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Color and lipid stability of dry aged beef during retail display

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ABSTRACT

The objective of this study was to determine color and lipid stability of steaks from dry-aged beef loins over 7 d of retail display (RD). Sixteen boneless strip loins were assigned to one of four treatments: wet-aging, dry aging at 50% relative humidity (RH), dry aging at 70% RH, or dry aging at 85% RH and aged for 42 days at 2 °C. Dry aging of beef resulted in decreased lightness (L*), redness (a*), and yellowness (b*) values and increased lipid oxidation compared to wet-aged counterparts ($P < 0.05$). Dry-aged steaks had greater discoloration ($P < 0.05$) than wet-aged steaks from d 4 to d 7 of RD. Results suggest that under prolonged RD, dry aging of beef has the potential to reduce color and lipid stability compared to wet aging and thus reduce display life. Color and lipid stability were not affected by RH during dry aging.

1. Introduction

There has been increased interest in merchandising dry-aged steaks at the retail consumer level due to its unique flavor (Campbell, Hunt, Levis, & Chambers IV, 2001; Warren & Kastner, 1992). Although extended dry aging contributes to flavor development, evidence of its effects on color and lipid stability under retail display conditions is incomplete. Previous studies have reported that extended wet aging in vacuum packages negatively impacts color and lipid oxidative stability, resulting in accelerated rates of surface discoloration and oxidized off-flavor development in meat (Ribeiro et al., 2018).

Dry-aged beef is darker and less red compared with wet-aged beef due to lower moisture content and surface drying after aging resulting in less light reflection (Dikeman, Obuz, Gök, Akkaya, & Stroda, 2013; Kim & Hunt, 2011; Kim, Kemp, & Samuelsson, 2016). Discoloration caused by accumulation of metmyoglobin due to the oxidation of myoglobin and darkening due to surface dehydration may result in economic losses since meat purchasing decisions at the retail level are mainly influenced by color (Mancini & Hunt, 2005; Smith, Belk, Sofos, Tatum, & Williams, 2000). Evidence for an interaction between the processes of myoglobin oxidation and lipid oxidation in meat has been demonstrated by a number of studies. The biochemical reactions that occur in each of these processes generate products that can further accelerate oxidation in a reciprocal manner (Faustman, Sun, Mancini, & Suman, 2010). While color is the first criterion used by the consumer when deciding on a meat purchase, lipid oxidation is a major cause of deterioration in meat quality. There are several factors that can promote lipid oxidation. Substrates necessary for this deteriorative reaction include fat content, fatty acid composition, metal ions that accelerate oxidation (e.g., iron), and oxygen (Faustman et al., 2010). Extended aging periods accelerate oxidized off-flavor development in meat, when repackaged and displayed under retail light conditions (Ribeiro et al., 2018). Under normal conditions, consumers typically perceive oxidized flavors in a negative way (Campo et al., 2006). The nature of dry aging exposes beef to oxygen and increases in oxidized compounds occur. Studies focused on the direct comparison of dry aging to wet aging color have not been the subject of much scientific research. Further understanding of the influence of the dry aging process on meat color and lipid stability is needed to ensure dry-aged beef products can be merchandised without adverse impacts on retail

Abbreviations: C, Celsius; D, day(s); g, Gram(s); kg, Kilogram(s); MRA, Metmyoglobin reducing activity; μL, Microliter(s); mg, Milligram(s); min, Minute(s); NADH, Nicotinamide adenine dinucleotide hydride; nm, Nanometer(s); RD, Retail display; RH, Relative humidity; RH50, Dry aging at 50% relative humidity; RH70, Dry aging at 70% relative humidity; RH85, Dry aging at 85% relative humidity; RPM, Revolutions per minute; s, Second(s); SEM, Standard error of the mean; TBA, Thiobarbituric acid; TBARS, Thiobarbituric acid reactive substances; TCA, Trichloroacetic acid; WET, Wet aging

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display life. Therefore, this study aimed to determine color and lipid stability of steaks from dry-aged beef loins over 7 d of retail display (RD).

