



## Retail Display Properties and Consumer Perception of Extended Aged Beef Topically Treated with Ascorbic Acid and Rosemary Extract<sup>1</sup>

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**Abstract:** Extended aging increases browning and decreases redness in fresh beef. The goal of this study was to test an already proven combination of antioxidants (0.05% ascorbic acid + 0.1% rosemary extract) using a method that could be applied at the retail level to simply and effectively extend the shelf-life of extended aged beef. The specific objective was to analyze the effect of topical application of ascorbic acid and rosemary extract on color, lipid oxidation, microbial growth, and sensory perception of beef *longissimus lumborum* (LL;  $n = 12$ ) and *semimembranosus* (SM;  $n = 12$ ) muscles wet aged at 0°C for 14, 28, and 42 (extended aging period) days. After aging, steaks were cut, sprayed with 2 mL of a 0.05% ascorbic acid + 0.1% rosemary extract solution (treated) or untreated (control), and subjected to retail display. Antioxidant treated LL steaks had greater ( $P < 0.05$ )  $L^*$  (lightness) values, but lower ( $P < 0.05$ )  $a^*$  (redness) and  $b^*$  (yellowness) values than control steaks. Furthermore, antioxidant treatment decreased ( $P < 0.05$ ) browning on d 4 of retail display compared to control steaks. Consumers scored antioxidant treated SM steaks as less tender on d 28, more juicy on d 14 but less juicy on d 18 and 42. Antioxidant treatment did not affect lipid oxidation, microbial growth, or sensory flavor scores. As expected, longer aging periods resulted in less color stability of LL and SM steaks. Although the antioxidant treatment resulted in measurable subjective color improvements, these improvements are likely not detectable by the consumer.

**Keywords:** aging, ascorbic acid, beef, rosemary extract

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## Introduction

The 2015 National Beef Tenderness Survey reported that beef sold at retail establishments had post fabrication aging times ranging from 6 to 102 d

(Martinez et al., 2017). It is well accepted that aging improves beef tenderness (Eilers et al., 1996; Bratcher et al., 2005; Gruber et al., 2006; Dixon et al., 2012; Colle et al., 2015; 2016); however, aging also reduces the color stability of product during subsequent retail display (Colle et al., 2015; 2016). Color plays a critical role in a consumer's decision to purchase a meat product (Mancini and Hunt, 2005). Accordingly, discoloration causes meat to be perceived by consumers as less wholesome (Faustman and Cassens, 1990; Suman et al., 2014; Neethling et al., 2017).

Longer aging periods, such as the ones reported in the 2015 National Beef Tenderness Survey, result in product management issues at the retail level such as reduced color stability. Colle et al. (2015; 2016)

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aged *longissimus lumborum* (LL), *gluteus medius* (GM), *biceps femoris* (BF), and *semimembranosus* (SM) up to 63 d prior to 4 d of retail display. They reported that longer aging resulted in  $a^*$  values (redness) decreasing faster and to a greater extent. Additionally, the amount of browning increased more rapidly and to a greater extent during retail display with longer aging periods in the LL, BF, and SM. In the present study only the LL and SM were evaluated since our previous research demonstrated that these 2 muscles represent extremes for both tenderness and color stability in response to extended aging (> 28 d; Colle et al., 2015; 2016). Furthermore, McKenna et al. (2005) classified the *longissimus dorsi* (LD) and SM as “high” and “intermediate” color stability muscles, respectively.

The use of an antioxidant during retail display could improve color stability and therefore shelf-life of long aged product, while maintaining the positive sensory attributes associated with extended aged beef. Ascorbic acid and rosemary extract are 2 antioxidants that work together synergistically (Elliott, 1999). Ascorbic acid is a reducing and chelating compound that prevents myoglobin oxidation (Ahn and Nam, 2004; Brewer, 2008). Rosemary contains carnosol and carnosic acid which inhibit lipid oxidation by acting as peroxyl radical scavengers (Aruoma et al., 1992; Basaga et al., 1997).

Both ascorbic acid and rosemary extract have been used to improve beef color and reduce lipid oxidation. However, they have never been tested on extended aged (> 28 d) beef. Wheeler et al. (1996) noted that SM steaks injected with a sodium ascorbate solution resulted in improved retail display lean color stability after 15 d of aging and 5 d of retail display and after 17 d of aging and 7 d of retail display. Additionally, with longer retail display times the treated steaks were redder and had less surface discoloration (Wheeler et al., 1996). Ahn et al. (2007) observed that 1.0% rosemary solution lowered the thiobarbituric reactive substances (lipid oxidation) of cooked ground beef after 9 d of storage. Djenane et al. (2003) utilized a combination of ascorbic acid and rosemary extract to extend shelf-life. They reported that LD steaks sprayed with a 0.1% rosemary/0.05% ascorbic acid solution in combination with 1.5% lactic acid and stored for up to 27 d in a Modified Atmosphere Packaging (MAP) significantly delayed the decrease in Commission International de l’Eclairage (CIE)  $a^*$  values (redness) compared to steaks treated with 1.5% lactic acid and stored in a MAP. Furthermore, these authors observed a decrease in lipid oxidation, myoglobin oxidation, as well as less metmyoglobin formation in steaks treated with the antioxidant solution. The use of antioxidants, specifically ascorbic acid and rosemary extract, could increase the retail shelf-

life of long aged (> 28 d) LL and SM steaks by reducing lipid oxidation and improving color stability.

