12.90 ^d	14.56 ^c
Semimembranosus						
Grass-fed	3.20 ^{bc}	3.33 ^{cd}	23.90 ^a	31.88 ^d	18.51 ^d	19.96 ^{bc}
Short-fed	3.34 ^{cd}	3.19 ^{bc}	29.67 ^{cd}	26.26 ^{ab}	17.01 ^e	22.24 ^a
Long-fed	3.44 ^d	3.01 ^b	28.45 ^{bc}	32.30 ^d	16.92 ^e	20.16 ^b
Silage-fed	3.34 ^{cd}	2.78 ^a	30.71 ^{cd}	31.08 ^{cd}	14.50 ^f	18.68 ^{cd}

a-f Observations within the same muscle-color parameter block bearing similar superscript letters are not different ($P < .05$).

^g1=very bright red, 2=bright red, 3=slightly dark red or brown, 4=dark red or brown, 5=exceptionally dark red or brown.

^hCalculated according to Stewart *et al.* (1965)

ⁱFabricated after 48 hour carcass chill.

^jFabricated after 21 day storage in vacuum bag at 2°C.

k-n See footnotes f-i, table 3.

Table 6. Nutritional regime x packaging x day of display interaction on mean percent Mb+ and %R 630 nm - %R 580 nm of beef longissimus muscle.

	Percent Mb+ ^r , day					%R 630 nm - %R 580 nm, day				
	0	1	2	3	5	0	1	2	3	5
Grass-fed: ^s										
Non-vac ^w	8.26 ^{ab}	13.98 ^{def}	21.81 ^{hkl}	28.38 ⁿ		23.45 ^{ef}	23.39 ^f	21.51 ^{ghi}	19.22 ^{lmnop}	
Vac ^x	9.98 ^{abc}	16.56 ^{fg}	18.36 ^{ghi}	32.04 ^o		27.55 ^{ab}	26.43 ^{bcd}	24.95 ^{de}	18.74 ^{mnopq}	
Short-fed: ^t										
Non-vac	7.45 ^a	14.11 ^{def}	18.54 ^{ghi}	20.49 ^{hijk}		20.53 ^{ijkl}	23.05 ^f	21.46 ^{ghij}	21.22 ^{ghijkl}	
Vac	8.45 ^{ab}	16.20 ^{efg}	20.87 ^{ijk}	24.01 ^{klm}		25.59 ^{cd}	26.47 ^{bc}	22.53 ^{fg}	20.16 ^{ijklm}	
Long-fed: ^u										
Non-vac	11.75 ^{bcd}	18.83 ^{ghi}	21.42 ^{ijkl}	24.67 ^{lm}		21.53 ^{ghi}	22.14 ^{fgh}	21.44 ^{ghij}	18.56 ^{nopq}	
Vac	9.28 ^{abc}	17.13 ^{fgh}	24.56 ^{lm}	26.85 ^{mn}		29.00 ^a	25.67 ^{cd}	19.42 ^{lmnop}	18.35 ^{opq}	
Silage-fed: ^v										
Non-vac	12.62 ^{cde}	19.05 ^{ghi}	23.70 ^{klm}	27.26 ^{mn}		19.83 ^{klmno}	19.99 ^{ijklmn}	19.15 ^{lmnop}	17.59 ^q	
Vac	10.16 ^{abc}	17.87 ^{ghi}	21.82 ^{hkl}	26.75 ^{mn}		28.12 ^a	23.54 ^{ef}	20.95 ^{hijkl}	18.23 ^{pq}	

^{a-q}Observations within the same color parameter block bearing similar superscript letters are not different (P<.05).

^rCalculated according to Stewart et al. (1965).

^{s-v}See footnotes f-i, table 3.

^wFabricated after 48 hour carcass chill.

^xFabricated after 21 day storage in vacuum bag at 2°C.

Table 7. Nutritional regime x packaging x day of display interaction on mean percent Mb+ and %R 630 nm - %R 580 nm of beef semitendinosus muscle.

