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Evaluation of Beef Retail Shelf-Life Following Extended Storage at Low Temperature

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Abstract: The storage of meat at temperatures below regular chilling can extend the storage shelf-life of fresh beef. However, the retail shelf-life of beef after extended storage has not been thoroughly investigated. This study evaluated the retail shelf-life of steaks derived from 10 upper two-thirds Choice beef inside rounds, bone-in ribeyes, and striploins that had been stored at low temperature (LT; −2.7 ± 0.3°C) for different periods of time. The subprimals were fabricated into 3 pieces, vacuum packaged, and randomly allocated to an LT storage time of 60, 75, and 90 d. After each storage time, subprimal portions were fabricated into steaks, overwrapped, and placed in a retail display case (3°C) for 7 d. Steaks were evaluated daily for instrumental and visual color and microbial levels (aerobic plate counts [APC], lactic acid bacteria counts, and *Pseudomonas* spp. counts) on days 0, 2, 4, and 7. For all subprimals, the initial redness (*a** values) of LT75 and LT90 steaks was greater ($P < 0.05$) than that of LT60 steaks. In general, irrespective of LT storage time or retail display day, visual panelists did not detect differences in lean color and discoloration of steaks. For all subprimals, the APC of LT60 steaks on days 0, 2, and 4 of the retail display were lower ($P < 0.05$) than those of LT75 and LT90 samples. Samples from LT60 presented a longer microbial retail shelf-life than those from LT75 and LT90 due to lower initial microbial loads following LT storage. However, the retail shelf-life of samples from LT75 and LT90 was similar. Overall, these results demonstrated the impact of LT60, LT75, and LT90 on the retail shelf-life of different beef subprimals.

Key words: extended chilled storage, shelf-life, meat color, retail display
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Introduction

As the demand for fresh beef continues to grow worldwide, extending shelf-life has become very important for beef-producing and exporting countries such as the United States (Gonzalez et al., 2022). Although refrigerated storage (2–4°C) is commonly used to preserve the quality of fresh beef, microbial spoilage can limit the storage time (Hopkins and Thompson, 2002; Colle et al., 2015; Coombs et al., 2017). Freezing is an effective method to extend the storage time of beef products, but it may result in undesirable changes, such as decreased water-holding capacity and color stability (Coombs et al., 2017). However, recent studies have demonstrated that using temperatures lower than regular chilling (i.e., <2–4°C) but above freezing (>−3°C) can significantly extend the storage shelf-life of fresh beef to up to 20 wk, compared to an average of 6–8 wk in conventional chilled storage (Small et al., 2012; Chen et al., 2019; Chen et al., 2020). The use of temperatures below typical chilling can slow the growth rate of spoilage microflora and thus extend the storage shelf-life of beef products while maintaining their fresh status. Our previous study (Gonzalez et al., 2023) also showed that the Warner-Bratzler shear force values of steaks fabricated from beef inside rounds (IR), bone-in ribeyes (RE), and striploins (SL)

that had been stored at low temperature (LT; −2.7 ± 0.3°C) decreased with increased storage time of the subprimals (up to 90 d). Additionally, the perceived tenderness of the steaks by consumers increased with no adverse effect on juiciness, flavor, and overall liking when compared to 21-d wet-aged steaks. These findings suggest that using lower storage temperatures could be a promising approach for maintaining the quality of fresh beef during extended storage.

Meat color is a crucial quality attribute that heavily influences consumers' purchase decisions at the retail level (Tomasevic et al., 2021; Ramanathan et al., 2022). Consumers prefer a bright cherry red color in beef as it is associated with freshness and wholesomeness. Deviations from this desired color result in beef products at retail being initially discounted and eventually discarded if not sold, which generates food waste and economic losses for the meat industry (Mancini and Hunt, 2005; Ramanathan et al., 2022; King et al., 2023). External factors such as storage time and temperature can affect meat color and the growth rate of spoilage bacteria. A recent study (Zhang et al., 2023) evaluated the retail color of steaks fabricated from beef striploins after extended vacuum-packaged storage at −1°C. This study reported that the steaks from striploins stored for 12 wk had a retail shelf-life of at least 5 d, whereas it was shorter (3 d) for those stored for 16 to 20 wk (Zhang et al., 2023). However, the color stability and microbial shelf-life of beef subprimals after LT storage, which uses a slightly lower temperature (−2.7°C), have not been investigated. Therefore, the objective of this study was to evaluate the color and microbial shelf-life during retail display of beef IR, RE, and SL subprimals following 60, 75, and 90 d of LT storage. The overall hypothesis was that there were no differences between the storage shelf-life of beef subprimals that were stored for 60, 75, and 90 d at temperatures lower than typical chilling (−2.7°C).