The current study was designed to test an already proven combination of antioxidants (0.05% ascorbic acid + 0.1% rosemary extract; Djenane et al., 2003) using a method that could be applied at the retail level to simply and effectively help retailers manage and increase the shelf-life of extended aged beef whether the beef was intentionally or unintentionally aged for longer than 28 d. Increasing the shelf-life of beef aged longer than 28 d will allow retail stores more time to market the product at the normal price vs. having to mark the product as reduced for quick sale (Suman et al., 2014). In some cases, this mark down in price likely occurs within 24 h due to the poor color stability of extended aged product. To our knowledge, an antioxidant strategy to improve the shelf-life of extended aged beef has not been evaluated. The effects of ascorbic acid and rosemary extract on retail shelf-life of LL and SM steaks from USDA Choice carcasses at varying stages of wet aging were evaluated. Our objectives were to 1) determine the effect of ascorbic acid and rosemary extract on color stability, lipid oxidation, and microbial growth during retail display of LL and SM steaks wet aged for 14, 28, and 42 d; and 2) examine the consumer acceptability of cooked antioxidant treated LL and SM steaks subjected to extended aging. Antioxidant levels were based on previous studies (Wheeler et al., 1996; Djenane et al., 2003; Ahn et al., 2007) and from preliminary experiments using various levels of ascorbic acid and rosemary extract alone or in combination with one another.

## Materials and Methods

### *Human subject participation in consumer panel*

The University of Idaho Institutional Review Board certified this project as Exempt.

### *Product procurement*

Carcasses were fabricated 24 h postmortem. Cooler temperatures and electrical stimulation of the carcasses were the same as described in Colle et al. (2016). Briefly, drip and sales coolers were set at 0 to 1.1°C and 2.8°C, respectively. Carcasses were electrically stimulation for 1 min in each of two sections at 26 to 29 V and 26 V. At 48 h postmortem, strip loin (*longissimus lumborum*) [Institutional Meat Purchase Specifications (IMPS) 180] and top (inside) round (*semimembranosus*; IMPS 168) from one side of USDA Choice carcasses ( $n = 12$ )

were purchased from Washington Beef, a commercial beef processing facility (Toppenish, WA) and transported to the University of Idaho Meat Science Laboratory at refrigerated temperatures.

### Preparation of Product

The LL and SM were removed from their respective wholesale cuts for aging and subsequent analysis. The muscles were cut into 3 similar sized sections ( $\sim 10.5 \times 19 \times 7 \text{ cm}^3$ ). Each section was assigned to one of the 3 aging periods (14, 28, and 42 d post-fabrication) so that muscle location was equally represented in each of the aging periods. Sections were then vacuum shrink packaged (3 mil,  $7 \times 12$  Durashrink bags, 6 to  $7.5 \text{ g/m}^2$ , 24hr water vapor transmission rate, 50 to  $70 \text{ cc/m}^2$ , 24hr  $\text{O}_2$  transmission rate, Winpak Films, Senoia, GA) and subsequently aged at  $0^\circ\text{C}$ .

After the specified aging period, sections were cut into four 2.54 cm-thick steaks. Two steaks were designated to the untreated control group and the remaining 2 to the antioxidant treatment group. One steak in each of these groups was assigned to shelf-life and tenderness analysis, while the other steak was designated for consumer sensory analysis. Preliminary experiments along with previous literature determined the optimum concentration of ascorbic acid (AA) and rosemary extract (RE) on retail color stability. Antioxidant concentrations in solution of 0.05, 0.1, and 0.4% AA; 0.05, 0.1, and 0.25% RE; 0.05% AA + 0.05% RE and 0.05% AA + 0.1% RE were utilized in the preliminary experiments. The top surface of the steaks assigned to the antioxidant treatment group were sprayed with 2ml of the antioxidant (0.05% AA + 0.1% RE) solution and allowed to rest for  $\sim 10$  min, while control steaks were not sprayed. Steaks assigned to shelf-life were weighed then sampled for microbial analysis, thiobarbituric acid reactive substances analysis, metmyoglobin reducing activity, and oxygen consumption. Following procedures in Colle et al. (2016), steaks were overwrapped with an oxygen permeable PVC film,  $17 \text{ gm}/645 \text{ cm}^2$  per 24hr water vapor transmission rate,  $1200 \text{ cc}/645 \text{ cm}^2$  per 24hr  $\text{O}_2$  transmission rate, (Koch Industries, Inc. #7500-3815; Wichita, KS) with the freshly cut surface exposed to oxygen. Steaks were then displayed in a glass-fronted retail display case (Model GDM-69, True Manufacturing Co., O'Fallon, MO) with fluorescent lighting (408 lux) at  $3^\circ\text{C}$  for 4 d.

### pH

The pH was measured initially on the day of fabrication (d0) and then subsequently measured when the

assigned sections were being fabricated into steaks on d 14, 28, and 42 post-fabrication. pH was measured using a portable pH meter (Model SevenGo, Mettler Toledo, Woburn, MA) equipped with an InLab Solids Pro puncture-type electrode (Colle et al., 2015; 2016). The pH meter was calibrated in pH 4.0 and 7.0 buffers each day.