2. Materials and methods

2.1. Sample collection

All animals used in this study were slaughtered humanely, under USDA guidelines. Sixteen USDA low Choice strip loins [Institutional Meat Purchase Specifications number 180 (USDA, 1996)] were collected at a commercial beef harvest facility, vacuum-packaged, and transported to the University of Nebraska meat laboratory where they were immediately removed from the vacuum packages. Boneless loins were assigned to one of the four aging treatments: vacuum (WET), dry aging at 50% relative humidity (RH; RH50), dry aging at 70% RH (RH70), or dry aging at 85% RH (RH85). Strip loins were placed in each assigned dry aging chamber (1 loin per chamber) and aged for 42 days at 2 °C. The dry aging chambers (86 cm length x 47.6 cm width x 33 cm height) are capable of measuring and precisely controlling relative humidity (± 1%) and air velocity (± 50 RPM). The chambers monitor temperature (± 0.5 °C) and have built-in sensors that can continuously monitor weight loss (± 5 g). All measured data were saved on the connected computer every 15 s. A full description of this dry aging system called Agenator has been previously published by Lau, Ribeiro, Subbiah, and Calkins (2019). After aging, loins were subject to retail display (RD) for 7 d. Loins assigned to wet aging were individually vacuum packaged and kept intact over 42 days at 2 °C in the same cooler where the dry aged chambers were located.

Dry-aged loins were trimmed of dried surface and subcutaneous fat, and fabricated anterior to posterior into 3 steaks: One 2.54 cm-thick steak for objective color and subjective discoloration with 7 d RD, and two 1.27 cm-thick steaks for lipid oxidation (one steak for lipid oxidation at 0 d RD and one steak was split in half for lipid oxidation after 4 and 7 d RD).

After fabrication, steaks assigned to lipid oxidation at 0 d of RD were vacuum packaged and frozen at −80 °C. Steaks assigned to 4 and 7 d of RD were placed on foam trays (21.6 × 15.9 × 2.1 cm, Styro-Tech, Denver, CO), overwrapped with oxygen permeable film (Prime Source PSM 18 #75003815, Bunzl Processors Division, North Kansas City, MO; oxygen transmission rate = 2.25 mL/cm^2/24 h at 23 °C and 0% relative humidity; water vapor transfer rate = 496 g/m^2/24 h at 37.8 °C and 90% relative humidity), and placed under RD conditions for 7 d (continuous white fluorescence lighting at 1000 to 1800 lx; F32T8/TL741, 700 series, 32 W, Philips, USA) at 2 °C. After 4 or 7 d of RD steaks were vacuum packaged and stored at −80 °C. Steaks were randomly rotated daily to minimize any possible location effects within the display.

2.2. Objective color

Objective color was measured using the L*, a*, b* scales using a Minolta CR-400 colorimeter (Illuminant D65, 8 mm diameter aperture, 2° standard observer angle; Minolta, Osaka, Japan). The calibration process was done on a daily basis using a white ceramic tile provided by the manufacturer (Calibration Plate, Serial No. 14933058, Konica Minolta, Japan) and the D65 settings were set as follows: Y = 93.13, x = 0.3164 and y = 0.3330. Six measurements were made per steak through the overwrap film once a day at a standardized time from d 0 to 7 of RD. Readings were averaged by steak for statistical analysis.

2.3. Subjective discoloration

Six trained panelists evaluated surface discoloration daily during the 7 d of RD according to the procedure of Senaratne-Lenagala (2012). A reference guide of ten steak images ranging from 0% to 100% surface discoloration with increments of 10% were provided to panelists to ensure consistent evaluations. A percentage continuous scale was used where 0% meant no discoloration and 100% meant complete surface discoloration.

