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Effects of Dark Storage and Retail Display on Beef Chunk and Round Muscles Enhanced with Ammonium Hydroxide, Salt, and Carbon Monoxide

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Effects of dark storage and retail display on beef chuck and round muscles enhanced with ammonium hydroxide, salt, and carbon monoxide[1,2]

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ABSTRACT: The objective of this study was to determine the retail shelf stability of beef chuck and round muscles enhanced with ammonium hydroxide, salt, and carbon monoxide. A split plot design was used for each of 3 muscles [triceps brachii (TB), biceps femoris (BF), and rectus femoris (RF)] with 2 treatments (0 and 20% pump), 3 dark storage periods (1, 2, and 3 wk), and 3 replications in the whole plot and retail display period as the split plot. There were a total of 12 subprimals per treatment per dark storage period (n = 72 each). Individual steaks were cut to a thickness of 2.54 cm and packaged in a modified-atmosphere package (MAP). The TB was packaged in a high-oxygen MAP (80% oxygen, 20% carbon dioxide). The BF and RF were packaged in a low-oxygen MAP (100% carbon dioxide). At the completion of each dark storage period, steaks were subjected to 7 d of simulated retail display. Steaks were used for objective and subjective color measurements, total plate counts, and determination of retail purge and oxidation. For all muscles, total plate counts were always numerically greater in injected steaks. Triceps brachii steaks held in dark storage for 3 wk and displayed at retail for 4 or more days all exceeded 10^7 log of cfu/cm^2 for aerobic plate count. Biceps femoris and RF steaks packaged in a low-oxygen MAP had much lower bacterial counts, with levels below 4.2 log of cfu/cm^2, even after 7 d of retail display. Oxidation values for the TB were extremely high (ranging from 12.3 to 26.6), whereas the BF and RF had values that were much lower (≤1.0 mg of malonaldehyde/kg of muscle), likely due to the oxidation occurring in a high-oxygen MAP for the TB. Enhanced TB steaks proved to have greater color stability (less discoloration) than nonenhanced TB steaks. In addition, the BF and RF (low-oxygen MAP) steaks had better color stability (more stable redness values) than TB (high-oxygen MAP) steaks, although TB steaks initially exhibited a brighter red color. Retail display life was enhanced by packaging in 100% carbon dioxide, and enhanced steaks exhibited greater color stability in retail display than control steaks.

Key words: enhancement, modified-atmosphere packaging, retail display, shelf life

INTRODUCTION

Enhancement of meat pH with a solution containing ammonium hydroxide, carbon oxide, and salt has been shown to improve consumer palatability ratings (Everts et al., 2006; Nath et al., 2006). Little is known about the effect of this enhancement system (patent pending for Freezing Machines Inc., Dakota Dune, SD) on color and microbial stability of beef.

The presence of carbon monoxide in the enhancement solution could be expected to increase color stability and maintain red color because low levels of carbon monoxide in modified-atmosphere packages (**MAP**) often cause this effect in fresh beef (Luno et al., 2000; Jayasingh et al., 2001; John et al., 2005; Grobbel, 2007). Grobbel (2007) found injection-enhanced steaks packaged in a low-oxygen MAP discolored at a slower rate and to a lesser extent than steaks packaged without carbon monoxide. However, the enhancement system did not include pH adjustment from ammonium hydroxide.

Hand et al. (2006) reported lower levels of inoculated *Escherichia coli* O157:H7 on pH-enhanced round steaks after 0 or 2 d of anaerobic storage but higher levels after 14, 28, or 56 d of anaerobic storage. Under aerobic conditions, lower levels of inoculated *E. coli* O157:H7 were observed. These results raise some questions about the microbial stabililty of pH-adjusted meat.

Luno et al. (2000) reported that MAP of fresh beef in an atmosphere containing ≤1% carbon monoxide in

conjunction with 24% oxygen reduced total aerobic population numbers when compared with MAP with no carbon monoxide and 70% oxygen. There was no effect on lactic acid bacteria.

The objectives of this study were to evaluate the color and microbial stability of beef chuck and round muscles enhanced with ammonium hydroxide, salt, and carbon monoxide.

MATERIALS AND METHODS

Experimental Design

The 3 muscles (n = 72 for each muscle) studied were the triceps brachii (**TB**), biceps femoris (**BF**), and rectus femoris (**RF**). Subprimals were randomly assigned to treatment (0 or 20% pump level) and dark storage period (1, 2, and 3 wk). There were 3 replications.

Raw Meat Materials

No approval was obtained from the Institutional Animal Care and Use Committee because samples were obtained from a federally inspected slaughtering facility.

Vacuum-packaged subprimals containing the TB, BF, and RF muscles (72 clod hearts, NAMP #114E; 72 sirloin caps, NAMP #184D; and 72 peeled knuckles, NAMP #167A, respectivly; NAMP, 2007) from USDA Choice beef carcasses were obtained from Tyson Foods in Dakota City, Nebraska. Subprimals (2 to 3 d postmortem) were randomly assigned to 1 of 2 pump levels and 1 of 3 dark storage periods. For each muscle, there were 24 subprimals per each of 3 replications (12 per pump level, 4 per dark storage period). Preparation of the samples took place at the Beef Products Inc. facility (Dakota City, NE) on 3 dates, with 1 subprimal type tested on each date.