### Retail fluid loss

After steaks were treated and sampled, initial weights were recorded. Steaks were weighed following 4 d of retail display to calculate percent retail fluid loss (Colle et al., 2015; 2016).

$$\text{Percent retail fluid loss} = \frac{\text{initial weight} - \text{final weight}}{\text{initial weight}} \times 100$$

### Retail color

Steaks were allowed to bloom for 60 min. Objective ( $L^*$ ,  $a^*$ , and  $b^*$ ) and subjective (oxygenated lean color; 1 = extremely bright cherry-red, 2 = bright cherry-red, 3 = moderately bright cherry-red, 4 = slightly bright cherry-red, 5 = slightly dark cherry-red, 6 = moderately dark red, 7 = dark red, 8 = extremely dark red, amount of browning; 1 = no evidence of browning, 2 = dull, 3 = grayish, 4 = brownish-gray, 5 = brown, 6 = dark brown, discoloration; 1 = none, 2 = slight, 3 = small, 4 = moderate, 5 = extreme, surface discoloration; 1 = none (0%), 2 = slight (1 to 20%), 3 = small (21 to 40%), 4 = modest (41 to 60%), 5 = moderate (61 to 80%), 6 = extensive (81 to 100%), and color uniformity; 1 = uniform, 2 = slight two-toning, 3 = small amount of two-toning, 4 = moderate two-toning, 5 = extreme two-toning) color were measured daily (American Meat Science Association, 2012). Additionally, the procedure in Colle et al. (2016) was followed with the exception that 3 evaluators scored subjective color.

### Metmyoglobin reducing activity

Nitric oxide metmyoglobin reducing activity (MRA) was determined on d 0 and 4 of retail display for each of the three aging periods following the protocol provided in Section XI of the Meat Color Measurement Guidelines (American Meat Science Association, 2012). The percentage of metmyoglobin (MMb) was calculated using the formulas reported in Section IX of the Meat Color Measurement Guidelines (American Meat Science Association, 2012). MRA was calculated as follows:

$$\text{MRA} = \left[ \frac{\text{Initial \% MMb} - \text{Final \% MMb}}{\text{Initial \% MMb}} \right] \times 100$$

## Oxygen consumption

Oxygen consumption (OC) was determined on Day 0 of retail display for each of the 3 aging periods following the protocol provided in Section XI of the Meat Color Measurement Guidelines (American Meat Science Association, 2012). The percentage of oxymyoglobin (OMb) was calculated using the formulas reported in Section IX of the Meat Color Measurement Guidelines (American Meat Science Association, 2012). Oxygen consumption was calculated as follows:

$$OC = \left[ \frac{\text{Initial \% OMb} - \text{Ending \% OMb}}{\text{Initial \% OMb}} \right] \times 100$$

## Microbial growth

Steaks were swabbed on d 0, 2, and 4 and samples were diluted 1:100 using Lethen broth (3M, St. Paul, MN) before being plated (Colle et al., 2016). Aerobic plates were incubated at 35°C for 48 h before being counted by research personal.

## Lipid oxidation

Thiobarbituric acid reactive substances (TBARS) were analyzed on Days 0, 2, and 4 of retail display following the protocol provided in Section XI, Appendix O of the Meat Color Measurement Guidelines (American Meat Science Association, 2012). Steaks were sampled following the procedure in Colle et al. (2016). Briefly, the end (~1 cm) of the steak was discarded before ~0.5 cm wide, ~2.0 cm long, and ~1.27 cm thick samples were taken from the top half of the steak avoiding the steak edge.

## Cooking

Steaks were cooked on open-hearth broilers (Model BG-16, DeLonghi, Upper Saddle River, NJ) to an internal temperature of 40°C, then turned and cooked to a final internal temperature of 71°C (American Meat Science Association, 2015; Colle et al., 2016). Temperature was monitored with hypodermic temperature probes (Omega Engineering Co., Stamford, CT) coupled with a 12-channel scanning thermocouple thermometer (Digi-Sense, Cole-Parmer Instrument Co., Vernon Hills, IL). Percent cook loss was calculated as follows:

$$\text{Percent } \square \text{cook loss} = \frac{\text{precook } \square \text{weight} - \text{postcook } \square \text{weight}}{\text{precook } \square \text{weight}} \times 100$$

## Warner-Bratzler shear force

Warner-Bratzler shear force was conducted following the procedures in American Meat Science Association (2015) and Colle et al. (2016). After overnight storage at 3°C, 6 cores (1.27-cm diameter) were mechanically removed parallel with the muscle fiber orientation using a drill press-mounted coring device. Shear force was determined by shearing each core (200 mm/min) perpendicular to the muscle fibers using a Warner-Bratzler shear machine (GR Manufacturing, Manhattan, KS).

## Sensory analysis

Steaks designated for consumer acceptability were weighed and exposed to retail display conditions for 2 d, then reweighed, sampled for TBARS analysis, vacuum packaged and frozen at -20°C until completion of all aging periods. One consumer panel was conducted for each muscle following closely the procedure in American Meat Science Association (2015) and Colle et al. (2016). Steaks were thawed overnight at 4°C and subsequently cooked as described above. Samples were placed in insulated containers with hot packs to keep them warm until serving. Panelists were served samples in covered cups, labeled with a random number, and consumed samples in the order and pace that they desired. Panelists received 2 unsalted saltine crackers and a cup of distilled water. A panel of consumers ( $n = 60$  per muscle) evaluated cooked steaks from each aging time for overall acceptability, tenderness, juiciness, and flavor using a 9-point scale (9 = like extremely, extremely tender, extremely juicy, and like flavor extremely, respectively; 1 = dislike extremely, not at all tender, extremely dry, and dislike flavor extremely, respectively). Each consumer also evaluated one cube (1.27cm  $\times$  1.27cm  $\times$  steak thickness) from each of 6 steaks that represented all aging periods and treatments for a muscle.