2.4. Lipid oxidation

Steaks used for lipid oxidation were removed from the freezer, partially thawed at room temperature for 60 min, cut by hand into small cubes, frozen in liquid nitrogen, powdered in a metal cup blender (Model 51BL32, Waring Commercial, Torrington, CT), and stored at −80 °C until further analysis. Thiobarbituric acid reactive substance values (TBARS) were measured at 0, 4 and 7 d of RD according to the procedure of Ahn et al. (1998). Five grams of powdered meat from each steak were blended with 1 mL of butylated hydroxyanisole solution (10%) and 14 mL of distilled water. Samples were homogenized using a Polytron (Kinmatica AG, Lucern, Sui) for 15 s and centrifuged (2000 x *g* for 5 min). After centrifugation, 1 mL of supernatant was collected, mixed with 2 mL of thiobarbituric acid/trichloroacetic acid (TBA/TCA) solution (15% TCA and 20 mM TBA in deionized distiller water), and placed in a water bath at 70 °C for 30 min. Samples were cooled for 10 min in a water bath at 20 °C and centrifuged (2000 x *g* for 15 min). Two hundred microliters of supernatant were transferred to 96-well plates in duplicate. Absorbance was measured at 540 nm using a microplate spectrophotometer (Model Epoch, Biotek, Winooski, VT). Results were expressed in mg of malonaldehyde per kg of tissue.

2.5. Statistical analysis

Objective color data were analyzed as a split-plot repeated measures design with aging treatment as the whole-plot and RD time as the repeated measures, and chambers were considered a random effect. Subjective color data were analyzed as a split-plot repeated measures design with aging treatment as the whole-plot, steak as the split-plot, and RD time as the repeated measures. Color panelists were considered a random effect when analyzing subjective discoloration. The TBARS data were analyzed as a split-plot design with aging treatment as the whole-plot, and RD time as the split-plot. In this study, chamber (loin) was considered the experimental unit. Interactions between fixed effects were tested and when significant the interactions were reported. When the interactions were not significant, the main effects of each variable were reported. Data were analyzed using the PROC GLIMMIX procedure of SAS. All means were separated with the LS MEANS and PDIFF or SLICEDIFF functions with $\alpha = 0.05$.

3. Results and discussion

3.1. Instrumental color

For all three color readings, a RD effect was found ($P < 0.001$). In general, a* and b* values decreased as RD time increased, regardless of the aging treatment. The L* values were greater at day 0 RD in comparison to all other days of RD, regardless of aging treatment (Table 1). However, no differences in L* values among days were found during 1 to 7 d of RD ($P > 0.05$). For a* values, differences were found among all days as shown in Table 1. The b* values were greater at day 0 and 1 RD, followed by day 2 and 3, while no significant differences were observed on day 4 to 7 of retail display (Table 1).

In addition to a retail display effect, aging treatment also had an effect on all three color readings. Wet-aged steaks had higher L* ($P = 0.01$), a* ($P = 0.03$), and b* values ($P < 0.001$) than any dry-aged treatment (Table 2). No differences in L*, a*, and b* values among dry-aged treatments were found ($P < 0.05$).

Similar results were also observed in other studies, where dry-aged beef steaks were darker and less red compared with wet-aged steaks (Dikeman et al., 2013; Kim et al., 2016) due to moisture loss (Kim &

Table 1
Objective lightness (L*), redness (a*) and yellowness (b*) values of strip loin steaks (*Longissimus lumborum)* wet aged or dry aged for 42 days at 50, 70 or 85% relative humidity through 7 days of retail display.

Days on retail display	L*	a*	b*
0	46.32^{a}	17.94^{a}	7.93^{a}
1	44.97^{b}	16.99^{b}	7.70^{a}
2	44.98^{b}	15.46^{c}	6.97^{b}
3	43.94^{b}	14.35^{d}	6.83^{b}
4	44.01^{b}	13.36^{e}	6.43^{c}
5	44.07^{b}	12.12^{f}	6.35^{c}
6	44.23^{b}	10.82^{g}	6.30^{c}
7	43.98^{b}	9.28^{h}	6.32^{c}
SEM	0.6844	0.6516	0.1560
P-value	< 0.001	< 0.001	< 0.001

a-h Means in the same column with different superscripts differ ($P < 0.001$).