Injection of Samples

All operations occurred in a cool room (<10°C). For each replication, 12 subprimals assigned to the 0% pump level (**CON**, control) were unpackaged and cut. Subprimals (12) assigned to the 20% pump level (**PUMP**) were unpackaged and an initial weight was taken. Subprimals were injected with a solution containing 1% sodium chloride and sufficient ammonium hydroxide to bring the pH of the brine to 11.4. The solution also contained carbon monoxide, which was formulated so that a 20% pump would result in less than 0.4% CO in the finished product. This is patented technology from Freezing Machines Inc. (US Patents 7,022,361 and 7,094,435). Subprimals were injected to the target pump level using an 88-needle Fomaco injector (model #FGM-88SW, Koge, Denmark). Once injected, subprimals were weighed to determine the actual pump level. The second and third replications followed in the same manner. Actual pump levels achieved were 19.1, 19.5, and 19.6% for RF, TB, and BF, respectively.

Cutting Procedures

After a final weight was recorded and the pump level was determined, 3 to 4 steaks from each subprimal were cut parallel to the cut surface to a thickness of 2.54 cm. The TB was cut ventral to dorsal, the BF was cut dorsal to ventral, and the RF was cut proximal to distal. Steaks were then trimmed of excess fat and muscles, leaving the muscle of interest (TB, BF, and RF) intact.

Packaging Procedures

Individual TB steaks were placed in 3.81-cm-deep trays (#10 tray, Jamestown Plastics, Brockton, NY) containing an absorbent pad (PL-75, Sealed Air Corp., Duncan, SC). A Ross Jr. A-10 model packaging machine (Midland, VA) was used with premixed gas (80% oxygen, 20% carbon dioxide) to package the steaks. The trays were sealed with Cryovac lid 1050 film (Duncan, SC), sorted into boxes, and sent to the cold storage facility (−2°C) at Beef Products, Inc. (Dakota City, NE). On the following day, the boxes were shipped approximately 3 h in a refrigerated truck to the Loeffel Meat Laboratory at the University of Nebraska.

Pairs of BF steaks and individual RF steaks were placed in 0.635-cm-deep trays (#10 tray, Jamestown Plastics) containing an absorbent pad (PL-75, Sealed Air Corp.) and packaged in a low-oxygen MAP (100% carbon dioxide) on a Ross Jr. A-10 model packaging machine (Midland, VA). The trays were sealed with Cryovac lid 1050 film (Duncan, SC), sorted into boxes, and sent to the cold storage facility (−2°C) at Beef Products Inc. (Dakota City, NE). On the following day, the boxes were shipped approximately 3 h in a refrigerated truck to the Loeffel Meat Laboratory at the University of Nebraska.

Dark Storage

Subprimals were randomly assigned to 1 of 3 dark storage periods (1, 2, or 3 wk). Samples remained in their MAP and boxes for the assigned dark storage period. Boxes remained in the Loeffel Meat Laboratory cooler (4°C) for their assigned storage period before retail display.

Retail Display

Two Tyler retail display cases (model LNSC5, Tyler Refrigeration Corporation, Niles, MI) were used for retail display. Three 2-bulb (3000K Universal Fluorescent, Philips F32T8/ADV830/ALTO, Somerset, NJ) fixtures covered the whole display area to an intensity of 1,614 lx for 24 h/d. To minimize potential effects of variation in temperature throughout the retail display case, samples were randomly shifted around every day.

Table 1. Least squares means for total aerobic plate counts ($\log_{10}$ cfu/cm^2) of pumped and nonpumped (control) triceps brachii beef steaks as influenced by pumping and retail display

Dark storage, wk	Treatment	Retail display, d 0	4	7	SE
1	Control	2.20[b]	2.66[b,y]	3.66[a,y]	0.30
	Pump	2.58[c]	4.77[b,x]	6.76[a,x]	0.30
2	Control	2.86[b]	3.51[ab,y]	3.74[a,y]	0.30
	Pump	3.33[b]	6.20[a,x]	6.80[a,x]	0.30
3	Control	3.96[c,y]	4.78[b,y]	6.00[a,y]	0.30
	Pump	5.42[b,x]	7.22[a,x]	7.36[a,x]	0.30

[a–c]Means with different superscripts within the same row differ significantly ($P < 0.05$).

[x,y]Means with different superscripts within the same column differ significantly ($P < 0.05$).

The retail display cases were placed in a dark room and the only lighting came from the fluorescent lighting. The retail display cases operated at <4°C. On one occasion, during display of the RF steaks, a malfunction was discovered. One retail display case approached 10°C for less than a single day. Subsequent color data from those samples were deleted from the data set.

Assignment of Steaks

After the dark storage period, samples were placed in the retail display cases. The first steak from each sample was used for determination of total plate count, retail purge, and oxidation on d 0 of retail display and was not placed in the retail display case. Steak 2 was placed in retail display and removed on d 4 for determination of total plate counts and retail purge. Steak 3 from each sample received color measurements and discoloration scores on d 0 to 7. On d 7 of retail display, the steaks were removed and used for total plate counts, retail purge, and oxidation for d 7 of retail display.

Color Measurements

L*, a*, and b* measurements were taken every day of retail display on steaks displayed for 7 d by a Hunter

Lab Mini Scan XE Plus (Model 45/0-L, Reston, VA) hand-held colorimeter. The colorimeter was fitted with a 2.54-cm port and was standardized using a black tile and a white tile (X = 78.5, Y = 83.2, and Z = 88.7). Color measurements for L*, a*, and b* were taken with the colorimeter using illuminant A and 10° standard observer. Three measurements were taken across the whole steak and an average was recorded. The measurements were taken through the packaging film. To avoid getting purge on the surface of the steak, trays containing the TB steaks were tipped to the side, such that purge remained on one edge of the package and the meat touched the packaging film so that color measurements could be made.