## Statistical analysis

A split-split-plot with repeated measures was utilized. Aging time was the whole-plot factor, antioxidant treatment was the split-plot factor, and retail display time was the split-split-plot factor. Muscle from a carcass ( $n = 12$ ) served as a block. Data were analyzed using the Mixed Model procedure of the Statistical Analysis System (SAS Inst. Inc., Cary, NC). Muscle and muscle by age served as random variables, day of aging, day of retail display, antioxidant treatment, and the interaction between day of aging, day of retail display, and antioxidant treatment served as fixed variables. Color measurements, aerobic

**Table 1.** Fluid loss, cook time, cook loss, and WBSF of extended aged beef ( $n = 12$ )

| Characteristic              | Day of aging      |                   |                   | SEM  | P-value |
|-----------------------------|-------------------|-------------------|-------------------|------|---------|
|                             | 14                | 28                | 42                |      |         |
| Percent retail fluid loss   |                   |                   |                   |      |         |
| <i>Longissimus lumborum</i> | 0.90              | 0.98              | 0.92              | 0.10 | 0.82    |
| <i>Semimembranosus</i>      | 1.21 <sup>a</sup> | 0.90 <sup>b</sup> | 0.83 <sup>b</sup> | 0.06 | < 0.05  |
| Cook time <sup>1</sup>      |                   |                   |                   |      |         |
| <i>Longissimus lumborum</i> | 22                | 21                | 23                | 2    | 0.67    |
| <i>Semimembranosus</i>      | 30                | 30                | 25                | 2    | 0.12    |
| Percent cook loss           |                   |                   |                   |      |         |
| <i>Longissimus lumborum</i> | 22.7              | 23.1              | 24.7              | 1.1  | 0.40    |
| <i>Semimembranosus</i>      | 28.8              | 27.8              | 25.9              | 1.3  | 0.25    |
| WBSF (kg)                   |                   |                   |                   |      |         |
| <i>Longissimus lumborum</i> | 2.25              | 2.28              | 2.12              | 0.08 | 0.20    |
| <i>Semimembranosus</i>      | 3.16              | 3.16              | 2.73              | 0.15 | 0.11    |

<sup>a,b</sup>Within a row, means without a common superscript differ ( $P < 0.05$ ).

<sup>1</sup>Minutes to 71°.

plate counts, and TBARS values were analyzed as repeated measures with a compound symmetry covariance structure. Aerobic Plate Counts were  $\log_{10}$  transformed prior to analysis. Differences in least squares means (LSM) were compared by the DIFF option. P-values of  $\leq 0.05$  were considered statistically significant and P-values  $\leq 0.10$  were considered trends in the data.

## Results

Antioxidant treatment did not affect LL or SM retail fluid loss ( $P = 0.39$  and  $0.16$ , respectively), cook time ( $P = 0.74$  and  $0.15$ , respectively), cooking loss ( $P = 0.48$  and  $0.23$ , respectively), or WBSF ( $P = 0.71$  and  $0.66$ , respectively) values (data not shown). Additionally, aging period did not affect LL or SM cook time, cooking loss, or WBSF values (Table 1). Furthermore, aging period did not affect percent retail fluid loss in LL but did decrease the percent retail fluid loss ( $P < 0.05$ ) of the SM from d 14 to 28 of aging (Table 1).

The pH values were higher ( $P < 0.05$ ) on d 28 for both the LL and SM compared to the other aging periods. Values for the LL on d 0, 14, 28, and 42 were 5.49, 5.51, 5.65, and 5.50, respectively. Values for the SM on d 1, 14, 28, and 42 were 5.41, 5.41, 5.58, and 5.44.

Aerobic plate counts for the LL ( $P = 0.30$ ) and SM ( $P = 0.17$ ) and lipid oxidation for the LL ( $P = 0.15$ ) and SM ( $P = 0.12$ ) were not affected by the antioxidant treatment (data not shown). However, an aging period by day of retail display interaction ( $P < 0.05$ ) was observed for microbial growth and lipid oxidation for both muscles (Table 2).

