Effects of different packaging atmospheres and injection-enhancement on beef tenderness, sensory attributes, desmin degradation, and display color1,2

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ABSTRACT: The objectives were to determine the effects of packaging atmosphere and injection-enhancement on tenderness, sensory traits, desmin degradation, and display color of different beef muscles. Longissimus lumborum (LL; n = 12 pairs), semitendinosus (ST; n = 12 pairs), and triceps brachii (TB; n = 24 pairs; 12 from the same carcasses as the LL and ST and 12 additional pairs) were obtained from the same USDA Select, A-maturity carcasses. On d 7 postmortem, each muscle from one side of the carcass was injection-enhanced, and each muscle from the other side was nonenhanced. Steaks 2.54-cm thick were cut from the muscles and packaged in vacuum packaging (VP), ultra-low oxygen with CO (ULO2CO; 0.4% CO/35% CO2/69.6% N2) modified atmosphere packaging (MAP), or high-oxygen MAP (HiO2; 80% O2/20% CO2) and assigned to 14-d tenderness or display followed by 18- or 28-d tenderness measurement. Steaks packaged in HiO2 MAP were in dark storage (2°C) for 4 d and all other steaks for 14 d. Steaks for Warner-Bratzler shear force, sensory panel (n = 8 trained panelists), and desmin degradation were cooked to 70°C. Steaks were displayed under fluorescent lighting (2,153 lx, 3,000 K) for 7 d. Trained color panelists (n = 10) assigned display color scores. Enhanced steaks had lower (P < 0.05) Warner-Bratzler shear force values than nonenhanced steaks. Sensory panelists found that nonenhanced steaks packaged in ULO2CO MAP or VP were more tender (P < 0.05), had more (P < 0.05) beef flavor, and had less (P < 0.05) off-flavors than steaks packaged in HiO2 MAP. The LL and TB were more tender (P < 0.05) according to myofibrillar tenderness than the ST. Nonenhanced steaks were less (P < 0.05) juicy than enhanced steaks. The most common off-flavors associated with steaks packaged in HiO2 MAP were oxidative or rancid. Enhanced steaks had more (P < 0.05) off-flavors than nonenhanced steaks, with typical descriptors of salty, metallic, or chemical, in addition to an undesirable mushy texture. Desmin degradation increased (P < 0.05) from 7 to 14 d postmortem and differed among muscles but was not affected (P > 0.05) by packaging or enhancement. Enhanced steaks were darker (P < 0.05) initially than nonenhanced steaks. Steaks packaged in HiO2 MAP discolored faster (P < 0.05) and to a greater extent (P < 0.05) than steaks packaged in VP or ULO2CO MAP. Nonenhanced muscles packaged in VP and ULO2CO MAP had more stable display color and very desirable tenderness and flavor compared with those packaged in HiO2 MAP.

Key words: beef, display color, injection-enhancement, modified atmosphere packaging, postmortem proteolysis, sensory attribute

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INTRODUCTION

Case-ready meat provides many benefits, including quality and safety. Packaging meat in high-oxygen (HiO2) modified atmosphere packaging (MAP) results in a desirable bright red display color (Behrends et al., 2003; Seyfert et al., 2005) but may have increased off-flavors and decreased tenderness. Steaks aged and packaged in HiO2 MAP had more off-flavor, including warmed-over flavor, and were less tender and juicy than steaks aged in vacuum packaging (VP; Tørngren, 2003; Sorheim et al., 2004; Clausen, 2004; Madsen and

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Intra-muscular injection-enhancement improves tenderness and juiciness while decreasing variation in these traits, and it is used in conjunction with MAP. Enhancement of beef resulted in more tender and juicy steaks than none-enhanced steaks (Vote et al., 2000; Lawrence et al., 2003a; Wicklund et al., 2005; Hoffman, 2006). Several researchers have found an increase in beef flavor associated with enhanced steaks (Vote et al., 2000; Knock et al., 2006a), whereas others have reported a decrease or no change in beef flavor (Robbins et al., 2003; Molina et al., 2005; Hoffman, 2006; Stetzer et al., 2007). Off-flavors have been associated with enhanced beef, such as salty and oxidative (Seyfert et al., 2005; Knock et al., 2006a). The objectives of our study were to determine the effects of packaging atmosphere and injection-enhancement on beef longissimus lumborum (LL), semitendinosus (ST), and triceps brachii (TB) tenderness, sensory traits, desmin degradation, and display color.

MATERIALS AND METHODS

Animal Care and Use Committee approval was not obtained for this study because the samples were obtained from a federally inspected slaughter facility (Tyson Fresh Meats).