Discoloration scores were assigned by an experienced panelist (a graduate student trained to do so and who had been involved with previous color research) to steaks under retail display for 7 d. Discoloration scores ranged from 1 to 11 (0, 1 to 10, 11 to 20, 21 to 30, 31 to 40, 41 to 50, 51 to 60, 61 to 70, 71 to 80, 81 to 90, and 91 to 100%). Scores for discoloration were determined immediately after the color measurements were taken with the colorimeter.

Total Plate Counts

Steaks pulled on d 0, 4, and 7 of retail display were sampled for total plate counts. A 6.45-cm^2 area was swabbed in 3 directions with a BD BBL CultureSwab (BD Biosciences, Sparks, MD). Swabs were then sent to the microbiology lab in Food Science at the University of Nebraska. Aerobic plate counts (TB steaks) were obtained using the pour plate method. Swabs were vortexed for 10 to 15 s in 10 mL of sterile PBS in 16 × 125-mL tubes for the first 10^1 dilution. Serial dilutions were then made from this solution. Initially, samples were plated at dilutions of 1:10, 1:100, and 1:1,000 (vol/vol) and plated on plate count agar (Difco, Becton Dickinson and Company, Sparks, MD). Dilutions were increased if necessary for the next sampling period. Plates for aerobic plate counts were incubated at 32 ± 1°C for 48 h. Plates for anaerobic plate counts (BF and RF steaks) were plated on plate count agar and incubated at 32 ± 1°C for 48 h in rectangular jars (Mitsubishi Gas Chemi-

Table 2. Least squares means for total anaerobic plate counts ($\log_{10}$ cfu/cm^2) of pumped and nonpumped (control) biceps femoris beef steaks as influenced by pumping, dark storage, and retail display

Treatment	Retail display, d	Dark storage, wk 1	2	3	SE	Retail display, d 0	4	7	SE
Control		0.22[b]	1.01[a]	1.01[a,y]	0.20	0.34[b]	0.68[b,y]	1.21[a,y]	0.18
Pump		0.57[c]	1.25[b]	3.12[a,x]	0.20	0.63[c]	1.85[b,x]	2.46[a,x]	0.18
	0	0.22[b]	0.20[b,z]	1.03[a,y]	0.22				
	4	0.41[c]	1.06[b,y]	2.34[a,x]	0.22				
	7	0.56[c]	2.14[b,x]	2.82[a,x]	0.22				

[a–c]Means with different superscripts within the same row differ significantly ($P < 0.05$).
[x–z]Means with different superscripts within the same column differ significantly ($P < 0.05$).

Table 3. Least squares means for total anaerobic plate counts ($\log_{10}$ cfu/cm^2) of pumped and non-pumped (control) rectus femoris beef steaks as influenced by pumping, dark storage, and retail display

| Treatment | Retail display, d | Dark storage, wk | | | | Retail display, d | | | |
		1	2	3	SE	0	4	7	SE
Control		0.77[a]	1.45[b]	2.14[c,x]	0.18	1.08[a]	1.25[b]	2.02[c,x]	0.17
Pump		0.98[a]	1.73[b]	3.20[c,y]	0.18	1.14[a]	1.46[b]	3.32[c,y]	0.17
	0	0.30[a,x]	0.38[a,x]	1.76[b,x]	0.21				
	4	1.20[a,y]	1.62[b,y]	2.15[b,x]	0.21				
	7	1.13[a,y]	2.77[b,z]	4.11[c,y]	0.21				

[a–c]Means with different superscripts within the same row differ significantly ($P < 0.05$).
[x–z]Means with different superscripts within the same column differ significantly ($P < 0.05$).

cal, New York, NY) containing Anaero-packs (Mitsubishi Gas Chemical Company Inc., Tokyo, Japan) for anaerobic atmosphere generation.

Results were reported as the number of $\log_{10}$ colony forming units per square centimeter (cfu/cm^2).

Retail Purge

Purge was determined on steaks pulled on d 0, 4, and 7 of retail display. The entire package (steak included) was weighed for a primary weight. The steak was removed, the surface blotted with a paper towel, and the steak was weighed to determine the weight. These weights were used, in addition to the weight of a package, film, and unused absorbent pad (tare weight), to calculate the amount of purge incurred during the retail display.

The percentage purge was calculated by the following equation: [primary weight – steak weight – tare weight)/(primary weight – tare weight)] × 100.

Oxidation

The thiobarbituric acid assay (**TBA**) of Ahn et al. (1998) was used, with the modifications of Buege and Aust (1978).

Thiobarbituric acid values were determined for steaks pulled after 0 and 7 d of retail display. Fourteen milliliters of double-distilled water was added to a 5-g

sample randomly obtained from steak powdered in liquid nitrogen and 1.0 mL of butylated hydroxyanisole [10% (vol/vol) stock solution dissolved in 90% ethanol]. The mixture was homogenized and then centrifuged at $2,000 \times g$ for 5 min. Two milliliters of TBA/TCA was added to 1 mL of homogenate, vortexed, and then incubated in a 70°C water bath for 30 min to develop color. Samples were then allowed to cool in a cold water bath (13°C) for 10 min before centrifuging at $3,000 \times g$ for 15 min. Duplicate aliquots of 200 µL were transferred to a 96-well plate. Absorbance was read at 540 nm on a Dynatech Laboratories MR5000 plate reader (Chantilly, VA) and analyzed by BioLinx assay management software (Dynatech Laboratories).