	Percent Mb+ ^P , day				%R 630 nm - %R 580 nm, day			
	0	1	2	5	0	1	2	5
Grass-fed ^q , Non-vac ^u Vac ^v	2.44 ^{ab} 2.47 ^{ab}	7.50 ^{cd} 7.72 ^{cd}	10.90 ^{de} 12.04 ^{ef}	12.84 ^{efg} 15.31 ^{feh}	31.82 ^{fg} 38.67 ^a	33.17 ^e 35.63 ^{bc}	32.99 ^e 32.59 ^{ef}	31.81 ^{fg} 29.62 ^{hg}
Short-fed ^r , Non-vac Vac	1.49 ^a 8.14 ^{cd}	7.57 ^{cd} 15.47 ^{feh}	14.93 ^{feh} 25.50 ^{lm}	17.88 ^{hij} 30.42 ⁿ	29.87 ^{hg} 36.79 ^b	33.53 ^{de} 30.96 ^{gh}	30.27 ^{hg} 24.68 ^{lm}	28.84 ^{gh} 21.36 ⁿ
Long-fed ^s , Non-vac Vac	3.40 ^{ab} 5.93 ^{bc}	13.38 ^{efg} 16.06 ^{ghij}	19.08 ^{ijk} 26.69 ^m	22.11 ^{kl} 33.78 ⁿ	28.09 ^h 36.92 ^b	31.35 ^e 30.91 ^{gh}	29.19 ^{gh} 23.22 ^m	25.22 ^{kl} 20.04 ^{no}
Silage-fed ^t , Non-vac Vac	4.97 ^{abc} 7.65 ^{cd}	12.36 ^{ef} 16.18 ^{ghij}	17.34 ^{hij} 23.74 ^{lm}	19.76 ^{jk} 30.79 ⁿ	26.91 ^{ij} 34.94 ^{cd}	29.97 ^{hg} 26.85 ^{gh}	27.81 ^{hi} 23.39 ^m	26.23 ^{jk} 19.24 ^o

^{a-o}Observations within the same color parameter block bearing similar superscript letters are not different (P<.05).

^PCalculated according to Stewart et al. (1965).

^{q-t}See footnotes f-i, table 3.

^uFabricated after 48 hour carcass chill.

^vFabricated after 21 day storage in vacuum bag at 2°C.

Table 8. Nutritional regime x packaging x day of display interaction on mean percent Mb+ and %R 630 nm - %R 580 nm of beef biceps femoris muscle.

	Percent Mb ⁺ , day					%R 630 nm - %R 580 nm, day				
	0	1	2	3	4	0	1	2	3	4
Grass-fed ^s :										
Non-vac	15.15 ^a	28.38 ^{bc}	40.08 ^{gh}	46.59 ^{klm}		19.18 ^{cd}	16.36 ^{fg}	13.65 ^{ijk}	11.59 ^{lmn}	
Vac	13.21 ^a	28.13 ^{bc}	38.52 ^{fg}	54.43 ⁿ		27.37 ^a	18.00 ^{de}	14.23 ^{hij}	10.01 ^{opq}	
Short-fed ^t :										
Non-vac	12.40 ^a	32.37 ^{de}	42.61 ^{hi}	46.84 ^{klm}		18.58 ^{cde}	15.17 ^{gh}	12.75 ^{ijk}	11.31 ^{mno}	
Vac	14.85 ^a	25.92 ^b	38.09 ^{fg}	43.65 ^{hik}		26.76 ^a	19.78 ^c	14.92 ^{ghl}	12.19 ^{klm}	
Long-fed ^u :										
Non-vac	13.13 ^a	32.44 ^{de}	45.67 ^{ikl}	49.82 ^m		19.60 ^c	14.69 ^{hi}	11.32 ^{mno}	10.03 ^{opq}	
Vac	13.52 ^a	30.33 ^{cd}	48.63 ^{lm}	54.90 ⁿ		26.84 ^a	17.26 ^{ef}	10.44 ^{nop}	9.18 ^{pq}	
Silage-fed ^v :										
Non-vac	13.00 ^a	35.26 ^{ef}	46.61 ^{klm}	50.12 ^m		18.84 ^{cd}	12.93 ^{jkl}	10.58 ^{nop}	9.26 ^{pq}	
Vac	15.56 ^a	33.23 ^{de}	46.95 ^{klm}	54.47 ⁿ		24.45 ^b	14.84 ^{hi}	10.18 ^{nopq}	8.78 ^q	

^{a-q}Observations within the same color parameter block bearing similar superscript letters are not different (P<.05).

^rCalculated according to Stewart et al. (1965).

^{s-v}See footnotes f-i, table 3.

^wFabricated after 48 hour carcass chill.

^xFabricated after 21 day storage in vacuum bag at 2°C.