Materials and Methods

Sample collection and processing

The Colorado State University Institutional Review Board approved the procedures used in this study (IRB exemption #2784). Beef subprimals (IR [IMPS#169], RE [IMPS#109E], and SL [IMPS#180]) were collected from upper two-thirds Choice beef carcasses ($n = 10$) in a commercial beef processing facility. The subprimals were transported in a refrigerated truck to the Department of Animal Sciences Global Food Innovation Center (GFIC) at Colorado State University (Fort Collins, CO). Upon arrival, each subprimal was portioned, individually vacuum packaged, and randomly assigned to an LT storage period (60, 75, or 90 d; −2.7 ± 0.3°C). The packaged pieces were then separated by subprimals and LT storage time, boxed, and transported overnight in a refrigerated truck to an LT storage facility. Following each storage period, the products were shipped overnight to the GFIC. Immediately after arrival, each vacuum-packaged subprimal was weighed, aseptically removed from its packaging, and placed on a sanitized tray to obtain the weight of the meat without the packaging and any product purge. Five 1.27-cm steaks were cut from each portion and were placed on individual white Styrofoam trays lined with absorbent pads and were overwrapped with oxygen-permeable polyvinyl chloride (PVC) packaging film (O_2 transmission = 23,250 mL x m^2 x d^{-1}, 72 gauge; Resinite Packaging Films, Borden, Inc., North Andover, MA). Overwrapped steaks were placed in a commercial multi-deck retail display case (Hussman Model No. M3X8GEP) under continuous, cool-white, fluorescent lighting (2,200 to 2,500 lx) at a temperature of 3°C (±1°C) for 7 d (the first day of retail display was designated as d-0). Each sample was identified with a random four-digit number for visual and instrumental color evaluation. Trays were rotated within the display case once a day to account for light intensity and temperature variation within the display case.

Purge loss

Purge loss (PL) for each subprimal portion after LT storage was determined by taking the weight of the vacuum-packed subprimal portion (total weight), the weight of the meat without packaging (meat weight), and the weight of the dry package. The PL was expressed as a percentage relative to total weight using the following formula:

$$\%\mathrm{PL} = [(\text{total weight} - [\text{meat weight} + \text{package weight}])/(\text{total weight})] \times 100.$$

Color evaluation

Instrumental lean color measurements were obtained with a portable HunterLab MiniScan LabScan EZ4500 colorimeter (Hunter Associates Laboratory, Reston, VA) equipped with a 6-mm measurement port (2.54-cm diameter aperture, illuminant

A, and 10° standard observer). The instrument was standardized before each use, using standard tiles covered with overwrap film. Color measurements (6 technical replicate readings), CIE L^* (lightness), a^* (redness), and b^* (yellowness) were obtained once every day from the same steak ($n = 10$) for the duration of the retail display from day 0 to day 7. The color measurements of each steak were taken at random locations for the duration of retail display through the overwrap film. Averages of the L^*, a^*, and b^* values were used for statistical analysis. In addition, a minimum of 6 trained panelists evaluated the percent of metmyoglobin formation (lean discoloration) and lean color daily using a continuous 8-point scale in which 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, and 8 = extremely dark red. Panelists were selected and trained following the American Meat Science Association Meat Color Measurement Guidelines (King et al., 2023). Ratings of individual panelists were collected using Qualtrics software (Provo, UT) and averaged to obtain a single panel rating for each sample and visual attribute.