Results were expressed as milligrams of malonaldehyde per kilogram of sample.

Statistical Analysis

The experimental design for total plate counts, TBA values, and retail purge was a split plot, with treatment and period of dark storage in the whole plot and day of retail display as the split plot. Replication was used as the block. Color data were analyzed as a repeated measures design, with treatment and period serving

Table 4. Least squares means for retail purge[1] of pumped and nonpumped (control) triceps brachii beef steaks as influenced by retail display, dark storage, and pumping

| Retail display, d | Dark storage, wk | | | | Treatment | |
	1	2	3	SE		
0	3.55[b,z]	5.15[a,y]	4.76[a,z]	0.36	Control	4.89[b]
4	7.29[y]	8.04[x]	7.78[y]	0.36	Pump	9.06[a]
7	8.64[x]	8.31[x]	9.23[x]	0.36	SE	0.23

[a,b]Means with different superscripts within the same row differ significantly ($P < 0.05$).
[x–z]Means with different superscripts within the same column differ significantly ($P < 0.05$).
[1]Retail purge expressed as percentage.

Table 5. Least squares means for retail purge[1] of pumped and nonpumped (control) biceps femoris beef steaks as influenced by pumping, dark storage, and retail display

| Treatment | Retail display, d | | | |
	0	4	7	SE
Control	11.59[x]	12.18	12.60	0.51
Pump	10.18[c,y]	12.25[b]	13.27[a]	0.51

| | Dark storage, wk | | | |
	1	2	3	SE
	11.52[b]	11.51[b]	13.00[a]	0.47

[a,b]Means with different superscripts within the same row differ significantly ($P < 0.05$).
[x,y]Means with different superscripts within the same column differ significantly ($P < 0.05$).
[1]Retail purge expressed as percentage.

Table 6. Least squares means for retail purge[1] from pumped and nonpumped (control) rectus femoris beef steaks as influenced by pumping, dark storage, and retail display

Treatment	Dark storage, wk			SE
	1	2	3	
Control	$6.51^{a,x}$	$6.88^{a,x}$	$8.25^{b,x}$	0.35
Pump	$10.40^{a,y}$	$13.79^{b,y}$	$13.46^{b,y}$	0.35
Treatment	Retail display, d			SE
	0	4	7	
Control	6.99^{x}	7.55^{x}	7.11^{x}	0.31
Pump	$11.33^{a,y}$	$11.67^{a,y}$	$14.65^{b,y}$	0.31

[a,b]Means with different superscripts within the same row differ significantly ($P < 0.05$).
[x,y]Means with different superscripts within the same column differ significantly ($P < 0.05$).
[1]Retail purge expressed as a percentage.

as the whole plot, retail display day as the repeated measure, and sample as the block. Treatments, dark storage periods, and retail display times were considered fixed effects and replication was considered a random effect.

Data were analyzed using the GLIMMIX procedure (SAS Inst. Inc., Cary, NC). When significance ($P \leq 0.05$) was indicated by ANOVA, means separations were performed using the LSMEANS and DIFF functions of SAS.

RESULTS AND DISCUSSION

Bacterial Counts

For the TB (high-oxygen MAP), CON steaks always had numerically lower counts aerobic plate counts than PUMP steaks (Table 1). There was a 3-way interaction between treatment, storage period, and retail display time for the TB. Total plate counts increased as retail display time and dark storage time increased. Dainty and Mackey (1992) found that bacterial spoilage occurs when the number of bacteria reaches 10^7 logs of cfu/cm^2. The enhanced TB steaks did not reach the spoilage level until d 4 of retail display after 3 wk of dark storage

($7.22 \log_{10}$ cfu/cm^2). Thus, maximum storage for an enhanced steak in high-oxygen MAP, based on aerobic plate count, was between 21 and 25 d.

The CON BF and RF (packaged in low-oxygen MAP) steaks had lower anaerobic plate counts for every dark storage period and retail display time (Tables 2 and 3). The greatest increase in total anaerobic plate counts occurred with the PUMP steaks between wk 2 and 3 of dark storage. Total anaerobic plate counts increased as dark storage time and retail display time increased. For the BF, total anaerobic plate counts increased from 0.22 to 2.82 $\log_{10}$ cfu/cm^2 during this study. The RF total plate counts increased from 0.30 to 4.11 $\log_{10}$ cfu/cm^2. The difference between the final total plate counts of these muscles may be because, during the study of the RF, one of the retail display cases malfunctioned and some steaks were exposed to warmer temperatures (up to 10°C) for less than 1 d. Steaks packaged in the low-oxygen MAP did not reach the spoilage level previously indicated for aerobic microbial growth thresholds of spoilage, suggesting that maximum storage time was more than 28 d.

Hand et al. (2006) found that pH-enhanced steaks had lower counts than nonenhanced controls that were inoculated, vacuum-packaged, and stored for 0 and 2 d. However, nonenhanced steaks held in anaerobic storage for 14, 28, and 56 d had lower *E. coli* O157:H7 counts than the pH-enhanced steaks. In addition, Hand et al. (2006) found the enhanced steaks to have lower *E. coli* O157:H7 counts than nonenhanced steaks that were retail-wrapped and displayed for 0 and 2 d. On d 14 of retail display, there was no difference in *E. coli* O157:H7 counts between the enhanced and nonenhanced steaks. Hand et al. (2006) also determined the pH of the enhanced steaks to be 7.72 and that of nonenhanced steaks to be 5.61. The results from Hand et al. (2006), along with the results from this study, suggest that the sudden change of pH from enhancement disrupts the growth and amount of bacteria on steaks. However, bacteria appear to grow faster as the incubation time increases with the higher pH from enhancement than at the lower pH found in controls.