Samples, Packaging, and Injection-Enhancement

Paired LL (n = 12 pairs), ST (n = 12 pairs), and TB (n = 24 pairs; 12 from same carcass as the LL and ST plus 12 additional pairs) from the same USDA Select, A-maturity carcasses were obtained at a commercial abattoir at 2 d postmortem. One steak from each muscle was vacuum packaged and used for initial Warner-Bratzler shear force (WBSF) on d 7 postmortem. On d 7 postmortem, each muscle from one side of the carcass was enhanced (Schröder Injector 50, Wolf-Tec Inc., Kingston, NY) with beef broth, potassium lactate, sodium phosphate, salt, and natural flavoring (rosemary) solution (proprietary formulation), with a targeted 10% enhancement. Each muscle from the opposite side was nonenhanced. Steaks 2.54-cm thick were cut from the muscles and packaged in treatments of 1) vacuum packaging (VP; 62.2 cm Hg vac; Multivac C500; Multivac Inc., Kansas City, MO); 2) ultra-low oxygen modified atmosphere packaging (MAP) with CO (ULO2CO; 64.6% N2, 35% CO2, 0.4% CO); or 3) high-oxygen (HiO2; 80% O2, 20% CO2; AirGas certified gas, MidSouth Inc., Tulsa, OK) MAP and assigned to either 14-d tenderness measurement or to display followed by 18- or 28-d tenderness measurement. Steaks packaged in MAP (Ross Jr. S-3180, Ross, Midland, VA) were packaged in 4.32-cm-deep rigid plastic trays (CS977, Cryovac Sealed Air Corp., Duncan, SC) and covered with oxygen-barrier film (Lid 550; 1.0 mils; less than 20.0 oxygen transmission mL/24 h/m2 at 4.4°C with 100% relative humidity (RH); and moisture vapor transmission less than 0.1 g/24 h/645.2 cm2 at 4.4°C and 100% RH; Cryovac Sealed Air Corp., Duncan, SC). High-oxygen MAP were held in dark storage (2°C) for 4 d and then put into simulated retail display and removed on d 18 postmortem. All packaging treatments without O2 were held in dark storage (2°C) for 14 d and then put into simulated retail display and removed on d 28 postmortem. Steaks packaged in all packaging treatments used for 14 d postmortem WBSF were held for 7 d in the dark after packaging and then cooked for WBSF measurement. Dark storage times were developed to mimic what would happen in industry. An activated oxygen scavenger (ActiveTech; Pactiv, Chicago, IL) was included in each of the ULO2 packages to eliminate any residual O2.

pH

The pH of steaks was measured on d 14 postmortem by inserting the tip of a pH probe (MPI pH probe, glass electrode, Meat Probes Inc., Topeka, KS) into 1 location of each muscle.

Cooking of Steaks

Steaks for WBSF or internal cooked color were cooked in a forced-air convection oven (Blodgett, model DFG-102 CH3, G. S. Blodgett Co., Burlington, VT) set at 163°C. Steaks were turned at an internal temperature of 40°C and cooked to an internal temperature of 70°C, as monitored with copper-constantan thermocouples in the approximate geometric center of each steak.

Warner-Bratzler Shear Force

On d 7 postmortem, d 14 postmortem, and at the end of each display (d 18 or 28 postmortem), steaks from all packaging treatments were cooked, cooled to room temperature, and stored at 2°C overnight. Eight 1.27-cm cores were removed from each steak parallel to the muscle fibers using a 1.27-cm corer (G-R Manufacturing Co., Manhattan, KS) attached to an electric drill (Craftsman 3/8” Electric Drill, Sears, Hoffman Estates, IL). Cores were then sheared once perpendicular to the muscle fibers using a Warner-Bratzler V-shaped blunt blade (G-R Manufacturing Co.) attached to a 50-kg load cell of an Instron Universal Testing Machine (model 4201, Instron Corp., Canton, MA) with a crosshead speed of 250 mm/min. Peak shear force was recorded in kilograms, and values from the 8 cores were averaged.
**Sensory Analysis**

One steak from each muscle and each packaging atmosphere was removed from MAP on d 18, vacuum packaged (62.2 cm Hg vac; Multivac C500, Multivac Inc., Kansas City, MO) and frozen at −20°C for later sensory analysis. Steaks already packaged in VP also were frozen. Panelists (n = 8) were trained according to AMSA guidelines (1995) for evaluation of steaks. Steaks were thawed overnight (2°C), cooked to 70°C, sliced into 2.54-cm × 1.27-cm × 1.27-cm samples, and served warm to panelists. Samples were kept warm in blue enamel double boiler pans with warm water in the bottom portion of the pan. Panelists evaluated samples in duplicate for myofibrillar tenderness, juiciness, beef flavor intensity, amount of connective tissue, overall tenderness, and off-flavor using an 8-point scale. The scale used for myofibrillar and overall tenderness was 1 = extremely tough, 2 = very tough, 3 = moderately tough, 4 = slightly tough, 5 = slightly tender, 6 = moderately tender, 7 = very tender, and 8 = extremely tender. For juiciness, the scale was 1 = extremely dry, 2 = very dry, 3 = moderately dry, 4 = slightly dry, 5 = slightly juicy, 6 = moderately juicy, 7 = very juicy, and 8 = extremely juicy. The scale used for beef flavor was 1 = very bland, 2 = very bland, 3 = moderately bland, 4 = slightly bland, 5 = slightly intense, 6 = moderately intense, 7 = very intense, and 8 = extremely intense. The scale used for connective tissue and off-flavor intensity was 1 = abundant, 2 = moderately abundant, 3 = slightly abundant, 4 = moderate, 5 = slight, 6 = traces, 7 = practically none, and 8 = none. Scores were given to the nearest half-point increment.