Table 9. Nutritional regime x packaging x day of display interaction on mean percent Mb+ and %R 630 nm - %R 580 nm of beef semimembranosus muscle.

	Percent Mb ⁺ , day					%R 630 nm - %R 580 nm, day				
	0	1	2	3	5	0	1	2	3	5
Grass-fed ^q :										
Non-vac ^u	14.6 ^{8cd}	19.12 ^e	27.20 ^f	34.58 ^h		20.27 ^{ef}	20.29 ^{ef}	17.96 ^{gh}	15.49 ^{ijk}	
Vac ^v	11.30 ^{abc}	25.70 ^f	35.82 ^{hi}	54.72 ⁿ		30.04 ^b	21.78 ^{de}	16.56 ^{hi}	11.43 ^o	
Short-fed ^r :										
Non-vac	13.44 ^{bcd}	25.69 ^f	36.38 ^{hi}	43.15 ^{kl}		20.30 ^{ef}	18.21 ^e	15.51 ^{ijk}	14.00 ^{klm}	
Vac	9.27 ^a	23.92 ^f	34.42 ^h	37.41 ^{hij}		31.75 ^a	23.24 ^d	17.91 ^{gh}	16.07 ^{ij}	
Long-fed ^s :										
Non-vac	17.00 ^{de}	25.69 ^f	34.46 ^h	36.84 ^{hi}		19.19 ^f	18.58 ^e	15.19 ^{ijk}	14.73 ^{jkl}	
Vac	10.81 ^{ab}	25.59 ^f	44.54 ^l	48.26 ^m		30.99 ^{ab}	21.73 ^{de}	14.53 ^{kl}	13.39 ^{lmn}	
Silage-fed ^t :										
Non-vac	16.26 ^{de}	27.74 ^e	38.32 ^{ij}	40.51 ^{jk}		17.93 ^{gh}	15.38 ^{ijk}	12.63 ^{mno}	12.06 ^{no}	
Vac	13.98 ^{bcd}	28.15 ^e	37.69 ^{hij}	44.50 ^l		27.50 ^c	18.98 ^f	15.28 ^{ijk}	12.95 ^{mn}	

^{a-o} Observations within the same color parameter block bearing similar superscript letters are not different (P<.05).

^p Calculated according to Stewart et al. (1965).

^{q-t} See footnotes f-i, table 3.

^u Fabricated after 48 hour carcass chill.

^v Fabricated after 21 day storage in vacuum bag at 2°C.

SUMMARY

A comparison of visual color scores of four muscles from cattle fed grass only, a 60% forage ration for 98 days in drylot (silage fed) or a 75% grain ration in drylot for 49 (short-fed) and 98 days (long-fed), respectively, showed no difference ($P < .05$) due to nutritional regime. Percent metmyoglobin (Mb^+) determined spectrophotometrically was greatest ($P < .05$) in steaks from the longer fed cattle and least in the steaks from short-fed and grass-fed cattle. %R 630 nm - %R 580 nm values coincided with Mb^+ levels.

Longissimus (L) steaks from all nutritional regimes had acceptable visual scores through the third day (day 3) of display, but those from short-fed and grass-fed cattle were marginally acceptable by the fifth day (day 5). L steaks from silage-fed cattle were brightest ($P < .05$) at days 3 and 5. In addition, L steaks from grass-fed cattle had the highest percent Mb^+ at day 5.

Semitendinosus (ST) muscle from all nutritional regimes had brighter visual scores and higher %R 630 nm - %R 580 nm values through the display period indicating ST muscle was less affected over a lengthy display. Nutritional background does not present problems in display color stability of ST steaks.

Biceps femoris (BF) steaks from grass-fed cattle had lower ($P < .05$) visual scores at day 1 than BF steaks from cattle fed grain. Color of BF steaks from all nutritional treatments was acceptable through day 1, but all were unacceptable or marginally so by the third display day.

BF muscle from those cattle fed a high forage or high grain ration for 98 days had the highest ($P < .05$) Mb^+ levels at day 3. A large increase in Mb^+ for BF steaks from grass-fed cattle was noted between days 3 and 5 which coincided with possible development of bacterial contamination on the steak surface. %R 630 nm - %R 580 nm values for BF steaks followed the pattern established by Mb^+ percentages as a function of length of feeding.