Microbiological analyses

On days 0, 2, 4, and 7 of retail display, randomly selected IR, RE, and SL steaks ($n = 10$, total $N = 40$) were analyzed for bacterial population levels. A 4 × 4 cm^2 sample was aseptically excised from the center of each steak using a disposable scalpel. The excised samples were placed into a Whirl-Pak filter bag (710 mL; Nasco, Pleasant Prairie, WI) with 50 mL of Maximum Recovery Diluent (MRD; Neogen Culture Media, Lansing, MI) and then mechanically pummeled for 2 min (Masticator, IUL Instruments, Barcelona, Spain). Samples were serially diluted in MRD, and aliquots of appropriate dilutions were surface plated, in duplicate, onto Tryptic Soy Agar (TSA; Neogen Culture Media) to enumerate aerobic bacterial populations (aerobic plate counts [APC]) and on *Pseudomonas* Agar Base with *Pseudomonas* CFC (cetrimide, fucidin, and cephalosporin) Selective Agar Supplement (PSA; Oxoid Ltd., Basingstoke, UK) to obtain *Pseudomonas* spp. counts. Samples were also analyzed for lactic acid bacteria (LAB) counts using the pour plate method with an overlay. Specifically, 1 mL of appropriate sample dilutions were mixed in 10 mL of molten (<45°C) Lactobacilli MRS Agar (Becton, Dickinson, and Company [BD], Sparks, MD); this was done in duplicate. After the agar had set, a 10-mL overlay of molten Lactobacilli MRS Agar was added to each plate to generate an anaerobic environment. All plates were incubated at 25°C, and colonies were counted after 72 ± 1 h (PSA plates) or 72 to 96 h ± 1 h (TSA and Lactobacilli MRS plates) of incubation.

Statistical analysis

All statistical analyses were conducted using R statistical software version 4.0.3 (R Core Team, 2020) within each subprimal because it is known that different subprimals/muscles will differ in color performance during retail display (McKenna et al., 2005). Purge loss as well as instrumental and visual color were analyzed using the *lme4* package (Bates et al., 2015) as a mixed model, where LT storage time (LT60, LT75, and LT90), retail display day, and their interaction were fixed effects, and the individual carcass was set as a random blocking factor. For the microbiological analysis, the experiment was designed as a 3 (LT storage times) × 4 (sampling times) factorial for each subprimal and bacterial count type (APC, LAB count, and *Pseudomonas* spp. count). Bacterial populations were expressed as least-squares means for log CFU/cm^2 under the assumption of a log-normal distribution of plate counts. All least-squares means were calculated using the *emmeans* package (Lenth, 2020). The differences between least-squares means are reported using a significance level of $\alpha = 0.05$ with Tukey's multiple comparison adjustment.

Results

Purge loss

The percentage of PL by subprimal and LT storage time is shown in Figure 1. Among the muscles examined, only IR had differences ($P < 0.05$) in PL with duration of LT storage, where the PL of LT60 samples (4.9%) was less ($P < 0.05$) than that of LT90 samples (8.8%), with LT75 being intermediate.

Color evaluation

Instrumental color values, lightness (L^*), redness (a^*), and yellowness (b^*) for IR, RE, and SL steaks for all LT storage times are presented in Tables 1, 2, and 3, respectively. For IR (Table 1), there was an interaction ($P < 0.05$) between LT storage time and display day for a^* and b^* values. However, there was no effect ($P \geq 0.05$) of LT storage time or display day for L^* values of IR steaks. On day 0 of retail display, LT75 and