**Cooking Loss**

Steaks used for sensory analysis were weighed before cooking (initial weight), allowed to cool for approximately 5 min at room temperature, and weighed again (final weight). Cooking loss was calculated by [(initial weight − final weight)/initial weight]·100.

**Immunoblotting**

Desmin degradation was used as a measure of postmortem proteolysis. Extraction, electrophoresis, Western blotting, and quantification of desmin was measured on 7 and 14 d postmortem samples at the USDA, ARS, Roman L. Hruska US Meat Animal Research Center, Clay Center, NE, according to procedures outlined by Wheeler and Koohmaraie (1999) and Wheeler et al. (2002) with the following modifications. Samples were loaded at 15 g of protein per lane. Gels were transferred to membranes for 1 h. The primary antibody [monoclonal anti-desmin (clone D3; developed by D. A. Fischman, Cornell University Medical College, New York, NY, and obtained from the Developmental Studies Hybridomal Bank)] was diluted as follows: 1:300 for ST and TB or 1:100 for LL. Bound primary antibodies were labeled with Immunopure goat anti-mouse IgG horseradish peroxidase conjugated secondary antibodies diluted 1:10,000 (Pierce, Rockford, IL). Detection of antibody binding was done by incubating the membranes for 5 min using the SuperSignal West Dura Extended Duration Substrate (Pierce) and exposing the membrane for 5 min with a ChemiImager 4000 digital imaging analysis system (Alpha Innotech, San Leandro, CA).

**Display Case**

Packages were displayed (Unit model DMF8, Tyler Refrigeration Corp., Niles, MI) under continuous fluorescent lighting (2,153 lx, 3,000 K and CRI = 85, Bulb model 32T8/ADV830/Alto, Philips, Bloomfield, NJ) for 7 d at 2°C. Packages were rotated twice daily to maintain a random sample placement.

**Color Measurements**

Trained visual color panelists (n = 10) evaluated initial color on d 0 of display and display color and surface discoloration on d 0 to 7 of display once each day. Initial color was determined using the following scale: 1 = purplish red or reddish tan, 2 = bleached, pale red, 3 = slightly cherry red, 4 = moderately light cherry red, 5 = cherry red, 6 = slightly dark red, 7 = moderately dark red, 8 = dark red, 9 = very dark red. The color scale used by panelists for steaks packaged in MAP was 1 = very bright red or very bright pinkish red, 2 = bright red or bright pinkish red, 3 = dull red or dull pinkish red, 4 = slightly dark red or slightly dark pinkish red, 5 = reddish tan or pinkish tan, 6 = moderately dark red or reddish tan or moderately dark pinkish red or pinkish tan, 7 = tannish red or tannish pink, 8 = tan to brown. Steaks packaged in VP were evaluated with the following scale: 1 = very bright purplish pink or very bright purplish pink, 2 = bright purplish pink or bright purplish pink, 3 = dull purplish red or dull purplish pink, 4 = slightly dark purplish red or slightly dark purplish pink, 5 = purplish tan or pinkish tan, 6 = moderately dark purplish red or moderately dark purplish pink, 7 = tannish purple red or tannish purple pink, and 8 = tan to brown. For all steaks, discoloration was considered as a percentage of surface metmyoglobin, and the following scale was used to evaluate this: 1 = none (0%), 2 = slight discoloration (1 to 19%), 3 = small discoloration (20 to 39%), 4 = modest discoloration (40 to 59%), 5 = moderate discoloration (60 to 79%), 6 = extensive discoloration (80 to 99%), and 7 = total discoloration (100%). Color scales were used to half-point increments, and discoloration was scored to whole-point increments.

**Statistical Analysis**

The experimental design was a split plot design, with block being enhancement for carcass side. The subplot
Table 1. pH muscle × enhancement treatment means and SE\textsuperscript{1} for steaks

<table>
<thead>
<tr>
<th>Muscle</th>
<th>Nonenhanced</th>
<th>Enhanced</th>
</tr>
</thead>
<tbody>
<tr>
<td>Longissimus lumborum</td>
<td>5.5\textsuperscript{a}</td>
<td>5.8\textsuperscript{b}</td>
</tr>
<tr>
<td>Semitendinosus</td>
<td>5.6\textsuperscript{a}</td>
<td>5.8\textsuperscript{b}</td>
</tr>
<tr>
<td>Triceps brachii</td>
<td>5.6\textsuperscript{a}</td>
<td>5.8\textsuperscript{b}</td>
</tr>
</tbody>
</table>

\textsuperscript{a,b}Means within a row with different superscript letters differ (P < 0.05).
\textsuperscript{1}SE = 0.02.