**Table 2.** Microbial counts and lipid oxidation of extended aged beef ( $n = 12$ )

| Characteristic                      | Day of display | Day of aging       |                    |                    | SEM  | P-value             |
|-------------------------------------|----------------|--------------------|--------------------|--------------------|------|---------------------|
|                                     |                | 14                 | 28                 | 42                 |      |                     |
| Aerobic plate counts <sup>1,2</sup> |                |                    |                    |                    |      |                     |
| <i>Longissimus lumborum</i>         | 0              | 2.8 <sup>ax</sup>  | 4.8 <sup>by</sup>  | 2.4 <sup>x</sup>   | 0.3  | < 0.05 <sup>4</sup> |
|                                     | 2              | 2.4 <sup>ax</sup>  | 4.2 <sup>bey</sup> | 1.8 <sup>x</sup>   | 0.3  |                     |
|                                     | 4              | 3.6 <sup>bx</sup>  | 4.1 <sup>cx</sup>  | 2.1 <sup>y</sup>   | 0.3  |                     |
| <i>Semimembranosus</i>              | 0              | 1.9 <sup>x</sup>   | 2.6 <sup>y</sup>   | 2.5 <sup>ay</sup>  | 0.2  | < 0.05 <sup>4</sup> |
|                                     | 2              | 1.8 <sup>x</sup>   | 2.8 <sup>y</sup>   | 0.3 <sup>bz</sup>  | 0.2  |                     |
|                                     | 4              | 1.5 <sup>x</sup>   | 3.1 <sup>y</sup>   | 0.8 <sup>bz</sup>  | 0.2  |                     |
| Lipid oxidation <sup>3</sup>        |                |                    |                    |                    |      |                     |
| <i>Longissimus lumborum</i>         | 0              | 0.73               | 0.85 <sup>a</sup>  | 0.78 <sup>a</sup>  | 0.12 | < 0.05 <sup>4</sup> |
|                                     | 2              | 0.75 <sup>x</sup>  | 1.25 <sup>by</sup> | 1.11 <sup>by</sup> | 0.12 |                     |
|                                     | 4              | 0.89 <sup>x</sup>  | 1.40 <sup>by</sup> | 1.16 <sup>bz</sup> | 0.12 |                     |
| <i>Semimembranosus</i>              | 0              | 0.30 <sup>a</sup>  | 0.34 <sup>a</sup>  | 0.25 <sup>a</sup>  | 0.09 | < 0.05 <sup>4</sup> |
|                                     | 2              | 0.29 <sup>ax</sup> | 0.80 <sup>by</sup> | 0.46 <sup>bz</sup> | 0.09 |                     |
|                                     | 4              | 0.42 <sup>bx</sup> | 0.72 <sup>by</sup> | 0.74 <sup>cy</sup> | 0.09 |                     |

<sup>a,b</sup>Within a column, trait, and muscle, means without a common superscript differ ( $P < 0.05$ ); means without <sup>ab</sup> did not differ within a column, trait, and muscle.

<sup>x-z</sup>Within a row, means without a common superscript differ ( $P < 0.05$ ); means without <sup>x-z</sup> did not differ within a row.

<sup>1</sup> $\log_{10}$  colony-forming units/cm<sup>2</sup>.

<sup>2</sup>Samples were diluted 1:100 with Lethen Broth and plates were estimated following the 3M Interpretation Guide.

<sup>3</sup>mg malondialdehyde/kg meat.

<sup>4</sup>Aging period by day of retail display interaction.

Antioxidant treated LL steaks were lighter (higher L\*), less red (lower a\*), and less yellow (lower b\*) than control steaks ( $P < 0.05$ ; Table 3). However, objective color measurements did not differ between SM treated and control steaks (data not shown). Longer aging periods and retail display times resulted in darker, less-red, and less-yellow LL and SM steaks ( $P < 0.5$ ).

An antioxidant treatment by day of retail display interaction ( $P < 0.05$ ) was observed for LL amount of browning (Table 4). Control steaks were more brown than antioxidant treated steaks on d 4 of retail display. Additionally, antioxidant treated steaks tended to have brighter oxygenated lean color than control steaks for the LL ( $P = 0.069$ ) and SM ( $P = 0.061$ ; Table 5). All other subjective color measurements (discoloration, surface discoloration, and color uniformity) did not differ between antioxidant treated and control steaks (data not shown). An aging period by day of retail display interaction ( $P < 0.01$ ) was observed for all subjective color measurements for both muscles (data not shown). Steak color became less desirable with longer aging periods and retail display times.

An antioxidant treatment by aging period interaction ( $P < 0.05$ ) was observed for LL MRA (Fig. 1).

**Table 3.** *Longissimus lumborum* objective color across all aging periods and retail display times ( $n = 12$ )

| Color measure | Antioxidant <sup>1</sup> | Control            | SEM  | <i>P</i> -value |
|---------------|--------------------------|--------------------|------|-----------------|
| L*            | 39.48 <sup>a</sup>       | 38.48 <sup>b</sup> | 0.87 | < 0.05          |
| a*            | 30.25 <sup>a</sup>       | 30.79 <sup>b</sup> | 0.24 | < 0.05          |
| b*            | 25.86 <sup>a</sup>       | 26.37 <sup>b</sup> | 0.24 | < 0.05          |

<sup>a,b</sup>Within a row, means without a common superscript differ ( $P < 0.05$ ).

<sup>1</sup>0.05% ascorbic acid + 0.1% rosemary extract.

**Table 4.** *Longissimus lumborum* amount of browning<sup>1</sup> across all aging periods ( $n = 12$ )

| Day of display | Control          | Antioxidant <sup>2</sup> | SEM | <i>P</i> -value     |
|----------------|------------------|--------------------------|-----|---------------------|
| 0              | 1.0 <sup>a</sup> | 1.0 <sup>a</sup>         | 0.2 | < 0.05 <sup>3</sup> |
| 1              | 1.2 <sup>a</sup> | 1.2 <sup>a</sup>         | 0.2 |                     |
| 2              | 2.3 <sup>b</sup> | 2.4 <sup>b</sup>         | 0.2 |                     |
| 3              | 2.7 <sup>c</sup> | 2.8 <sup>c</sup>         | 0.2 |                     |
| 4              | 4.5 <sup>d</sup> | 3.9 <sup>c</sup>         | 0.2 |                     |

<sup>a-c</sup>Within the table, means without a common superscript differ ( $P < 0.05$ ).

<sup>1</sup>1 = no evidence of browning, 2 = dull, 3 = grayish, 4 = brownish-gray, 5 = brown, 6 = dark brown.

<sup>2</sup>0.05% ascorbic acid + 0.1% rosemary extract.

<sup>3</sup>Antioxidant treatment by day of retail display interaction.