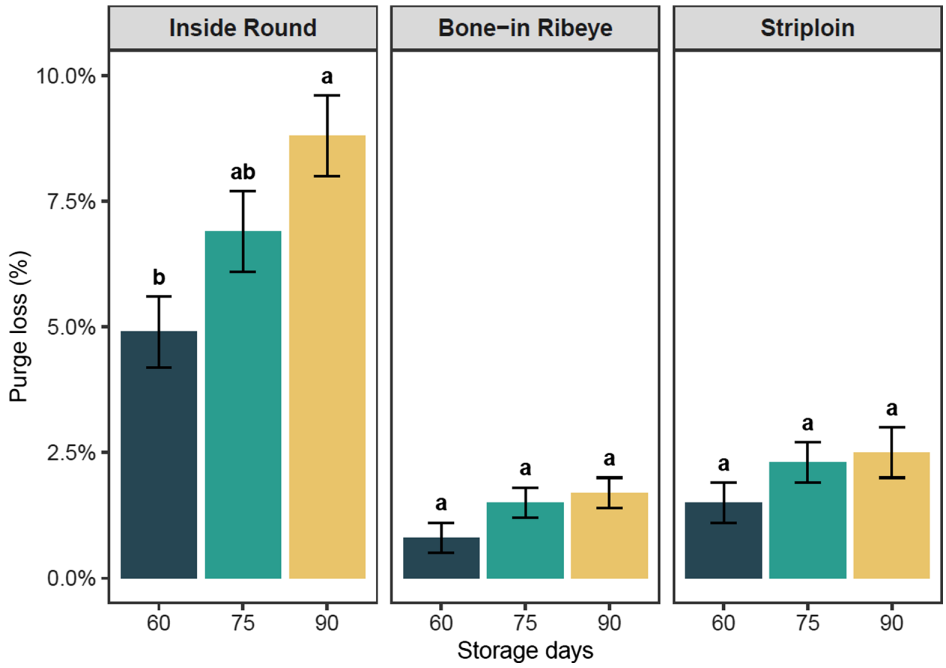

Figure 1. Effect of low-temperature (−2.7 ± 0.3°C) storage times (60, 75, or 90 d) on the percentage of purge loss of inside round, bone-in ribeye, and striploin subprimals ($n = 10$). Different letters (a,b) within each subprimal indicate significant differences ($P < 0.05$). Error bars represent the standard error of the mean.

Table 1. Effect of low-temperature (LT; −2.7 ± 0.3°C) storage times (60, 75, or 90 d) on surface lightness (*L** value), redness (*a** value), and yellowness (*b** value), and percentage of discoloration (metmyoglobin formation) evaluation by a trained panel ($n = 6$), of inside round steaks (IR; $n = 10$) during retail display (3°C) for 7 d under aerobic packaging

	Days in LT storage	Days of retail display								SEM[1]
		0	1	2	3	4	5	6	7	
L*	60	35.4	33.7	33.6	33.4	33.6	34.2	34.6	34.9	0.9
	75	37.6	35.7	34.7	34.5	34.8	34.2	34.6	35.0	0.9
	90	36.7	37.0	35.6	35.6	35.2	35.5	35.3	34.9	1.0
a*	60	19.3^{B}	17.7^{BCD}	15.7^{DEF}	14.6^{FG}	13.3^{GHI}	12.2^{HIJK}	10.5^{KLM}	9.5^{M}	0.5
	75	22.1^{A}	18.9^{BC}	16.9^{CDE}	15.6^{EF}	14.4^{FG}	13.4^{GHI}	11.7^{IJKL}	9.9^{LM}	0.5
	90	22.0^{A}	18.8^{BC}	16.0^{DEF}	14.0^{FGH}	12.8^{GHIJ}	11.1^{JKLM}	10.1^{LM}	8.9^{M}	0.5
b*	60	15.6^{CDE}	15.4^{CDEF}	14.6^{DEFGHI}	14.8^{CDEFGH}	13.9^{FGHI}	13.6^{GHI}	12.9^{I}	13.2^{HI}	0.4
	75	17.7^{A}	16.5^{ABC}	15.7^{BCD}	15.4^{CDEF}	15.1^{CDEFG}	15.2^{CDEFG}	14.9^{CDEFGH}	14.4^{DEFGHI}	0.4
	90	18.2^{A}	17.5^{AB}	15.3^{CDEFG}	14.0^{DEFGHI}	13.8^{EFGHI}	13.4^{GHI}	13.7^{FGHI}	13.8^{EFGHI}	0.4
%Dis[2]	60	1.6^{M}	7.2^{KLM}	25.7^{HIJKL}	38.2^{FGHI}	43.5^{EFGH}	49.8^{DEFG}	67.4^{BCD}	74.3^{ABC}	5.6
	75	0.1^{M}	7.5^{KLM}	20.9^{IJKLM}	23.9^{HIJKL}	32.0^{GHIJ}	45.9^{DEFGH}	59.2^{CDEF}	79.6^{ABC}	5.7
	90	4.0^{LM}	10.7^{JKLM}	20.8^{IJKLM}	28.4^{GHIJK}	48.6^{DEFG}	61.4^{BCDE}	84.4^{AB}	94.5^{A}	5.9

[1]SEM: standard error of the mean.