RESULTS AND DISCUSSION

Injection-enhancement targeted pump levels were 10%. After approximately 30 min post initial injection and just before fabrication, pump level was 10.7% for the LL, 8.2% for the ST, and 13.0% for the TB, calculated by [(pumped weight – prepumped weight)/prepumped weight]. Differences may have occurred due to inherent water holding capacity, muscle fiber type, collagen content, muscle fiber orientation, or a combination of these relative to the injection needles.

pH

There was a muscle × enhancement treatment interaction (P < 0.05) for pH (Table 1). Enhanced steaks had a higher (P < 0.05) pH than nonenhanced steaks regardless of packaging treatment. The lactate, phosphate, or both in the enhancement solution caused an increase in muscle pH. The increased pH in enhanced steaks may be responsible for decreased cook loss and increased juiciness found in enhanced steaks compared with nonenhanced steaks. Water-holding capacity of meat increases the farther the pH is from the isoelectric point (5.1) of meat.

Warner-Bratzler Shear Force

Tenderness, according to WBSF, resulted in a packaging treatment × day interaction (P = 0.002; Figure 1). Steaks packaged in HiO\textsubscript{2} MAP were less tender at the end of their display (d 18 postmortem) than steaks packaged in VP or ULO\textsubscript{2}CO MAP. This is due to storage differences in tenderness due to packaging environment. Results of other studies show more distinct differences in tenderness due to packaging environment. Steaks packaged in HiO\textsubscript{2} MAP have been shown to be less tender after 7 to 14 d than steaks packaged in VP or ULO\textsubscript{2}, with or without CO MAP, by instrumental or trained sensory panelists, or both (Tørngren, 2003; Sørheim et al., 2004; Clausen, 2004; and Madsen and Clausen, 2006). These studies used steaks from heif-
ers, cows, or bulls and were most likely fed different
types of diets than the traditional grain-fed diets most
often found in US cattle slaughtered, such as the A-ma-
turity, USDA Select carcasses used in our study. The
differences in cattle gender, age, and feeding regimens
between our study and others might have played a role
in the results we saw compared with the other studies.
Claussen (2004) believes that the detrimental effects of
O2 on tenderness may be caused by protein oxidation.
Rowe et al. (2004) found that oxidation of beef muscle
proteins early postmortem inactivated μ-calpain and
decreased myofibrillar proteolysis, thus potentially
limiting tenderization.

Although there was a trend (P = 0.057) for a muscle ×
enhancement × packaging treatment × day interaction
for WBSF, there was a muscle × enhancement treatment × day interaction (P < 0.05) in which steaks from
enhanced muscles were more (P < 0.05) tender than
nonenhanced steaks (Figure 2). Tenderness increased
with time postmortem (d 14 to 18/28) in enhanced LL
and TB steaks but not in ST steaks. Nonenhanced
steaks were similar in tenderness on d 7 and 14 post-
mortem but were more tender on d 18/28 postmortem
for all muscles. Enhanced LL steaks were more (P <
0.05) tender than nonenhanced steaks on d 7 postmor-
tem, which was d 0 of packaging. This indicates that
injection enhancement has an immediate effect on ten-
derness. Injection-enhancement may increase tenderness through a dilution effect or through physically
altering the muscle structure with the injection nee-
dling process; however, the exact method of action is
currently unknown. Due to time constraints and cook-
ing capabilities, we were unable to determine WBSF on
d 7 postmortem (d of fabrication and d 0 of packaging)
for enhanced ST and TB muscles.

Other studies have found that injection-enhance-
ment of beef muscles results in decreased shear force
values compared with nonenhanced steaks. Wicklund
et al. (2005) reported that enhanced beef strip loin
steaks had lower WBSF values than nonenhanced
steaks when loins were aged before enhancement and
when loins were enhanced before aging. Vote et al. (2000) and Robbins et al. (2003) reported lower shear
force in enhanced beef strip loin steaks than in nonen-
hanced steaks. Lawrence et al. (2004) evaluated injec-
tion of beef LM with a phosphate and salt solution or
a calcium lactate plus beef broth or carrageenan with
rosemary extract solution and found no differences in
WBSF among all treatments. Knock et al. (2006a) re-
ported that steaks enhanced with sodium acetate had
lower shear force than control steaks or steaks en-
hanced with potassium lactate, but the mechanism for
this difference is unknown.