Retail Purge

For the TB (high-oxygen MAP), PUMP steaks had nearly twice as much retail purge (Table 4) as CON

Table 7. Least squares means for thiobarbituric acid values[1] from pumped and nonpumped (control) triceps brachii beef steaks as influenced by pumping, retail display, and dark storage

Retail display, d	Dark storage, wk				Treatment		
	1	2	3	SE	Control	Pump	SE
0	12.31	13.06	13.73^{y}	2.26	13.44^{y}	12.58	1.53
7	12.73^{b}	15.65^{b}	$26.61^{a,x}$	2.21	$21.73^{s,x}$	14.99^{t}	1.57

[a,b]Means with different superscripts within the same row differ significantly ($P < 0.05$).
[s,t]Means with different superscripts within the same row differ significantly ($P < 0.05$).
[x,y]Means with different superscripts within the same column differ significantly ($P < 0.05$).
[1]Values are expressed as milligrams of malonaldehyde per kilogram of muscle tissue.

Table 8. Least squares means for thiobarbituric values[1] from pumped and nonpumped (control) biceps femoris beef steaks as influenced by pumping, dark storage, and retail display

	Treatment		Retail display, d	
Item	Control	Pump	0	7
Concentration	4.65[a]	1.84[b]	2.12[b]	4.37[a]
SE	0.66	0.65	0.61	0.58

[a,b]Means with different superscripts within the same row differ significantly ($P < 0.05$).
[1]Values expressed as milligrams of malonaldehyde per kilogram of muscle tissue.

(9.06 to 4.89%). Also, the amount of purge increased as dark storage lengthened and retail display increased. Generally, there was no significant increase in retail purge between samples after 4 d of retail display and 7 d of retail display, regardless of dark storage time. This suggests that most of the retail purge has occurred by 4 d of retail display.

For the BF (Table 5), there was no significant difference for CON steaks as retail display time increased ($P \leq 0.050$). Enhanced steaks had less retail purge at d 0 of retail display, but were not different from CON at 4 and 7 d of retail display. Moreover, there was no difference between 1 and 2 wk of dark storage, with 3 wk of dark storage exhibiting the most retail purge (13.00%).

The CON steaks of the RF (Table 6) always had less retail purge than PUMP. There was no difference in purge for the CON as retail display time increased, whereas PUMP steaks increased from d 0 to 4 of retail display. Also, PUMP steaks at 2 and 3 wk of dark storage had the most retail purge.

Oxidation Values

Because the TB were packaged in a high-oxygen MAP, one would expect oxidation (TBA) values to be much greater than the BF and RF. This was the case even though the 3 muscles were not statistically compared. The TB exhibited very high TBA values ranging from 12.31 to 26.61 mg of malonaldehyde/kg of muscle tissue (Table 7). All values were much greater than the baseline of 1.0 mg of malonaldehyde/kg of muscle tissue (Tarladgis et al., 1960), which indicates spoilage.

Both PUMP BF and RF steaks had much lower TBA values than the TB (Tables 8 and 9). This is likely because the BF and RF were packaged in a low-oxygen MAP. Enhanced RF steaks did not show a significant increase in TBA values as dark storage increased ($P \geq 0.05$). The TBA values doubled for the BF steaks between 0 and 7 d of retail display. The nonenhanced RF TBA values tripled over the same period (0.76 to 2.13 mg of malonaldehyde/kg of muscle tissue), whereas the PUMP RF steaks did not significantly change (0.53 to 0.63 mg of malonaldehyde/kg of muscle tissue). Moreover, all values for PUMP RF steaks were less than 1.0, suggesting that the enhancement procedure helps prevent oxidation when in a low-oxygen MAP. The TBA values for the CON steaks more than doubled for each week of dark storage: 0.60, 1.22, and 2.52 for 1, 2, and 3 wk, respectively.

Color

Triceps brachii steaks, regardless of dark storage period, gradually got darker as retail display time progressed until d 4 (Figure 1). The L* values then slightly increased (lighter color) until d 7. Enhanced TB steaks tended to be darker (lower L*) and were always redder (a*) than CON steaks at the same sampling time. Values for a* gradually decreased over the 7-d retail display period (Figure 2). Steaks (TB) in 1 wk of dark storage before retail display had the lowest discoloration scores up to d 3 (Figure 3). Between d 3 and 4 of retail display, there was a sharp increase for discoloration scores. Enhanced steaks almost always had less discoloration than their CON counterparts.