[2]%Dis: Percentage of discoloration.

[A–M]Least-squares means with different superscripts are different ($P < 0.05$).

LT90 IR steaks had greater ($P < 0.05$) *a** values than those in LT storage for 60 d. However, by day 1, the *a** values of LT75 and LT90 steaks were similar ($P \geq 0.05$) to those of LT60 samples. Following day 1, *a** values of all samples decreased in a similar ($P \geq 0.05$) manner until day 4 of retail display. On day 5, *a** values of LT90 steaks were less ($P < 0.05$) than those of LT75 steaks but similar ($P \geq 0.05$) to the redness of LT60 steaks. By the end of retail display (day 6 and 7), all samples had similar ($P \geq 0.05$) *a** values, regardless of LT storage time. The *b** values of LT75 and LT90 IR steaks on days 0 and 1 were greater

Table 2. Effect of low-temperature (LT; −2.7 ± 0.3°C) storage times (60, 75, or 90 d) on surface lightness (*L** value), redness (*a** value), and yellowness (*b** value) of bone-in ribeye steaks (RE; $n = 10$) during retail display (3°C) for 7 d under aerobic packaging

	Days in LT storage	Days of retail display								SEM
		0	1	2	3	4	5	6	7	
L*	60	34.5^{EFGHI}	33.6^{GHI}	33.6^{GHI}	32.0^{I}	33.6^{GHI}	33.8^{FGHI}	33.4^{GHI}	33.3^{HI}	0.7
	75	39.2^{AB}	38.3^{ABC}	36.8^{ABCDE}	37.4^{ABCDE}	37.7^{ABCD}	36.3^{CDEFG}	35.4^{DEFGH}	35.8^{CDEFGH}	0.6
	90	39.5^{A}	38.0^{ABCD}	36.8^{BCDE}	36.7^{BCDEF}	36.7^{BCDEF}	36.8^{BCDE}	35.0^{EFGHI}	33.9^{GHI}	0.6
a*	60	17.6^{ay}	16.2^{abx}	14.1^{bx}	13.5^{bcx}	11.0^{cdx}	8.7^{dey}	7.0^{ey}	7.7^{ey}	0.8
	75	19.9^{ax}	17.5^{bx}	15.4^{bcx}	14.1^{cdx}	12.8^{dex}	11.0^{efx}	10.1^{fx}	10.3^{fx}	0.7
	90	20.0^{ax}	18.0^{abx}	15.8^{bcx}	14.1^{cdx}	12.3^{dex}	10.4^{exy}	10.1^{ex}	10.7^{ex}	0.7
b*	60	14.2^{ay}	13.2^{aby}	11.9^{bcx}	12.1^{bcx}	11.0^{cdx}	10.7^{cdx}	9.3^{dy}	9.4^{dx}	0.5
	75	15.0^{axy}	13.6^{aby}	12.6^{bcx}	12.2^{bcdx}	12.0^{bcdx}	11.4^{cdex}	10.8^{dex}	9.9^{ex}	0.5
	90	15.8^{ax}	15.1^{ax}	13.2^{bx}	12.0^{bcx}	11.2^{cdx}	10.4^{cdx}	10.0^{dxy}	9.7^{dx}	0.5

SEM: standard error of the mean.

$^{A–I}$Least-squares means with different superscripts are different ($P < 0.05$).

$^{a–f}$Least-squares means with different superscripts within a row are different ($P < 0.05$).

^{x,y}Least-squares means with different superscripts within a column and color parameter are different ($P < 0.05$).

($P < 0.05$) than those of LT60 samples. However, from day 2 and until day 5, all *b** values of IR steaks were similar ($P \geq 0.05$). On day 6, LT75 steaks had greater ($P < 0.05$) *b** values than LT60 steaks; however, they were similar ($P \geq 0.05$) to *b** values of LT90 samples.