MRA of treated steaks was numerically less than control steaks on d 14 of aging, while after 28 and 42 d of aging treated steaks had a higher MRA than control steaks. Additionally, a day of retail display by aging period interaction ( $P < 0.001$ ) was observed for LL MRA (Fig. 1). There was a large decrease in MRA

**Table 5.** Oxygenated lean color<sup>1</sup> across all aging and retail display times ( $n = 12$ )

| Muscle                      | Antioxidant <sup>2</sup> | Control | SEM  | <i>P</i> -value |
|-----------------------------|--------------------------|---------|------|-----------------|
| <i>Longissimus lumborum</i> | 3.64                     | 3.76    | 0.17 | 0.069           |
| <i>Semimembranosus</i>      | 4.43                     | 4.52    | 0.23 | 0.061           |

<sup>1</sup>1 = extremely bright cherry-red, 2 = bright cherry-red, 3 = moderately bright cherry-red, 4 = slightly bright cherry-red, 5 = slightly dark cherry-red, 6 = moderately dark red, 7 = dark red, 8 = Extremely dark red.

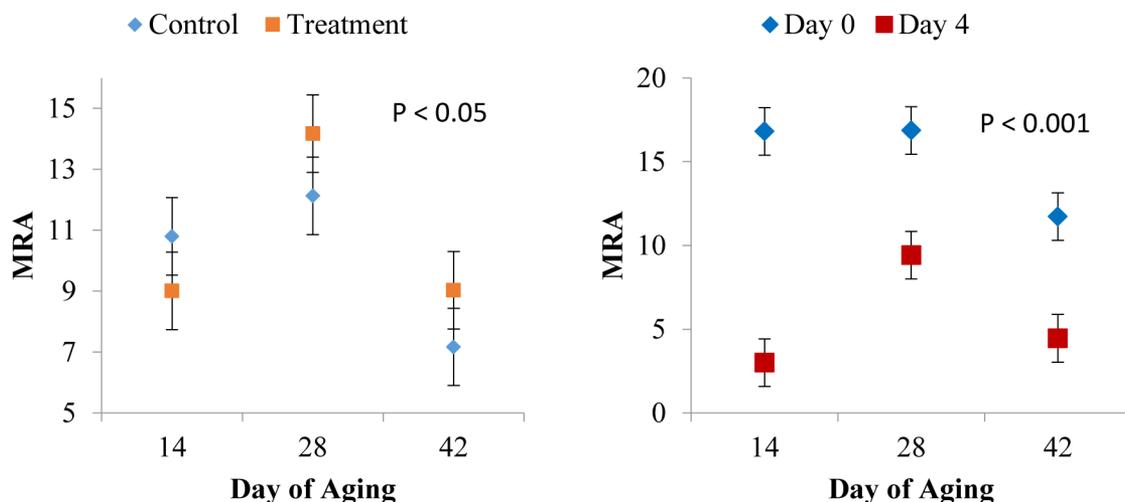
<sup>2</sup>0.05% ascorbic acid + 0.1% rosemary extract.

from d 0 to d 4 of retail display of the d 14 aging period with similar but smaller decreases for the longer aging periods.

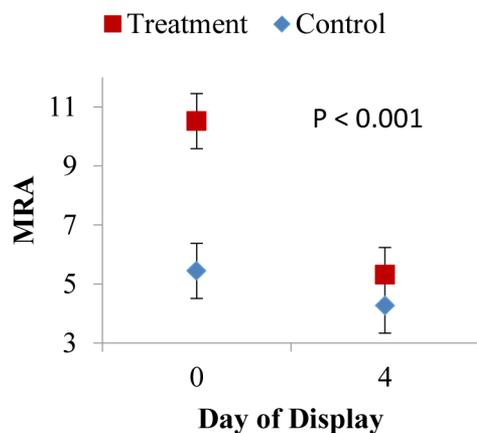
An antioxidant treatment by day of retail display interaction ( $P < 0.001$ ) was observed for SM MRA (Fig. 2). Antioxidant treated SM steaks had higher MRA than control steaks on d 0 of retail display, while no difference was observed on d 4 of retail display. Furthermore, SM MRA was higher ( $P < 0.001$ ) following 28 d of aging than the other 2 aging periods.

Oxygen consumption was not affected by antioxidant treatment for either muscle (data not shown). However, LL OC decreased ( $P < 0.05$ ) from 18.0% on d 14 of aging to 12.3% on d 28, and finally 8.0% on d 42 of aging. Additionally, SM OC remained constant ( $P = 0.30$ ) from d 14 to 28 of aging (9.9 and 11.1%, respectively) and subsequently decreased ( $P < 0.05$ ) to 5.2% on d 42 of aging.

Demographics of consumer panelists are shown in Table 6. An aging period by antioxidant treatment inter-



**Figure 1.** Metmyoglobin reducing activity (MRA) values for antioxidant treatment × day of aging (left) and day of retail display × day of aging (right) for the *longissimus lumborum* ( $n = 12$ ). Steaks assigned to the antioxidant treatment group were sprayed (~2ml) with the antioxidant (0.05% AA + 0.1% RE) solution, while control steaks were not sprayed. Steaks were overwrapped with an oxygen permeable PVC film, and displayed in a glass-fronted retail display case at 3°C for 4 d. MRA was determined on d 0 and 4 of retail display for each of the 3 aging periods with the following equation:  $MRA = [(Initial \% \text{ metmyoglobin} - Final \% \text{ metmyoglobin}) \div Initial \% \text{ metmyoglobin}] \times 100$ . Values are shown as least square means ± SE.