Visual color of semimembranosus (SM) steaks from all nutritional regimes was rated as unacceptable or closely unacceptable after three days display. Only small differences in percent Mb^+ in SM steaks were noticed through day 1, but by the third day of display, SM steaks from the longer fed groups had the most ($P < .05$) Mb^+ . Again, a large increase in Mb^+ occurred between days 3 and 5 in conjunction with the appearance of apparent bacterial contamination. Decreased %R 630 nm - %R 580 nm values were in general agreement with increases in Mb^+ .

Vacuum packaging resulted in brighter visual color for L muscle from all nutritional regimes at the initial reading compared to conventional packaging. L muscle from grass-fed cattle had the darkest (highest) ($P < .05$) visual score. Within nutritional regimes, vacuum packaging had no effect on percent Mb^+ . %R 630 nm - %R 580 nm values were higher for vacuum packaged L muscle.

Only ST steaks from grass-fed cattle had lower ($P < .05$) visual scores as a result of vacuum packaging. Except for the grass-

fed group, ST muscle from all nutritional regimes showed an increase in Mb^+ when vacuum packaged. It appears that vacuum packaging may cause Mb in ST muscle to be more susceptible to oxidation in cattle fed on a high energy level. This is supported by increased ($P<.05$) %R 630 nm - %R 580 nm values for grass-fed ST muscle.

Vacuum packaging tended to lower BF visual scores for each nutritional regime. Packaging did not alter Mb^+ percentages in BF muscle for grass-fed, long-fed and silage-fed cattle, but did lower Mb^+ in short-fed BF muscle. BF muscle from longer fed groups (long-fed and silage-fed) tended to have the greatest Mb^+ levels within both packaging treatments. %R 630 nm - %R 580 nm values for BF muscle from each nutritional regime were increased ($P<.05$) by vacuum packaging which supported visual observations.

Visual scores of SM muscle from long-fed and silage-fed cattle were improved ($P<.05$) as a result of vacuum packaging. All nutritional treatments except silage-fed demonstrated increased ($P<.05$) Mb^+ in SM muscle when vacuum packaged. Increased ($P<.05$) in %R 630 nm - %R 580 nm occurred for SM muscle from all nutritional groups.

Mb^+ percentages were not different between packaging treatments for each nutritional regime through the display period for L steaks from short-fed, long-fed and silage-fed cattle. Steaks from grass-fed cattle showed similar behavior through day 3, but at day 5 the vacuum packaged L steaks from grass-fed cattle had more ($P<.05$) Mb^+ than the non-vacuum packaged steaks.

Through day 1 vacuum packaged L steaks from all nutritional regimes had higher ($P<.05$) %R 630 nm - %R 580 nm values than the non-vacuum L steaks. By day 5 there was no difference due to packaging within nutritional regime.

After day 1, ST muscle from grass-fed cattle definitely had the least Mb⁺ for all nutritional regimes. ST steaks from short-fed cattle showed higher ($P<.05$) Mb⁺ levels for the vacuum groups at each evaluation period which means that short-fed ST was most adversely affected by vacuum packaging. Overall, ST muscle from grass-fed cattle had higher ($P<.05$) %R 630 nm - %R 580 nm values than those from other nutritional treatments, indicating ST steaks from grass-fed cattle have the most stable display color.

Although vacuum packaging seemed to retard Mb⁺ formation early in the display period, vacuum packaged BF muscle from all nutritional regimes appears to be more susceptible to oxidation over a long display. Within each nutritional regime the vacuum packaged BF had a higher ($P<.05$) %R 630 nm - %R 580 nm value than non-vacuum at days 0 and 1.

Vacuum packaged SM from groups fed a high grain ration showed lower ($P<.05$) Mb⁺ levels at day 0 than non vacuum. By day 5, vacuum SM from all nutritional regimes except short-fed had a higher ($P<.05$) Mb⁺ level than the non-vacuum packaged SM steaks. Generally, SM muscle from short-fed cattle seemed to be less affected by vacuum packaging than the other nutritional treatments. %R 630 nm - %R 580 nm values were higher ($P<.05$) for vacuum packaged SM within each nutritional treatment at day 0.

Overall, SM muscle from silage-fed cattle tended to have the lowest %R 630 nm - %R 580 nm values at each day of display when similar packaging treatments were compared.

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CHAPTER 5

THE RELATIONSHIP OF VARIOUS SPECTROPHOTOMETRIC REFLECTANCE MEASUREMENTS TO BEEF MUSCLE VISUAL COLOR SCORE.