An interaction between LT storage time and display day was observed ($P < 0.05$) for *L** values of RE steaks (Table 2). The *L** values of LT90 and LT75 steaks were greater ($P < 0.05$) than those of LT60 steaks at the beginning of the display (days 0 to 4). There was no ($P \geq 0.05$) change in *L** values of LT60 steaks during display, whereas values of LT75 and LT90 samples decreased ($P < 0.05$) over the retail display period. Storage time in LT and display day influenced ($P < 0.05$) *a** and *b** values of RE steaks, but there was no interaction ($P \geq 0.05$). Steaks from LT75 and LT90 RE had a greater ($P < 0.05$) initial and final *a** value than steaks from LT60 RE. Overall, *a** values of all RE samples decreased ($P < 0.05$) over time regardless of the LT storage time. Yellowness (*b** values) of LT90 RE samples was higher ($P < 0.05$) than that of LT60 steaks at the beginning of the retail display, but by day 2 and until day 7 of the retail display, *b** values of steaks from all LT storage times were generally similar ($P \geq 0.05$).

For SL steaks (Table 3), there was an interaction between LT storage time and display day ($P < 0.05$) for *L** and *b** values, while only the main effects influenced ($P < 0.05$) redness (*a** values). The *L** values of LT60 SL steaks were less ($P < 0.05$) than those of LT75 and LT90 samples on day 0 of retail display, but these values increased ($P < 0.05$) slightly during retail display. There were no ($P \geq 0.05$) changes over time in the *L** values of LT75 and LT90 steaks. The *a** values of all SL samples decreased ($P < 0.05$) as the display day increased, irrespective of the LT storage time. Similar to IR and RE steaks, the initial redness of LT75 and LT90 SL samples was greater ($P < 0.05$) than that of LT60 SL steaks. Additionally, the initial *b** values of LT90 SL steaks were greater ($P < 0.05$) than LT60 steaks and similar ($P \geq 0.05$) to LT75 samples. Samples from LT60 had similar ($P \geq 0.05$) *b** values from day 0 to day 3. On day 4, *b** values of LT60 steaks decreased ($P < 0.05$) compared with the beginning of the display (days 0 and 1) and stayed similar ($P \geq 0.05$) thereafter until day 7. For LT75 samples, *b** values declined ($P < 0.05$) on day 2, compared with day 0, and stayed similar ($P \geq 0.05$) during the rest of the retail display. Similar to LT75 steaks, *b** values of samples from LT90 decreased ($P < 0.05$) on day 2. By the end of the retail display (days 6 and 7), LT90 samples had greater ($P < 0.05$) *b** values than LT60 steaks but were similar ($P \geq 0.05$) to LT75 samples.

Visual lean color and percentage of discoloration for IR are presented in Figure 2 and Table 1, respectively. There was no LT storage time × display day interaction ($P \geq 0.05$) or LT storage time effect ($P \geq 0.05$) on IR lean color. However, a display day effect ($P < 0.05$) was observed (Figure 2). Lean color scores of IR steaks increased ($P < 0.05$) during retail display. On day 1 of the retail display, lean color scores of samples were greater ($P < 0.05$) than day 0 scores, going from moderately bright cherry red to slightly bright

Table 3. Effect of low-temperature (LT; −2.7 ± 0.3°C) storage times (60, 75, or 90 d) on surface lightness (*L** value), redness (*a** value), and yellowness (*b** value), and percentage of discoloration (metmyoglobin formation) evaluation by a trained panel ($n = 6$), of striploin steaks (SL; $n = 10$) during retail display (3°C) for 7 d under aerobic packaging