**Sensory Analysis**

There was an enhancement treatment × packaging
interaction for myofibrillar tenderness (P <
0.05), beef flavor and off-flavor (P < 0.01), and over-
all tenderness (P < 0.05) (Figures 3 and 4). According
to sensory panelists, nonenhanced steaks packaged in
HiO2 MAP were less tender, had less beef flavor, and
had more \((P < 0.05)\) off-flavors than those packaged in ULO\(_2\)CO MAP and VP. The LL (5.9 ± 0.1) and TB (6.0 ± 0.1) were more \((P < 0.05)\) tender according to myofibrillar tenderness than the ST (5.1 ± 0.1). Enhanced steaks packaged in VP had more \((P < 0.05)\) beef flavor than enhanced steaks packaged in HiO\(_2\) MAP.

**Figure 2.** Muscle × enhancement × day [day postmortem = 7, 14, or 18/28 (d 18 postmortem for the HiO\(_2\) treatment and d 28 postmortem for the ULO\(_2\)CO and VP treatments)] Warner-Bratzler shear force means and SE for longissimus lumborum (LL), semitendinosus (ST), and triceps brachii (TB). \(^{a–h}\)Means with different letters differ \((P < 0.05)\).

**Figure 3.** Enhancement × packaging treatment (HiO\(_2\) = 80% O\(_2\), 20% CO\(_2\); ULO\(_2\)CO = 0.4% CO/35% CO\(_2)/64.6% N\(_2\); VP = vacuum packaging) myofibrillar tenderness and overall tenderness (1 = extremely tough, 4 = slightly tough, 6 = moderately tender, 8 = extremely tender) means and SE for longissimus lumborum, semitendinosus, and triceps brachii steaks. \(^{a–c}\)Means with different letters within sensory traits differ \((P < 0.05)\).
The main effect ($P < 0.01$) for juiciness revealed that enhanced steaks ($5.7 \pm 0.1$) were juicier ($P < 0.05$) than nonenhanced steaks ($5.1 \pm 0.1$). These results agree with cooking loss data and were expected because enhanced steaks have additional moisture added to them at the time of injection. The muscle main effect ($P < 0.01$) for juiciness resulted in steaks from LL ($5.5 \pm 0.2$) and TB ($5.9 \pm 0.1$) muscles being juicier ($P < 0.05$) than steaks from ST ($5.0 \pm 0.2$) muscles. There was a packaging treatment main effect ($P < 0.01$) for juiciness. Steaks packaged in HiO$_2$ MAP ($5.3 \pm 0.1$) were less juicy ($P < 0.05$) than steaks packaged in ULO$_2$CO MAP ($5.6 \pm 0.1$), whereas steaks packaged in VP ($5.4 \pm 0.1$) were intermediate and not different in juiciness from steaks packaged in HiO$_2$ and ULO$_2$CO MAP.

There was a main effect ($P < 0.01$) for connective tissue for the enhancement treatment and muscle. Enhanced ($6.6 \pm 0.1$) steaks had less ($P < 0.05$) perceptible connective tissue than nonenhanced ($6.1 \pm 0.1$) steaks. The ST ($5.9 \pm 0.1$) had more ($P < 0.05$) perceptible connective tissue than the TB ($6.4 \pm 0.1$), which had more ($P < 0.05$) perceptible connective tissue than the LL ($6.7 \pm 0.1$). There was also a main effect ($P < 0.01$) for packaging treatment for connective tissue in which steaks packaged in HiO$_2$ MAP ($6.2 \pm 0.1$) had more ($P < 0.05$) perceptible connective tissue than steaks packaged in ULO$_2$CO MAP ($6.4 \pm 0.1$) and VP ($6.4 \pm 0.1$).

The most common off-flavors associated with steaks packaged in HiO$_2$ MAP were oxidative or rancid. Enhanced steaks had more ($P < 0.05$) off-flavors than nonenhanced steaks, with typical descriptors of salty and metallic or chemical. There were also comments on many of the enhanced steaks that indicated an undesirable mushy texture. There was a muscle $\times$ enhancement treatment interaction for beef flavor ($P < 0.05$) and off-flavor ($P < 0.05$; Figure 5). Enhanced TB steaks had more ($P < 0.05$) beef flavor than enhanced ST steaks. Oxidative off-flavors associated with steaks packaged in HiO$_2$ MAP were expected because the O$_2$ present in the package atmosphere allows for more rapid and a greater extent of oxidation of proteins and lipids found in meat. Eliminating O$_2$ from the package environment, as accomplished with VP or ULO$_2$CO MAP, drastically decreases the rate and extent of oxidation, thus resulting in fewer off-flavors and increased beef flavor taste perception.