INTRODUCTION

Greater efficiencies in production, marketing and distribution of beef can result from modification of feeding, processing and packaging methods. Whether the increased efficiencies will be realized is primarily dependent on consumer acceptance of beef produced utilizing alternative methods.

Color is a major criteria for consumer acceptance of beef. Any undesirable beef color changes resulting from alternative methods of production, processing or packaging may cause consumer rejection.

Correlation coefficients presented in this chapter give information on objective predictors of muscle color from cattle produced on widely different nutritional regimes using four muscles packaged in two ways. Correlations gathered using cattle from such diverse backgrounds may result in a single objective measurement that can be used as a predictor of visual muscle color from beef produced and processed in different ways.

MATERIALS AND METHODS

Longissimus (L), semitendinosus (ST), biceps femoris (BF) and semimembranosus (SM) muscles from cattle on widely different nutritional regimes (Materials and Methods, Chapter 3) were used. Intact muscles were removed 48 hours post-slaughter and transversely halved. The anterior (proximal) half was fabricated into 2.54 cm thick steaks. The posterior (distal) half was placed into a vacuum bag and stored 21 days at 2°C before fabrication. Steaks were displayed in a styrofoam tray overwrapped with PVC (polyvinyl chloride) film for five days at 2°C under continuous (24 hours/day) General Electric Delux Warm White lighting at an intensity of 1076 lumens/m² (100 foot candles) at meat surface level. Lighting consisted of two 40 watt tubes 126 cm from the muscle surface.

Subjective muscle color was scored individually by five panelists under display lighting at initial display (day 0) and at one, three and five days of display to the nearest .5 point using a scale of 1 = very bright red, 2 = bright red, 3 = slightly dark red or brown, 4 = dark red or brown and 5 = extremely dark red or brown (Kansas State University).

Objective color was measured at days 0, 1, 3 and 5 days of display with a Bausch and Lomb 600 Spectrophotometer with reflectance with MgCO₃ blocks. Reflectance spectra were scanned from 400 nm to 700 nm at a speed of 50 nm/min. Reflectance was measured at 10 nm intervals and printed out with an Autolab Minigrator connected to the spectrophotometer and set for a peak height mode. Reflectance at intermediate points was determined by interpolation.

Numerous reflectance parameters were determined, including seven reflectance differences or ratios (%R 630 nm - %R 580 nm, %R 630 nm/%R 580 nm, %R 632 nm - %R 614 nm, %R 582 nm/%R 525 nm, %R 572 nm/%R 525 nm, %R 507 nm/%R 572 nm and %R 474 nm/%R 597 nm) as well as absorption coefficient/scattering coefficient according to Judd and Wysecki (1963).

Pooled within cell correlation coefficients of selected objective reflectance measurements to average visual score of five panelists were calculated. Considering each muscle separately, multiple regression (stepwise deletion) was applied to the same data to select the objective measurement(s) most closely related to subjective scores (Snedecor and Cochran, 1967).

RESULTS AND DISCUSSION

Pooled within cell correlation coefficients were high for all reflectance parameters studied except for %R 474 nm/%R 597 nm and its K/S adjustment (table 10). Since this reflectance value in K/S form had originally been suggested by Dean and Ball (1960) as being useful for determining proportion of reduced myoglobin (Mb), the low correlations should be expected when measuring color deterioration caused largely by metmyoglobin (Mb⁺) and perhaps other oxidized pigment forms. The ratios may be of use very early in the display, but should be poor as Mb is converted to MbO₂ and Mb⁺. The very low coefficients presented in table 10 for %R 474 nm/%R 597 nm (-.05) and the K/S adjustment (.03) disagree with the findings of Leising (1975) who reported values of -.65 to -.66 and .64 to .68, respectively, for frozen beef longissimus muscle after seven days display. Possibly at that point in display a reducing atmosphere existed which encouraged the conversion of pigment forms to Mb.

Highest correlation coefficients were recorded for unadjusted reflectance at 630 nm/580 nm (-.84), 507 nm/572 nm (-.84), 572 nm/525 nm (.83), 630 nm - 580 nm (-.82) and K/S adjustment of 572 nm/525 nm (-.85) and 507 nm/572 nm (.83).

Strange et al. (1974) reported simple correlation coefficients to visual score of -.86 for both %R 630 nm - %R 580 nm and %R 630 nm/%R 580 nm using a 50 point visual scale and a ten member consumer scoring panel. They did not report correlations to visual scores for those reflectance measurements adjusted for

K/S. Correlation coefficients of $-.82$ and $-.81$ for %R 630 nm - %R 580 nm to visual scores of two panelists were determined by Leising (1975) on frozen beef longissimus muscle.