Table 2
Color measurements and lipid oxidation value (TBARS; mg malonaldehyde /kg of meat) of strip loins steaks (*Longissimus lumborum*) wet aged or dry aged for 42 days at 50, 70 or 85% relative humidity (RH).

Quality trait	Treatments					
	WET	RH50%	RH70%	RH85%	SEM	*P*-value
L*	46.40^{a}	43.64^{b}	43.74^{b}	43.87^{b}	0.790	0.01
a*	16.79^{a}	12.51^{b}	12.87^{b}	12.15^{b}	1.599	0.03
b*	8.22^{a}	6.35^{b}	6.44^{b}	6.42^{b}	0.306	< 0.001
TBARS	2.03^{a}	2.97^{b}	2.81^{b}	3.17^{b}	0.355	0.03

a,b Means in the same row with different superscripts differ ($P < 0.05$).

Hunt, 2011). The darker color in the dry-aged loins compared to the wet-aged loins may be explained by less light reflection associated with lower moisture content.

Detection of color by the human eye is influenced by light reflection. When light strikes meat, it can be absorbed, reflected, or scattered. Light must reflect off the object being viewed and return to the eye in order to be detected. The reflected light is perceived by the eye, captured and transmitted to the brain, where color is interpreted. Therefore, the wavelengths of light that are absorbed by the meat are not perceptible to the eye (AMSA, 2012). Dry aging results in shrinkage during cooler storage as moisture is lost over time. Both shrinkage in height and ribeye area have been observed (Calkins & Ribeiro, 2019). Shrinkage of myofibrils increases the space available for light absorption, thus decreasing the light scattering power of the meat and contributing to darkening of color intensity.

Holman, van de Ven, Mao, Coombs, and Hopkins (2017) reported the relationship between a* and consumer acceptance of beef color. Beef color was considered acceptable (with 95% acceptance) when a* values were equal to or above 14.5. In this study, the days required for each sample to reach an a* value of 14.5 was calculated by interpolating between the two days with values above and below 14.5. The 14.5 color threshold value was met by steaks from the dry aging treatments RH50, RH70, and RH85 after 2.4, 2.7, and 2.0 days of RD, respectively. Steaks from the WET group were statistically different from steaks of dry aging groups and reached the color threshold after 5.2 days of RD ($P \leq 0.05$; SEM = 1.11). A similar pattern was found in our discoloration data using the 20% surface discoloration threshold. Perhaps, a* values can be used as an objective and practical tool by meat retailers to predict meat discoloration and accurately predict color display life.

3.2. Subjective color (discoloration)

A 2-way interaction between treatment and RD for discoloration was observed ($P = 0.03$; Fig. 1). No differences were found among treatments over the first 2 d of RD ($P > 0.05$). Samples began to diverge on day 3 of RD. Dry-aged steaks had greater discoloration scores ($P < 0.05$) than wet-aged steaks from day 4 onward. No differences in discoloration scores among dry-aged treatments were found.

A 50% decline in purchasing decisions with 20% surface discoloration on RD beef has been reported by Hood and Riordan (1973). Even at lower levels of discoloration, consumers begin to discriminate against discolored meat and will select non-discolored products if both packages are viewed in retail display. According to Smith et al. (2000) nearly 15% of retail beef is discounted in price due to surface discoloration, which corresponds to annual revenue losses of $1 billion.

The same strategy used to calculate the days required for each sample to reach an a* value of 14.5 was used to calculate the days required for each sample to reach 20% discoloration. If the value of 20% was never reached then a value of 7.0 d was used. The 20% discoloration threshold was met by steaks from the dry aging treatments RH50, RH70, and RH85 after 4.1, 3.9, and 4.7 days of RD, respectively. Steaks from the WET group reached the color threshold after 6.1 days of RD ($P = 0.06$; SEM = 0.55). The faster discoloration observed for dry-aged steaks when compared to wet-aged steaks could be attributed to the extent of exposure to oxygen and depletion of reducing compounds due to the number of times the meat went through the color cycle.