Our results for beef-flavor intensity are similar to those of Carmack et al. (1995) who reported that the TB was equal in beef-flavor intensity to the LL and the TB had more flavor than the ST. In our study, steaks from the ST had less beef-flavor than steaks from the TB and the LL, but Carmack et al. (1995) found the LL to have beef-flavor intensity similar to that of the ST. In our study, the TB was equal in tenderness to the
LL, and both were more tender than the ST according to trained sensory panelists. Carmack et al. (1995) reported that the LL was more tender than the TB, which was equal in tenderness to the ST. Our results indicate that the TB and LL also were juicier than the ST. Carmack et al. (1995) reported that the LL was juicier than the ST, with the TB being intermediate in juiciness.

Jackson et al. (1992) evaluated the volatile compounds from the headspace of beef strip loins vacuum packaged or in 100% CO₂ MAP, ULO₂ MAP, or HiO₂ MAP. They reported that steaks packaged in HiO₂ MAP developed strong off-odors and had methyl thirane, ethyl acetate, benzene, and 1-heptene in the packages after 7 and 14 d of storage, but steaks in the vacuum packaging or other MAP atmospheres did not.

Seyfert et al. (2005) reported that injection-enhanced beef quadriceps packaged in HiO₂ MAP were less tender and had more off-flavors than those in ULO₂ MAP. They also reported that increasing injection percentage from 6 to 10% in beef round muscles decreased oxidation but increased nontypical beef flavors. Hoffman (2006) found that enhancement of cow LL and ST muscles resulted in more tender and juicier steaks that were saltier and had less overall beef flavor than steaks that were nonenhanced. Wicklund et al. (2005) found similar results in beef loin steaks.

Knock et al. (2006a) found that adding potassium lactate to injection-enhanced beef packaged in HiO₂ MAP limited rancid flavor development while increasing brown-roasted and beef flavors. They also found that increasing the salt content in the injection-enhancement solution increased salty and rancid flavors. In addition, oxidized, stale, and rancid flavors increased as time in HiO₂ MAP increased. Vote et al. (2000) reported increased tenderness and juiciness for injected steaks compared with control, noninjected steaks. They also found a trend for increased cooked beef flavor, but when sodium tripolyphosphate was injected alone, soapy and sour off-flavors were detected. They also cooked steaks to 66 or 77°C and showed that the improvements in tenderness and juiciness compared with control steaks were even greater at 77°C. Thus, injection-enhancement may be beneficial when consumers overcook steaks, and it helps processors deliver more consistent products to consumers.

Lawrence et al. (2003a) evaluated injection marination of beef LL muscles with calcium ascorbate, calcium chloride, or calcium lactate and reported that calcium lactate increased beef flavor and had no effect on off-flavors compared with control samples. Lawrence et al. (2003b) injected beef LL and ST muscles with calcium lactate followed by a phosphate and salt injection solution. They found no differences in tenderness or sensory traits for the ST muscles but found increased tenderness for injected LL muscles compared with control muscles. They reported that LL steaks from muscles injected with phosphate and salt in addition to calcium lactate had less beef flavor and fewer complaints of samples being too salty than did steaks from control muscles or muscles injected only with calcium lactate. Trained sensory panelists found that steaks enhanced

Figure 5. Muscle × enhancement beef flavor (1 = extremely bland, 4 = slightly bland, 6 = moderately intense, 8 = abundant) and off-flavor (1 = abundant, 5 = slight, 6 = traces, 7 = practically none, 8 = none) means for longissimus lumborum (LL), semitendinosus (ST), and triceps brachii (TB) steaks. * a–dMeans with different letters within sensory traits differ (P < 0.05).
with calcium lactate plus rosemary were less tender than steaks enhanced with phosphate and salt plus rosemary, but off-flavors of metallic and salty were increased with the phosphate and salt plus rosemary treatment compared with the calcium lactate plus rosemary treatment (Lawrence et al., 2004). Stetzer et al. (2007) compared beef loin steaks injection-enhanced with phosphate, salt, and natural flavorings packaged in HiO₂ MAP and CO MAP. These researchers reported that consumer sensory panelists did not find differences due to packaging environment in beef flavor, off-flavor, or overall acceptability.

**Cooking Loss**

There were no main effect interactions ($P > 0.01$) for cooking loss, but there were main effects ($P < 0.01$) for muscle, enhancement treatment, and packaging treatment ($P < 0.01$). The LL (19.7% ± 0.5) had the least ($P < 0.05$) cooking loss; the TB (22.7% ± 0.5) had intermediate cooking loss; and the ST had the most ($P < 0.05$) cooking loss (26.9% ± 0.5). Enhanced steaks (19.9% ± 0.4) had less ($P < 0.05$) cooking loss than nonenhanced steaks (26.3% ± 0.4). Steaks packaged in HiO₂ MAP (22.1% ± 0.5) had the least ($P < 0.05$) cooking loss, and steaks packaged in ULO₂CO MAP (24.1% ± 0.5) had the most ($P < 0.05$) cooking loss. Cooking loss of vacuum packaged steaks (23.2% ± 0.5) was intermediate and not different ($P > 0.05$) than cooking loss of steaks packaged in HiO₂ or ULO₂CO MAP.