Color estimates using reflectance at 630 nm and 580 nm should be fairly accurate since the difference of these two measurements has been shown by Van den Oord and Wesdorp (1971) to be determined by the relative proportions of oxymyoglobin (MbO_2) and Mb^+ . The MbO_2 reflectance spectrum has a minima between 540 nm and 580 nm and a maxima in the 600 to 700 nm region. Mb^+ has increased reflectance at 540 to 580 nm and decreased reflectance at 630 nm.

The high correlation coefficients for %R 507 nm/%R 573 nm and its K/S adjustment are expected since this ratio has been shown by Dean and Ball (1960) and Clydesdale and Francis (1971) to be useful in measuring Mb^+ . In the same vein, K/S 572 nm/K/S 525 nm has been used by several researchers to determine Mb^+ (Stewart et al., 1965; Snyder and Armstrong, 1967; Zimmerman and Snyder, 1969; and Ledward, 1970) and would also be expected to give high correlations to visual scores.

Eagerman, Clydesdale and Francis (1974) used %R 614 nm to report proportions of Mb, MbO_2 and Mb^+ . The difference was greatest for Mb, intermediate for MbO_2 and least or negative for Mb^+ . Leising (1975) found correlations of $-.78$ and $-.77$ for %R 632 nm - %R 614 nm to two visual scorers using beef longissimus muscle. This compares with results in table 10.

Kropf et al. (1976) reported that %R 582 nm/%R 525 nm should be an indicator of Mb⁺ portion of total pigment; however, the correlation coefficients reported in table 10 suggest that it is not as good an indicator as other ratios mentioned previously for measuring Mb⁺.

Numerous factors likely contributed to failure to achieve higher correlation coefficients between objective and visual scores. Many steaks demonstrated uneven surface discoloration which presented the panelist with the problem of averaging. Because of the small light beam (.3 cm x .5 cm) of the spectrophotometer, it is unlikely that a representative area was covered by the light beam although great care was taken to select a representative area. Perhaps the use of an instrument that covers a larger surface area or one of the recently introduced photoacoustic machines would give higher correlation coefficients.

The amount of dried juices on the covering film or degree of film glossiness could affect both objective and subjective evaluations. In addition, consistent visual scoring requires that the type and intensity of lighting be the same at all time periods. Even with structured visual scales with reference pictures, scorer variability may have been a problem.

Snyder (1965) discussed problems of reflectance measurements. Percent reflectance will depend on pigment concentration, amount of intramuscular fat and/or moisture. Effects of sample presentation were discussed by Clydesdale and Francis (1975). Uneven, concave or convex surface and "pillowing" are possible with muscle samples. Fiber orientation will also affect reflectance.

Table 11 shows that %R 630 nm - %R 580 nm made the greatest contribution to R^2 in L, BF and SM muscle. Although %R 630 nm - %R 580 nm was not the greatest contributor to R^2 in ST muscle, it made a significant ($P < .05$) contribution.

Because %R 630 nm - %R 580 nm had a high correlation coefficient (-.82) to visual color score and was the greatest contributor to R^2 in three of four muscles, its use as an objective predictor of visual score is recommended.

Table 10. Pooled within cell correlation of reflectance parameters to visual color scores (unadjusted vs. adjusted for K/S) for unfrozen beef muscle.^a

<u>nm</u>	<u>R^b</u>	<u>K/S^c</u>
630 - 580	-.82	-.71
630/580	-.84	-.71
632 - 614	-.79	.79
582/525	.62	-.57
572/525	.83	-.85
507/572	-.84	.83
474/597	-.05	.03

^an=1185, color evaluated at days 0, 1, 3 and 5 of display under 1076 lm/m² for 24 hrs/day at 20°C.

^b% reflectance, unadjusted.

^c% reflectance, adjusted for absorption coefficient/scattering coefficient.

Table 11. Objective measurements contributing most to R^2 of equations predicting visual color score of four beef muscles.