Enhanced BF steaks were always lighter in color than CON steaks (Figure 4) for the first 4 d of retail display. In addition, steaks with 2 and 3 wk of dark storage had sharp increases in L* values between 1 and 2 d of retail display (Figure 5). Steaks with 3 wk of dark storage increased in L* value within the first day of retail display. The values for a* decreased for all BF treatments

Table 9. Least squares means for thiobarbituric acid values[1] from pumped and nonpumped (control) rectus femoris beef steaks as influenced by pumping, dark storage, and retail display

Treatment	Retail display, d	Dark storage, wk				Retail display, d		
		1	2	3	SE	0	7	SE
Control		0.60[b]	1.22[b,x]	2.52[a,x]	0.27	0.76[b]	2.13[a,x]	0.39
Pump		0.41	0.46[y]	0.87[y]	0.27	0.53	0.63[y]	0.39
	0	0.40	0.59	0.94[y]	0.27			
	7	0.61[b]	1.08[b]	2.45[a,x]	0.27			

[a,b]Means with different superscripts within the same row differ significantly ($P < 0.05$).
[x,y]Means with different superscripts within the same column differ significantly ($P < 0.05$).
[1]Values are expressed as milligrams of malonaldehyde per kilogram of muscle tissue.
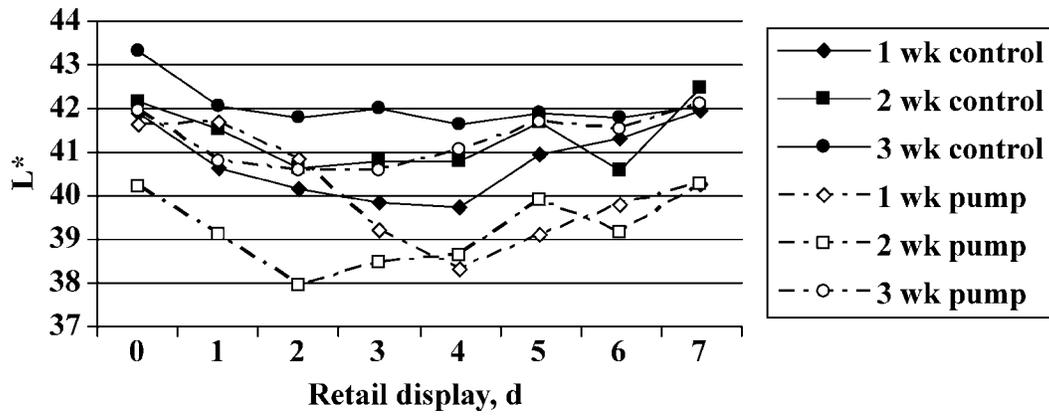
Figure 1. L* values for triceps brachii steaks in retail display as influenced by pumping and dark storage (1, 2, or 3 wk).
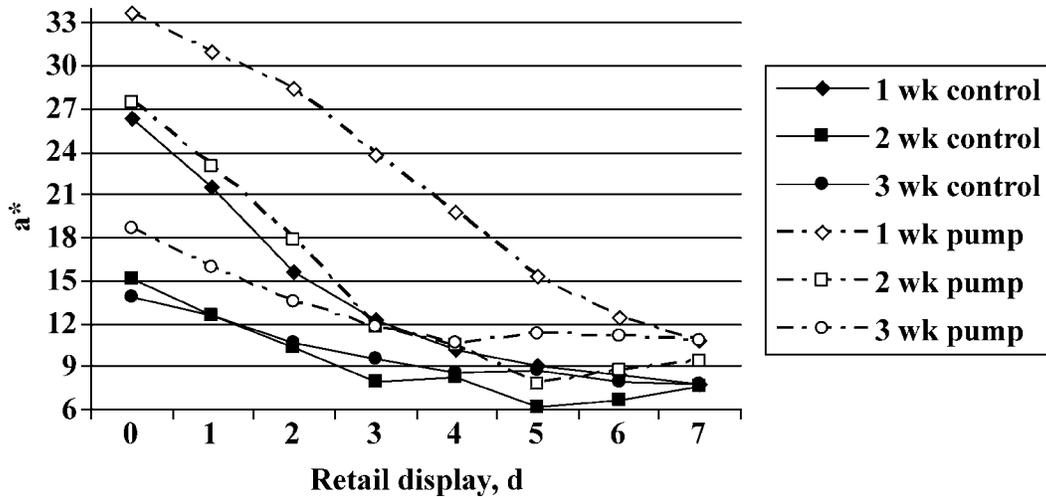

Figure 2. a* values for triceps brachii steaks in retail display as influenced by pumping and dark storage (1, 2, or 3 wk).
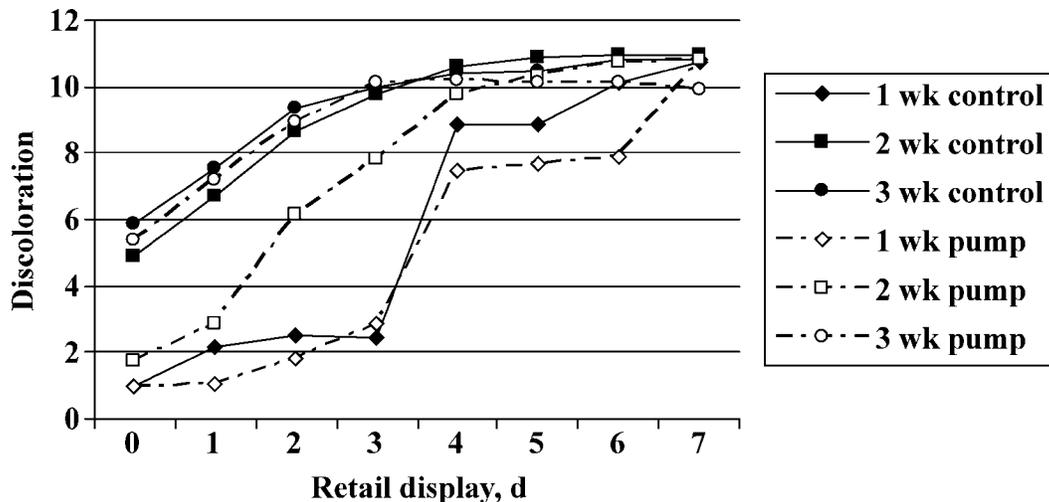