Wicklund et al. (2005) reported that enhanced beef strip loin steaks had less cooking loss on d 7 of storage but not on d 14, 21, or 28 than nonenhanced steaks. Molina et al. (2005) found that enhancement by marination, needle-pumping, and vacuum-tumbling resulted in decreased cooking loss compared with nonenhanced steaks from several different muscles from the beef chuck. Stetzer et al. (2007) compared beef loin steaks injection-enhanced with phosphate, salt, and natural flavorings packaged in HiO₂ MAP and CO MAP. These authors reported that there were no differences in purge loss or cooking loss when both packaging methods were stored for 14 d, but steaks in CO MAP had more cooking loss when stored for 28 d compared with steaks in both packaging types at 14 d of storage.

**Desmin Degradation**

There was a muscle × enhancement interaction ($P < 0.001$) for desmin degradation (Figure 6). Nonenhanced and enhanced steaks were not different ($P > 0.05$) in the amount of desmin degradation. Longissimus lumborum desmin degradation increased ($P < 0.05$) from d 7 to 14, regardless of enhancement treatment. Longissimus lumborum steaks had more ($P < 0.05$) degradation of desmin at d 14 than the ST or TB, regardless of enhancement treatment. Different muscles varied in rates and extent of postmortem proteolysis, of which desmin degradation is an index. Postmortem proteolysis is a key determinant of tenderness in some muscles (Rhee et al., 2004; Koohmaraie and Geesink, 2006). Desmin degradation was not affected ($P > 0.05$) by type of packaging (data not presented) but was affected ($P < 0.05$) by time postmortem. There was a day postmortem main effect ($P < 0.001$) for desmin degradation, with d 14 postmortem (36.09% ± 2.9) having more ($P < 0.05$).
Increased desmin degradation with increased day postmortem was expected because aging increases postmortem proteolysis and the breakdown of desmin. We did not expect to find differences in desmin degradation for enhancement treatments, and our results agree with this. We hypothesized that packaging treatment may alter desmin degradation through protein oxidation associated with HiO2 MAP, by slowing down or hindering postmortem proteolysis as indicated by Rowe et al. (2004). However, our results did not indicate this. Protein oxidation may be associated with muscle in the early stages after harvest. Muscles used in our study were aged for 7 d in vacuum before exposing steaks to different packaging treatments. To our knowledge, our study is the first to look at desmin degradation for enhanced and nonenhanced steaks from different muscles in different packaging atmospheres.

There was a more dramatic reduction ($P < 0.05$) in shear force between nonenhanced and enhanced steaks than was observed in the numerical increases ($P > 0.05$) of desmin degradation for enhanced steaks compared with nonenhanced steaks. We believe the large decrease in WBSF between nonenhanced and enhanced steaks, observed as early as d 0 of packaging in LL steaks, may be caused by the physical manipulation of the muscle through the needling process used with injection-enhancement or because of dilution of the muscle with enhancement solution. Relative to nonenhanced steaks, the numerical increase in desmin degradation in enhanced steaks argues against a role for oxidation through reduced $\mu$-calpain proteolysis (because enhancement solutions would cause much more oxidation than a packaging treatment). Our enhancement solution, potentially, could have affected other factors that would impact proteolysis.
Display Color and Discoloration

There was a muscle × packaging treatment interaction (P < 0.01) for initial color score (data not shown). The initial color for ST steaks was lighter cherry red (P < 0.05) than for LL or TB steaks. Initially, TB steaks packaged in HiO2 MAP were darker red (P < 0.05) than TB steaks packaged in ULO2CO MAP. Enhanced steaks were darker (P < 0.05) initially than nonenhanced steaks. Lactate in the enhancement solution is typically associated with increased color stability but also results in slightly darker muscle color (Kim et al., 2006). Nonenhanced TB steaks were darker (P < 0.05) than nonenhanced LL or ST steaks. Enhanced ST steaks were lighter (P < 0.05) than enhanced LL steaks, which were lighter (P < 0.05) than enhanced TB steaks. Differences in muscle fiber type most likely caused differences in initial color among muscles and within packaging treatments.

There was a muscle × enhancement treatment × packaging treatment × day interaction (P < 0.05) for display color scores (Figures 7, 8, and 9). Steaks became darker (P < 0.05) throughout the 7 d of display, but in general, steaks packaged in VP or ULO2CO MAP remained more stable than steaks packaged in HiO2 MAP. In general, TB steaks were darker than LL and ST steaks. Nonenhanced TB steaks packaged in HiO2 MAP became dramatically darker than those packaged in ULO2CO MAP and VP. Steaks packaged in HiO2 or ULO2CO MAP tended to become darker in color at a faster rate than steaks in VP.