**Figure 2.** Metmyoglobin reducing activity (MRA) values for antioxidant treatment by day of retail display for the *semimembranosus* ( $n = 12$ ). Steaks assigned to the antioxidant treatment group were sprayed (~2ml) with the antioxidant (0.05% AA + 0.1% RE) solution, while control steaks were not sprayed. Steaks were overwrapped with an oxygen permeable PVC film, and displayed in a glass-fronted retail display case at 3°C for 4 d. MRA was determined on d 0 and 4 of retail display for each of the 3 aging periods with the following equation:  $MRA = [(Initial \% \text{ metmyoglobin} - Final \% \text{ metmyoglobin}) \div Initial \% \text{ metmyoglobin}] \times 100$ . Values are shown as least square means  $\pm$  SE.

action ( $P < 0.05$ ) was observed for consumer panel SM sensory tenderness and juiciness (Table 7). Additionally, there was an antioxidant treatment by aging period interaction trend ( $P = 0.052$ ) for SM acceptability and

**Table 6.** Demographics of consumer panelists ( $n = 60/\text{panel}$ )

| Demographic                      | <i>Longissimus lumborum</i> |      | <i>Semimembranosus</i> |      |
|----------------------------------|-----------------------------|------|------------------------|------|
|                                  | <i>n</i>                    | %    | <i>n</i>               | %    |
| <b>Age</b>                       |                             |      |                        |      |
| 18–19                            | 2                           | 3.4  | 2                      | 3.4  |
| 20–29                            | 27                          | 45.8 | 36                     | 61.1 |
| 30–39                            | 10                          | 16.8 | 8                      | 13.5 |
| 40–49                            | 7                           | 11.8 | 4                      | 6.8  |
| 50+                              | 13                          | 22.0 | 9                      | 15.2 |
| <b>Gender</b>                    |                             |      |                        |      |
| Male                             | 26                          | 44.1 | 25                     | 41.0 |
| Female                           | 33                          | 55.9 | 36                     | 59.0 |
| <b>Beef meals/wk<sup>1</sup></b> |                             |      |                        |      |
| 0 to 1                           | 10                          | 16.7 | 10                     | 16.7 |
| 2 to 4                           | 34                          | 56.7 | 39                     | 65.0 |
| 5 to 7                           | 14                          | 23.3 | 10                     | 16.7 |
| 8+                               | 2                           | 3.3  | 1                      | 1.6  |
| <b>Most consumed<sup>2</sup></b> |                             |      |                        |      |
| Ground                           | 40                          | 61.5 | 38                     | 60.3 |
| Roast                            | 5                           | 7.7  | 3                      | 4.8  |
| Steak                            | 19                          | 29.3 | 22                     | 34.9 |
| Other                            | 1                           | 1.5  | 0                      | 0.0  |

<sup>1</sup>Please indicate the number of meals a week in which you consume beef: 0–1, 2–4, 5–7, or 8+.

<sup>2</sup>Please indicate the form in which you most commonly consume beef: ground, roast, steak, or other.

an aging period trend ( $P = 0.092$ ) for SM flavor (Table 7). The antioxidant treatment did not affect LL acceptability, flavor, tenderness, or juiciness (data not shown). Furthermore, aging period did not affect LL acceptability, flavor, tenderness, or juiciness (Table 7).

## Discussion

The present study evaluated the effects of topical application of a 0.05% ascorbic acid + 0.1% rosemary extract solution to steaks aged for 14, 28, or 42 d prior to retail display. Ascorbic acid and rosemary extract have been shown by several studies to improve various shelf-life attributes of beef (Wheeler et al., 1996; Sánchez-Escalante et al., 2001; Djenane et al., 2003; Ahn et al., 2007). The current study is the first to evaluate the effect of antioxidants on improving the shelf-life of steaks aged longer than 28 d. Increasing the shelf-life of beef aged longer than 28 d will allow retail stores more flexibility to market the product at the normal price vs. having to mark the product as reduced for quick sale (Suman et al., 2014). In some cases, this mark down

**Table 7.** Sensory analysis by consumer panelists ( $n = 60/\text{panel}$ )

| Sensory Trait               | Day of aging      |                   |                   | SEM | <i>P</i> -value     |
|-----------------------------|-------------------|-------------------|-------------------|-----|---------------------|
|                             | 14                | 28                | 42                |     |                     |
| <i>Longissimus lumborum</i> |                   |                   |                   |     |                     |
| Acceptability <sup>1</sup>  | 6.1               | 6.2               | 5.8               | 0.2 | 0.16                |
| Tenderness                  | 5.9               | 6.2               | 6.4               | 0.2 | 0.27                |
| Juiciness                   | 5.3               | 5.6               | 5.5               | 0.2 | 0.46                |
| Flavor                      | 6.0               | 6.2               | 5.5               | 0.3 | 0.13                |
| <i>Semimembranosus</i>      |                   |                   |                   |     |                     |
| Acceptability               |                   |                   |                   |     | 0.052 <sup>3</sup>  |
| Control                     | 5.0               | 5.5               | 5.1               | 0.3 |                     |
| Treatment <sup>2</sup>      | 5.6               | 5.1               | 4.9               | 0.3 |                     |
| Tenderness                  |                   |                   |                   |     | < 0.05 <sup>3</sup> |
| Control                     | 4.6 <sup>y</sup>  | 5.5 <sup>az</sup> | 5.4 <sup>yz</sup> | 0.3 |                     |
| Treatment                   | 5.2               | 4.6 <sup>b</sup>  | 5.0               | 0.3 |                     |
| Juiciness                   |                   |                   |                   |     | < 0.05 <sup>3</sup> |
| Control                     | 4.4 <sup>by</sup> | 5.3 <sup>az</sup> | 5.3 <sup>az</sup> | 0.3 |                     |
| Treatment                   | 5.3 <sup>a</sup>  | 4.6 <sup>b</sup>  | 4.6 <sup>b</sup>  | 0.3 |                     |
| Flavor                      | 5.4               | 5.4               | 4.7               | 0.2 | 0.09                |

<sup>a,b</sup>Within a column and trait, means without a common superscript differ ( $P < 0.05$ ); means without <sup>ab</sup> did not differ within a column and trait.