	Objective Measurement	R^2 contribution ^a
<u>Longissimus</u> ^b :	%R 630 nm - %R 580 nm	.334
	K/S 630 nm - K/S 580 nm	.018
	K/S 630 nm / K/S 580 nm	.009
	K/S 474 nm / K/S 597 nm	.003
	K/S 632 nm - K/S 614 nm	.008
	%R 632 nm - %R 614 nm	.017
	K/S 507 nm / K/S 572 nm	.003
<u>Semitendinosus</u> ^c :	%R 582 nm / %R 525 nm	.179
	%R 632 nm - %R 614 nm	.010
	K/S 632 nm - K/S 614 nm	.041
	%R 630 nm / %R 580 nm	.003
	%R 630 nm - %R 580 nm	.003
	%R 507 nm / %R 572 nm	.003
	K/S 474 nm / K/S 597 nm	.003
	%R 572 nm / %R 525 nm	.002
	K/S 507 nm / K/S 572 nm	.003
<u>Biceps femoris</u> ^d :	%R 630 nm - %R 580 nm	.180
	%R 630 nm / %R 580 nm	.002
	K/S 630 nm / K/S 580 nm	.006
	%R 582 nm / %R 525 nm	.002
	K/S 632 nm / K/S 614 nm	.001
	K/S 474 nm / K/S 597 nm	.001
<u>Semimembranosus</u> ^e :	%R 630 nm - %R 580 nm	.217
	K/S 632 nm - K/S 614 nm	.011
	%R 632 nm - %R 614 nm	.005
	%R 507 nm / %R 572 nm	.004
	K/S 582 nm / K/S 525 nm	.003

^aAll contributions are significant at .05 level.

^{b-e}_{n=608}, color evaluated at days 0, 1, 3 and 5 of display under 1076 lm/m² for 24 hrs/day at 2°C.

SUMMARY

Unadjusted reflectance parameters at 630 nm/580 nm, 507 nm/572 nm, 572 nm/525 nm, 630 nm - 580 nm and 507 nm/572 nm showed high pooled within cell correlation coefficients to visual color score. Adjustment of reflectance parameters to K/S values failed to improve correlation coefficients.

Multiple regression analysis showed that %R 630 nm - %R 580 nm made the greatest contribution to R^2 of prediction equations in three of four beef muscles.

The use of %R 630 nm - %R 580 nm as a predictor of visual color score is recommended.

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CHAPTER 6

GENERAL SUMMARY

Longissimus (L), semitendinosus (ST), biceps femoris (BF) and semimembranosus (SM) muscles from 32 steer carcasses representing four nutritional regimes (grass-, short-, long- and silage-fed) and fabricated after a 48 hour carcass chill or after vacuum aging for 21 days before cutting were used to evaluate nutritional regime and vacuum packaging effects on beef muscle color. Color reflectance and visual score were determined at days 0, 1, 3 and 5 of display and percent metmyoglobin (Mb^+) was calculated. Multiple regression analysis showed $\%R$ 630 nm - $\%R$ 580 nm to be the greatest objective contributor to R^2 in three of the four muscles; therefore, it was analyzed as the best indicator of visual score.

No difference in visual score existed due to nutritional regime, but percent Mb^+ increased and $\%R$ 630 nm - $\%R$ 580 nm values decreased as length of feeding increased.

Vacuum packaging for 21 days before cutting improved visual ~~score and increased~~ $\%R$ 630 nm - $\%R$ 580 nm values of L, BF and SM steaks. Conventionally packaged ST steaks had brighter visual scores, lower Mb^+ levels and higher $\%R$ 630 nm - $\%R$ 580 nm values.

Visual score became darker, percent Mb^+ increased and $\%R$ 630 nm - $\%R$ 580 nm values decreased as display time progressed.

L, BF and SM muscles had brighter visual scores and higher $\%R$ 630 nm - $\%R$ 580 nm values through day 3, but at day 5 conventionally packaged BF and SM steaks had darker visual scores.

Vacuum packaged steaks from each muscle had higher Mb⁺ levels at day 5. For ST muscle that was true at each day.

L steaks from all nutritional regimes had acceptable visual scores through day 3; those of ST steaks were acceptable through day 5. BF and SM steaks from all nutritional regimes had acceptable visual scores through day 1. By day 3 visual score was generally unacceptable for BF and SM steaks from all nutritional regimes. Only SM steaks from the silage-fed regime were clearly acceptable at day 3.

Through day 3 of display percent Mb⁺ was lowest in ST, BF and SM muscles from the grass-fed regime; however, from day 3 to day 5 L, BF and SM steaks from grass-fed cattle showed a large increase in percent Mb⁺.