There was a muscle × enhancement treatment × packaging treatment × day interaction (P < 0.001) for discoloration scores (Figure 10). Steaks packaged in HiO2 MAP discolored at a relatively faster rate (P < 0.05) and to a greater extent (P < 0.05) than steaks packaged in VP or ULO2CO MAP. Steaks packaged in VP or ULO2CO MAP had no (P < 0.05) discoloration throughout the 7-d display. Including O2 in the package allows for oxidation of myoglobin (main pigment in meat that gives it color) and thus resulted in a reddish tan color by d 7 of display. Excluding O2 from the package, as with VP or ULO2CO MAP treatments, allows myoglobin to remain in a more stable form longer.
and delays the onset of metmyoglobin (tan/brown) color formed through the oxidation of myoglobin. In general, TB steaks packaged in HiO2 MAP discolored at a faster rate and to a greater extent than LL and ST steaks packaged in HiO2 MAP, regardless of enhancement treatment.

Knock et al. (2006b) injected beef longissimus thoracis muscles with different combinations of potassium lactate, sodium chloride, sodium tripolyphosphate, and sodium acetate and packaged as steaks in HiO2 MAP. They found that steaks from muscles injected with potassium lactate, with or without sodium acetate, had increased color stability but were darker than control steaks. Lawrence et al. (2004) evaluated injection of beef LL muscles with calcium lactate and reported improvements in display color stability when rosemary extract was included in the enhancement solution with calcium lactate than when muscles were injected with phosphate and salt with rosemary.

There have been other reports of beef stored in ULO2CO MAP maintaining red color, whereas steaks packaged in HiO2 MAP discolor more rapidly. Behrends et al. (2003) reported acceptable color stability of steaks packaged in HiO2 MAP through d 5 of display.

Other researchers showed increased times of storage in HiO2 MAP for steaks being red in color; however, some of these steaks were stored in dark storage and not displayed under lights as in our study. The use of 0.4% CO in retail meat packages was approved by USFDA (2004). John et al. (2005) reported that steaks in HiO2 MAP were red in color through 14 d of storage and steaks in ULO2CO MAP were red through 21 d of storage. They also found that the majority of steaks stored in VP remained purple in color through d 21 of storage; however, some of their VP steaks must have had residual oxygen in the package and turned brown in color. In addition, Sørheim et al. (1999) and Hunt et al. (2004) reported bright red color and high a* values of steaks stored in ULO2CO MAP.

In summary, more off-flavors were associated with enhanced steaks than nonenhanced steaks. Enhanced steaks were darker in color, had a higher pH, were juicier, and had less perceptible connective tissue than nonenhanced steaks. Steaks packaged in HiO2 MAP were less tender according to sensory panelists and had more off-flavors than those packaged in either ULO2CO MAP or VP. Sensory panelists found steaks packaged in HiO2 MAP to be less tender than steaks packaged in ULO2CO MAP or VP. Sensory panelists found steaks packaged in HiO2 MAP to be less tender than steaks packaged in ULO2CO MAP or VP.
packaged in VP or ULO2CO MAP on d 18 postmortem, whereas WBSF results from steaks on d 14 postmortem were not different. Packaging treatment did not affect desmin degradation, which is a measure of tenderization during aging. Desmin degradation differed between LL and TB muscles, whereas these 2 muscles were equal in tenderness. Desmin degradation did not differ between control and enhanced muscles, whereas enhanced steaks were much more tender than control steaks. In general, steaks packaged in VP or ULO2CO MAP had more display color stability than steaks packaged in HiO2 MAP. Regardless of enhancement treatment, steaks packaged in VP or ULO2CO MAP did not discolor throughout 7 d of display, whereas steaks packaged in HiO2 MAP discolored. Although steaks packaged in VP did not discolor throughout display and had good color stability, they have a purplish red color that is not acceptable to most consumers. Packaging meat in ULO2CO MAP is an effective way to maintain red color and minimize any negative effects on tenderness or sensory attributes.

Figure 10. Discoloration score [1 = none (0%), 2 = slight discoloration (1 to 19%), 3 = small discoloration (20 to 39%), 4 = modest discoloration (40 to 59%), 5 = moderate discoloration (60 to 79%), 6 = extensive discoloration (80 to 99%), 7 = total discoloration (100%)] means and SE (0.12) for nonenhanced (NoEnh) or enhanced (Enh) longissimus (LL), semitendinosus (ST), and triceps brachii (TB) steaks packaged in different atmospheres (HiO2 = 80% O2, 20% CO2; ULO2CO = 64.6% N2, 35% CO2, 0.4% CO; and VP = vacuum packaging) and displayed. a–fMeans within the same treatment day with different letters differ \( P < 0.05 \); listed in order of data points from top to bottom in the corresponding day for HiO2 treatments only; all other treatments are a,z. y,zMeans within day within muscle and enhancement treatment with different letters differ \( P < 0.05 \); listed in order of data points from top to bottom in the corresponding day for HiO2 treatments only; all other treatments are a,z.

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