Discoloration is caused by an accumulation of metmyoglobin on the meat surface due to the oxidation of myoglobin. Muscle ability to convert metmyoglobin (ferric state) to reduced ferrous state through metmyoglobin reducing activity (MRA) is limited and is continually depleted as time postmortem progresses (Mancini & Hunt, 2005). Reduction of metmyoglobin is crucial to meat color life and greatly depends on muscle's oxygen scavenging enzymes, reducing enzyme systems, and the nicotinamide adenine dinucleotide hydride (NADH) pool (Mancini & Hunt, 2005; Renerre, 1990). Once NADH is depleted, MRA is limited and accumulation of metmyoglobin on the meat surface occurs (Kim & Hunt, 2011).

3.3. Lipid oxidation

There was a RD effect on TBARS values ($P < 0.001$). Greater TBARS values were seen as RD progressed from d 0 to d 4 and d 7, regardless the aging treatment (Table 3). A treatment effect was observed for lipid oxidation ($P = 0.03$). Dry-aged steaks had higher TBARS values than wet-aged steaks. No differences in TBARS values among dry aging treatments were found (Table 3).

Lipid oxidation is a three-step chemical process responsible for deterioration of meat (Descalzo et al., 2005; Pradhan, Rhee, & Hernández, 2000). The first step, initiation, begins with the removal of hydrogen from a carbon chain of a fatty acid (Morrissey, Sheehy, Galvin, Kerry, & Buckley, 1998). After initiation, a chain reaction is triggered where the free radicals react with oxygen to form peroxy radicals, which then react with unsaturated lipids, forming hydroperoxides. This step is known as propagation (Wong, 1989). Finally, in the termination step, radicals react among one other, resulting in radical combinations ultimately terminating in non-radical products (Wong, 1989).

In live muscle, enzymes are capable of eliminating free radicals formed during oxidation through the formation of water (Morrissey et al., 1998). However, after slaughter, the balance of prooxidants and antioxidants of living muscle is disrupted, favoring oxidation (Gray, Gomaa, & Buckley, 1996; Morrissey et al., 1998). Consequently, muscle cells become damaged and overall meat quality can be affected (Kanner, 1994). The larger detriment of lipid oxidation is perceived in meat color and flavor (Greene, 1969). Lipid and myoglobin oxidation in meat often appear to be linked and the oxidation of one leads to the formation of chemical species that can exacerbate oxidation of the other (Faustman et al., 2010). This is in agreement with our study, where greater TBARS values and discoloration scores were observed for dry-aged treatments in comparison with wet-aged counterparts.