Figure 3. Discoloration of triceps brachii steaks in retail display as influenced by pumping and dark storage (1, 2, or 3 wk).
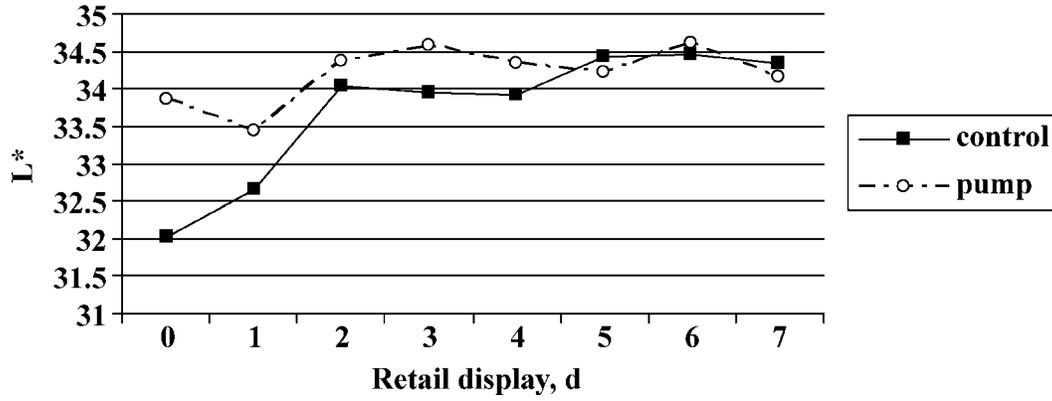
Figure 4. L* values for biceps femoris steaks in retail display as influenced by pumping.
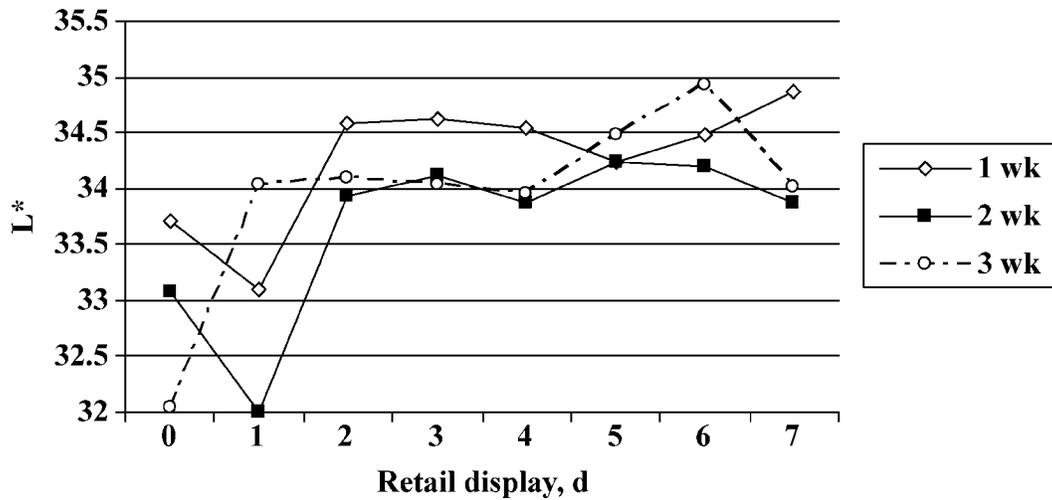

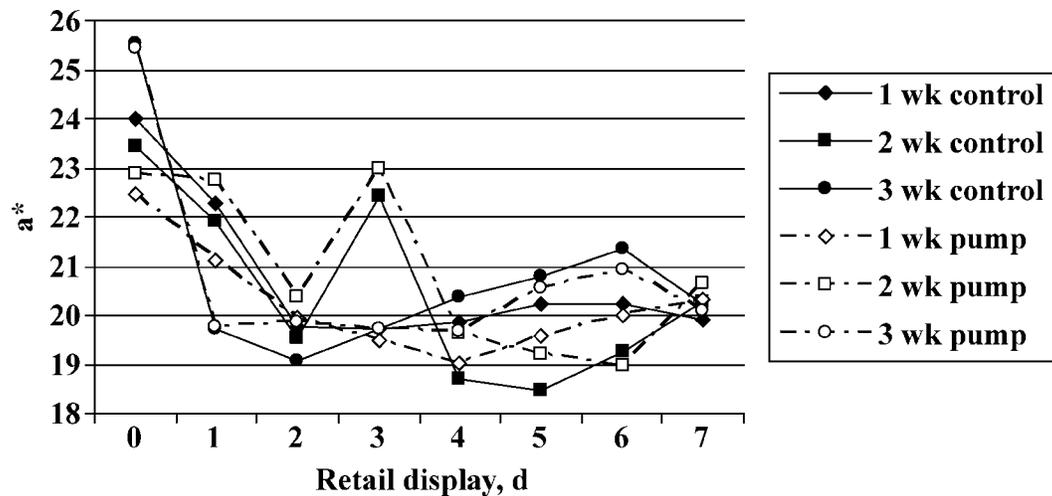**Figure 5.** L* values for biceps femoris steaks in retail display as influenced by dark storage.