	Days in LT storage	Days of retail display								SEM[1]
		0	1	2	3	4	5	6	7	
L*	60	31.6^{F}	32.7^{DEF}	32.8^{CDEF}	31.9^{EF}	33.4^{BCDEF}	33.6^{ABCDEF}	33.9^{ABCDE}	33.8^{ABCDE}	0.6
	75	35.0^{AB}	35.2^{AB}	34.4^{ABCD}	34.9^{AB}	34.4^{ABCD}	34.4^{ABCD}	34.4^{ABCD}	34.0^{ABCDE}	0.6
	90	35.7^{A}	34.1^{ABCDE}	34.6^{ABCD}	34.5^{ABCD}	33.8^{ABCDEF}	34.9^{ABCD}	35.1^{AB}	35.0^{ABC}	0.6
a*	60	17.3^{ay}	16.8^{ay}	15.7^{abxy}	14.6^{bcx}	13.1^{cdx}	11.8^{dx}	9.0^{ey}	7.2^{ex}	0.5
	75	19.0^{ax}	16.7^{by}	14.7^{cy}	13.6^{cdx}	12.2^{dex}	10.7^{efx}	8.8^{fgy}	8.4^{gx}	0.5
	90	19.9^{ax}	18.5^{abx}	16.3^{bcx}	15.1^{cdx}	13.7^{dex}	12.3^{efx}	11.0^{fx}	8.6^{gx}	0.6
b*	60	13.8^{BCD}	13.8^{BCD}	13.2^{CDEF}	12.9^{CDEFG}	11.9^{FGHI}	11.3^{HI}	10.7^{I}	10.7^{I}	0.4
	75	14.8^{AB}	13.5^{BCDE}	12.5^{DEFGH}	12.2^{EFGH}	11.6^{GHI}	11.6^{GHI}	11.2^{HI}	11.3^{HI}	0.4
	90	16.2^{A}	16.1^{A}	14.3^{BC}	13.6^{BCDE}	13.0^{CDEFG}	12.5^{DEFGH}	12.5^{DEFGH}	12.3^{EFGH}	0.4
%Dis[2]	60	0.4^{G}	0.4^{G}	5.8^{EFG}	9.9^{EFG}	20.6^{DE}	33.1^{CD}	64.4^{B}	88.7^{A}	3.5
	75	0.3^{G}	1.3^{FG}	3.9^{FG}	6.9^{EFG}	17.9^{DEF}	31.4^{CD}	64.5^{B}	85.0^{A}	3.5
	90	0.3^{FG}	2.2^{FG}	5.7^{EFG}	7.1^{EFG}	17.5^{DEFG}	30.8^{CD}	45.8^{C}	72.2^{AB}	3.8

[1]SEM: standard error of the mean.

[2]%Dis: Percentage of discoloration.

[A–I]Least-squares means with different superscripts are different ($P < 0.05$).

[a–g]Least-squares means with different superscripts within a row are different ($P < 0.05$).

[x–z]Least-squares means with different superscripts within a column and color parameter are different ($P < 0.05$).

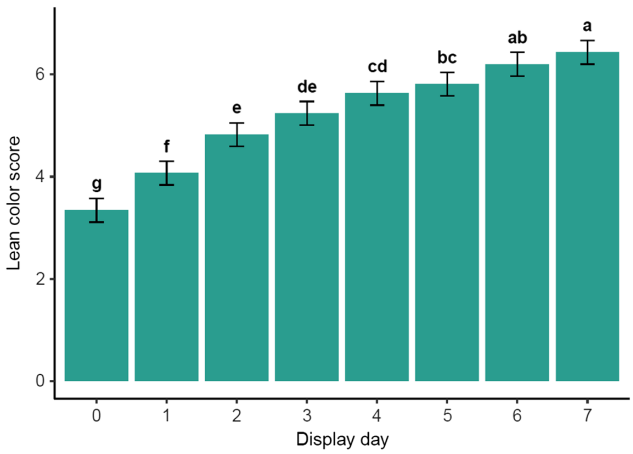

Figure 2. Effect of low-temperature (−2.7 ± 0.3°C) storage on visual lean color evaluation, by a trained panel ($n = 6$), of inside round steaks ($n = 10$) during retail display (3°C) for 7 d under aerobic packaging. Different letters (a–g) indicate significant differences ($P < 0.05$). Error bars represent the standard error of the mean. Panelists scored each steak to assess lean color using a continuous 8-point scale (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).

cherry red. Lean color scores of IR steaks increased ($P < 0.05$) from slightly bright cherry red to slightly dark cherry red by day 3, and on day 6, samples were scored as moderately dark red. There was an interaction ($P < 0.05$) between LT storage time × display day for the IR steak discoloration percentage (Table 1). From day 0 to day 2, samples from all LT storage times had similar ($P \geq 0.05$) discoloration percentages.