The rate of lipid oxidation depends on several conditions including

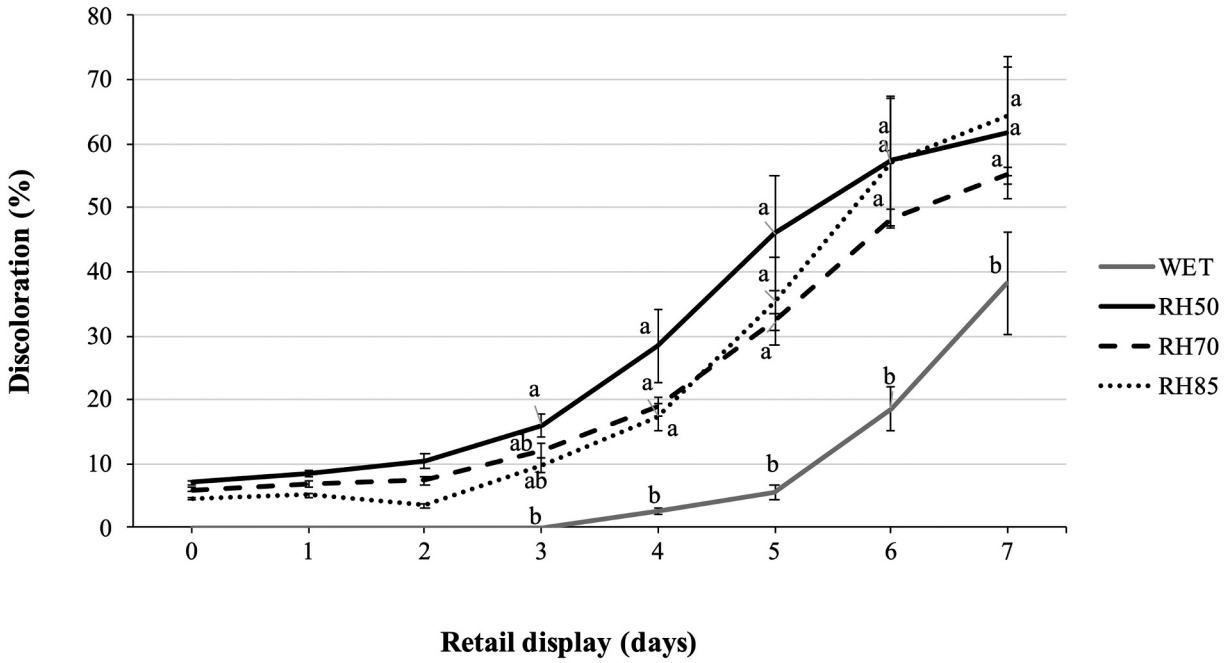

Fig. 1. Discoloration scores (%) of strip loins steaks wet or dry aged for 42 days at 50, 70 or 85% relative humidity (RH) through 7 days of retail display. [a,b] Means in the day with different superscripts differ ($P < 0.05$).

Table 3
Lipid oxidation value (TBARS; mg malonaldehyde /kg of meat) of strip loin steaks (*Longissimus lumborum)* wet or dry aged for 42 days at 50, 70 or 85% relative humidity with 0, 4 and 7 days retail display.

Days on retail display	TBARS	SEM	*P*-value
0	1.01[a]	0.39	< 0.001
4	2.22[b]		
7	3.53[c]		

a-c Means in the same column with different superscripts differ ($P < 0.001$).

length of storage and packaging. The importance of packaging on lipid oxidation has been well described in the literature. Faustman et al. (2010) indicated that the presence of oxygen catalyzes the formation of primary oxidative products that propagate to form secondary oxidative products that continue the oxidative chain reaction. Therefore, elimination of oxygen from meat packaging is a critical factor preventing lipid oxidation during storage. As expected, lipid oxidation was favored by long time storage under aerobic conditions (dry aging) and increased TBARS values were observed for dry-aged treatments in comparison with wet-aged counterparts.

Lipid oxidation increases beef flavor deterioration during aging, and this deterioration can be closely related to TBARS. Campo et al. (2006) considered TBARS values exceeding 2.28 mg of malonaldehyde per kg as unacceptable for beef because at this level rancid flavor overpowers beef flavor. Conversely, Hughes, McPhail, Kearney, Clarke, and Warner (2015) considered levels between 2.60 and 3.11 mg of malonaldehyde per kg as acceptable to consumers. In this study, the limiting threshold of 2 mg of malonaldehyde per kg was met by steaks from RH50 and RH85 dry-aged treatments at day 4 of RD, suggesting that dry-aged steaks aged for 42 days can be merchandised in the retail level for 3 d without detrimental effects on lipid oxidation.

4. Conclusion

Dry aging of beef resulted in decreased lightness and redness values and increased lipid oxidation compared to wet aging. With prolonged RD, dry aging of beef has the potential to reduce color and lipid stability compared to wet aging and thus reduce display life. In this study, the 14.5 color threshold for a* value was met by steaks from the RH70 and RH85 treatments after 2.7 days of RD, and after 2.4 days for the RH50 treatments, while the 20% discoloration threshold was met by steaks from the RH50 after 4.1 days of RD, and after 3.9 and 4.7 days for the RH70 and RH85 treatments, respectively. Dry-aged steaks from RH50 and RH85 treatments overtook the consumer thresholds for acceptable levels of lipid oxidation at d 4 of RD. Results suggest that dry-aged steaks aged for 42 days can be merchandised at retail level for 2 or more days without detrimental effects on color and lipid oxidation.

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Declaration of Competing Interest

None.

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