Figure 6. a* values for biceps femoris steaks in retail display as influenced by pumping and dark storage (1, 2, or 3 wk).
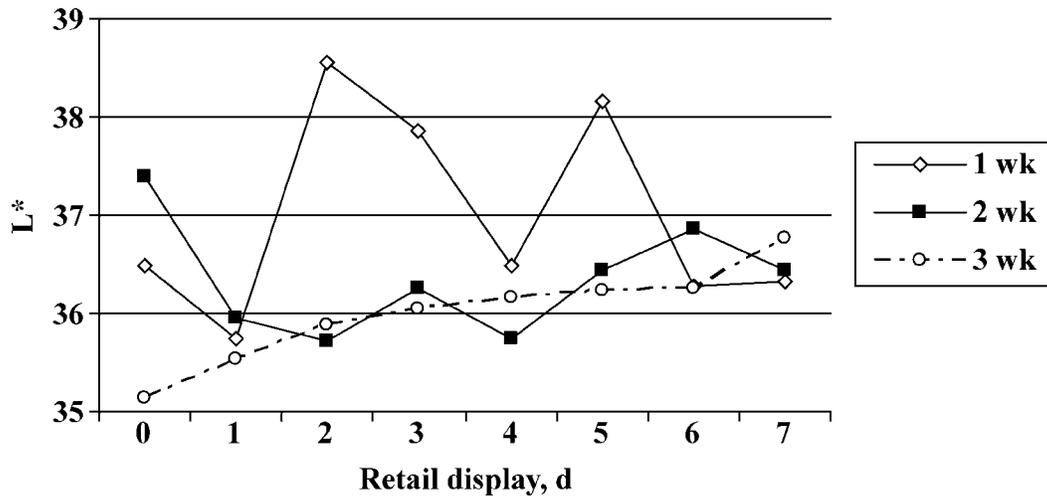
Figure 7. L* values for rectus femoris steaks in retail display as influenced by dark storage.

upon retail display (Figure 6). Steaks with 3 wk of dark storage had the sharpest decrease in a* within the first 24 h of retail display. Samples were variable throughout the display period but converged on d 7. It should be noted that the data sets for color and discoloration for the RF were smaller due to the fact that one of the retail display cases malfunctioned during the study. As a result, some of the RF steaks were deleted from the data set. Rectus femoris L* values for 1 wk of dark storage were highly variable (Figure 7). There was a large peak between d 1 and 2, which was also noted with the BF. Enhanced RF steaks were redder (higher a* values) throughout the whole retail display period (Figure 8).

Triceps brachii steaks had much higher L* values than both the BF and RF steaks. This could be because the TB was packaged in a high-oxygen MAP, whereas the BF and RF were packaged in a low-oxygen MAP.

Subjectively (data not shown), less discoloration was observed in the BF and RF steaks, probably because the steaks were in the deoxymyoglobin state. One would expect the steaks in the high-oxygen MAP early in retail display to have a more desirable, bright red oxymyoglobin color than the purple color of deoxymyoglobin in a low-oxygen MAP. However, oxidation quickly turns oxymyoglobin to metmyoglobin, producing an undesirable brown color.

In summary, when packaged in a high-oxygen MAP, the enhanced steaks proved to have less discoloration and oxidation than the nonenhanced steaks. However, with the extended dark storage period and retail display time in this study, both the oxidation levels and bacterial counts made all steaks unacceptable at the end of the 3-wk dark storage and 7-d retail display period. When packaged in a low-oxygen MAP, enhanced steaks proved to have less discoloration and oxidation than
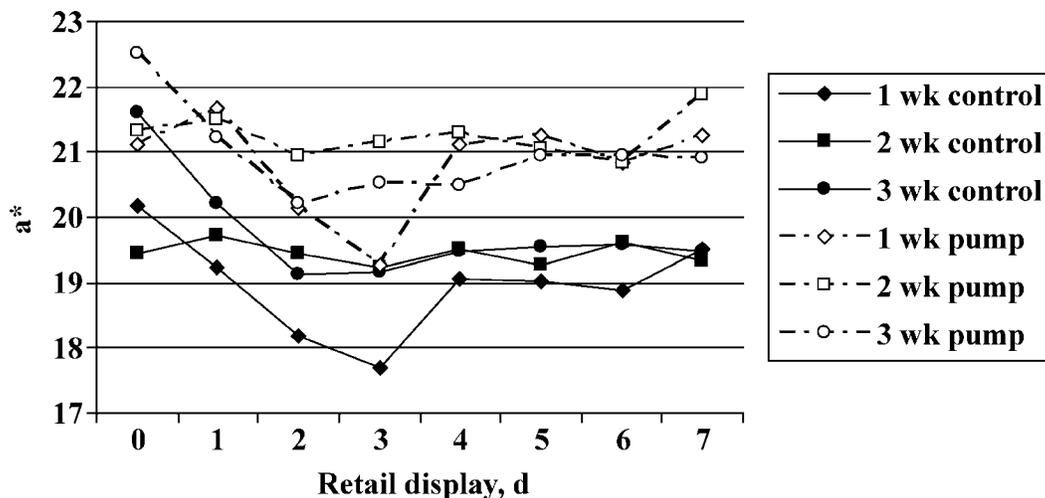

Figure 8. a* values for rectus femoris steaks in retail display as influenced by pumping and dark storage (1, 2, or 3 wk).

nonenhanced steaks. Enhanced steaks always had greater bacterial counts than the nonenhanced steaks. Data suggest that enhancement of beef chuck and round muscles can improve and extend shelf life under low-oxygen packaging conditions.

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