<sup>y,z</sup>Within a row, means without a common superscript differ ( $P < 0.05$ ); means without <sup>yz</sup> did not differ within a row.

<sup>1</sup>Scale, 9 = like extremely, extremely tender, extremely juicy, and like flavor extremely, respectively; 1 = dislike extremely, not at all tender, extremely dry, and dislike flavor extremely, respectively.

<sup>2</sup>0.05% ascorbic acid + 0.1% rosemary extract.

<sup>3</sup>Antioxidant treatment by aging period interaction.

in price likely occurs within 24 h due to the poor color stability of extended aged product (Colle et al., 2016).

In the current study, antioxidant treatment resulted in small subjective color improvements of LL steaks. The reduced browning of LL treated steaks compared to control steaks on d 4 of retail display was significant but is likely too small to be detected by consumers. Additionally, a majority of steaks in the retail case are likely sold prior to d 4. *Longissimus lumborum* treated steaks had higher L\* values and tended to have brighter oxygenated lean color. SM treated steaks also tended to have brighter oxygenated lean color. The added water on the surface of the steak from the antioxidant treatment may have led to increased light reflection and therefore a greater L\* value and brighter oxygenated lean color. The tendency of a brighter oxygenated lean color in both the LL and SM as well as the higher L\* value for the LL may lead to consumers perceiving this product as being fresher appearing than a darker product. In the current study, the antioxidant treatment decreased a\* values in LL. In contrast, at the same levels of antioxidants used in this experiment, Djenane et al. (2003) observed that antioxidant treated LD steaks significantly delayed the decrease in a\* values (redness) compared to untreated steaks. However, these authors used 0.05% ascorbic acid and 0.1% rosemary extract in combination with 1.5% lactic acid and then packaged the meat in modified atmospheric packaging. Therefore, the antioxidant treatment likely had a synergistic affect with the lactic acid or packaging to improve redness during shelf-life. The color differences noted in the present study may be too small for the consumer to detect.

The increased MRA in SM antioxidant treated steaks was intriguing since higher MRA values generally indicate improved color stability even though a color improvement was not observed in the SM. Additionally, improved color stability should correspond with reduced lipid oxidation since myoglobin oxidation and lipid oxidation are closely related (Faustman and Cassens, 1990; Faustman et al., 2010). The antioxidant treatment in the present study did not affect lipid oxidation of either muscle. This observation may be because the antioxidant was applied topically. Furthermore, the antioxidant treatment did not affect retail fluid loss microbial growth, LL sensory attributes or SM acceptability and flavor. Having panelists sample multiple cubes from the same antioxidant treatment as well as increasing the number of panelists would likely have resulted in more consistent and explainable results for SM tenderness and juiciness. One reason for the minimal significant differences could be that at low levels, ascorbic acid acts as a pro-oxidant (Buettner and Jurkiewicz, 1996; Wheeler et al., 1996). Ascorbic acid can act as a

pro-oxidant by reducing ferric iron to ferrous iron, with ferrous iron being a strong pro-oxidant (Buettner and Jurkiewicz, 1996). Higher levels of the antioxidants and/or a different application method such as dipping the steak in the antioxidant solution may result in larger improvements in color stability.

Aging time influenced several factors measured. Both MRA and OC generally decreased with longer aging periods and MRA decreased with retail display time. This was expected because metmyoglobin reductase decreases over time due to a lack of NADH being produced from the citric acid cycle (Sammel et al., 2002). English et al. (2016) also reported that MRA and OC decreased with increased aging time. McKenna et al. (2005) similarly noted that nitric oxide MRA decreased over retail display time in beef muscles. The large decrease in MRA from d 0 to d 4 of retail display in LL steaks of the d 14 aging period may be related to the higher levels of OC at this time point. Measuring pH on d 4 of retail display may have been helpful in interpreting these results.

Longer aging periods resulted in reduced SM retail fluid loss compared to the d 14 aging period. Cannon et al. (1996) as well as Colle et al. (2016) previously reported that drip loss and retail fluid loss, respectively, decrease with longer aging period's likely do to the fact that more moisture had been lost during the longer aging periods. An aging effect on LL sensory tenderness was not expected since a majority of USDA Choice LD aging response is completed by 14 d of aging (Gruber et al., 2006).

## Conclusions

A strategy to improve the shelf-life of extended aged beef is needed to provide retailers with an option to increase the time this product is in the retail case before it is reduced for quick sale. Although previous studies have shown substantial benefits of antioxidants on beef shelf-life, the current experiment found only limited improvements in LL color attributes as well as SM flavor in response to the ascorbic acid and rosemary extract treatment. Additional research to find a more effective application strategy of these and other antioxidants is needed to develop a simple but effective strategy to improve the shelf-life of extended aged beef.

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