All four muscles from the long- and silage-fed regimes and L steaks from each nutritional regime showed brighter visual scores as a result of vacuum packaging.

Vacuum packaging did not change Mb⁺ proportions in L muscle from any nutritional regime, but increased Mb⁺ percentage in ST muscle. Vacuum packaging generally increased $\%R$ 630 nm - $\%R$ 580 nm values for L, BF and SM muscles from all nutritional regimes.

Vacuum packaged L muscle from grass-fed cattle had increased Mb⁺ levels at day 5 compared with the same muscle packaged conventionally. $\%R$ 630 nm - $\%R$ 580 nm values were higher for vacuum packaged L muscle from all nutritional regimes through day 1.

Vacuum packaging did not change percent Mb⁺ of ST steaks from grass-fed cattle, but increased Mb⁺ in ST muscle from the

other regimes. $\%R$ 630 nm - $\%R$ 580 nm values showed an increase initially for ST muscle from all nutritional regimes. At days 3 and 5, these values were lower for vacuum packaged ST as opposed to conventionally packaged ST steaks.

Packaging method did not affect percent Mb^+ of BF muscle from any nutritional regime at day 0, but vacuum packaged BF muscle showed higher Mb^+ levels in three nutritional regimes at day 5. $\%R$ 630 nm - $\%R$ 580 nm values were higher for vacuum packaged BF steaks from all nutritional regimes through day 1.

Vacuum packaged SM muscle from all nutritional regimes had less Mb^+ at day 0 than conventionally packaged SM muscle, but was generally higher on day 5. Vacuum packaged SM muscle from the short-fed regime maintained the lower Mb^+ levels through the display compared with the same muscle conventionally packaged. $\%R$ 630 nm - $\%R$ 580 nm values of SM muscle from all nutritional regimes were increased by vacuum packaging through day 1. At days 3 and 5 these values were lower for vacuum packaged steaks from grass- and long-fed cattle.

Unadjusted reflectance parameters at 630 nm/580 nm, 507 nm/572 nm, 572 nm/525 nm, 630 nm - 580 nm and K/S adjustment of 572 nm/525 nm and 507 nm/572 nm showed high ($r = -.84, -.84, .83, -.82, -.85$ and $.83$, respectively) pooled within cell correlation coefficients to visual color score. Adjustment of reflectance parameters to K/S values failed to improve correlation coefficients.

BEEF MUSCLE COLOR AS AFFECTED BY NUTRITIONAL REGIME AND VACUUM PACKAGING

by

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Visual score became darker, percent Mb^+ increased and $\%R\ 630\text{ nm} - \%R\ 580\text{ nm}$ values decreased as display time progressed.

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Vacuum packaged L muscle from grass-fed cattle had increased Mb⁺ levels at day 5 compared with the same muscle packaged conventionally. %R 630 nm - %R 580 nm values were higher for vacuum packaged L muscle from all nutritional regimes through day 1.

Vacuum packaging did not change percent Mb⁺ of ST steaks from grass-fed cattle, but increased Mb⁺ in ST muscle from the other regimes. %R 630 nm - %R 580 nm values showed an increase initially for ST muscle from all nutritional regimes. At days

3 and 5, these values were lower for vacuum packaged ST as opposed to conventionally packaged ST steaks.

Packaging method did not affect percent Mb⁺ of BF muscle from any nutritional regime at day 0, but vacuum packaged BF muscle showed higher Mb⁺ levels in three nutritional regimes at day 5. %R 630 nm - %R 580 nm values were higher for vacuum packaged BF steaks from all nutritional regimes through day 1.

Vacuum packaged SM muscle from all nutritional regimes had less Mb⁺ at day 0 than conventionally packaged SM muscle, but was generally higher on day 5. Vacuum packaged SM muscle from the short-fed regime maintained the lower Mb⁺ levels through the display compared with the same muscle conventionally packaged. %R 630 nm - %R 580 nm values of SM muscle from all nutritional regimes were increased by vacuum packaging through day 1. At days 3 and 5 these values were lower for vacuum packaged steaks from grass- and long-fed cattle.

Unadjusted reflectance parameters at 630 nm/580 nm, 507 nm/572 nm showed high ($r = -.84, -.84, .83, -.82, -.85$ and $.83$, respectively) pooled within cell correlation coefficients to visual color score. Adjustment of reflectance parameters to K/S values failed to improve correlation coefficients.