There was no interaction ($P \geq 0.05$) between LT storage time and display day on lean color and discoloration percentage of RE steaks (Figure 3), and only the display day was significant ($P < 0.05$). On day 2 of the display, the lean color score of steaks was greater ($P < 0.05$) compared with the day 0 score, going from moderately bright cherry red to slightly bright cherry red. On day 4 of the retail display, the sample lean color score increased ($P < 0.05$) from slightly bright cherry red to slightly dark cherry red. By day 6, RE samples achieved a moderately dark red score. The discoloration percentage of RE steaks increased ($P < 0.05$) during retail display, with day 3 steaks showing a greater ($P < 0.05$) percentage of discoloration than day 0 steaks. On day 4, the discoloration percentage of steaks was more than double the previous day.

For the SL samples, an interaction between LT storage time and display day was observed ($P < 0.05$) for the percentage of discoloration (Table 3). However, only the display day was significant ($P < 0.05$) for the lean color (Figure 4). Similar to RE steaks, on day 2 the lean color score of SL samples was greater ($P < 0.05$) than on day 0. However, it was not until day 3 of the retail display that lean color score of SL steaks declined

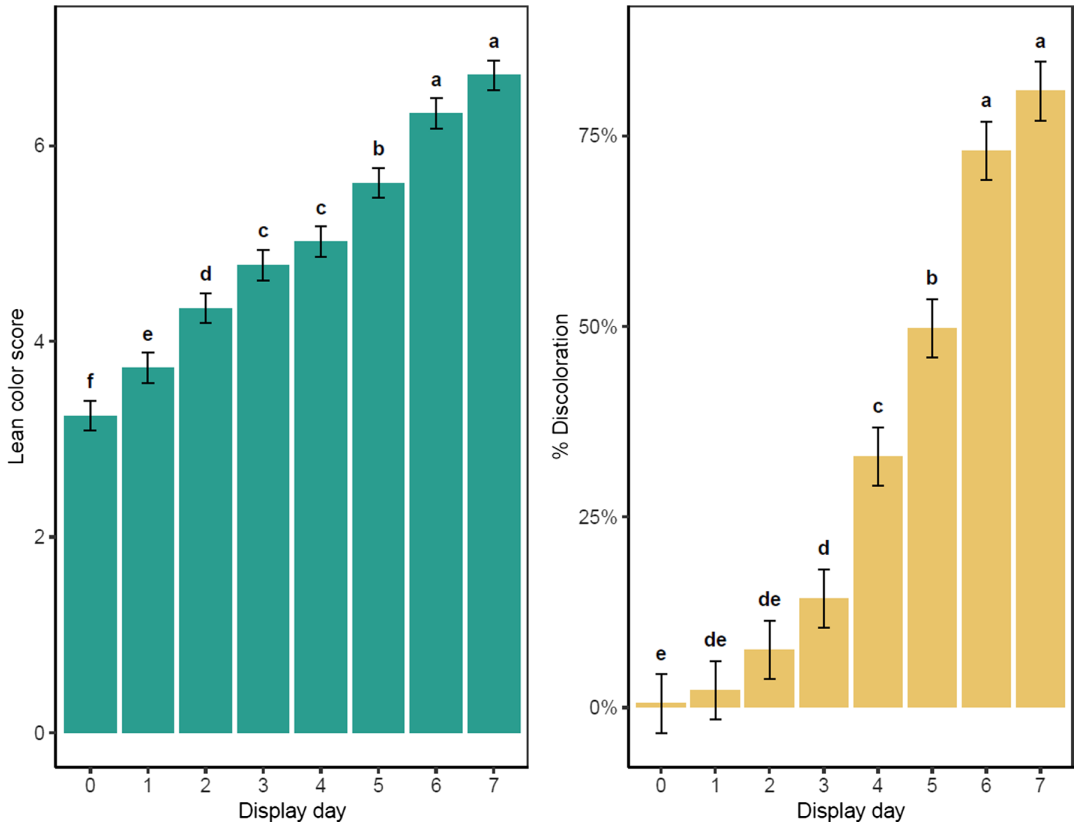

Figure 3. Effect of low-temperature (−2.7 ± 0.3°C) storage on visual lean color evaluation and percentage of discoloration (metmyoglobin formation) evaluation, by a trained panel ($n = 6$), of bone-in ribeye steaks ($n = 10$) during retail display (3°C) for 7 d under aerobic packaging. Different letters (a–f) indicate significant differences ($P < 0.05$). Error bars represent the standard error of the mean. Panelists scored each steak to assess lean color using a continuous 8-point scale (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).

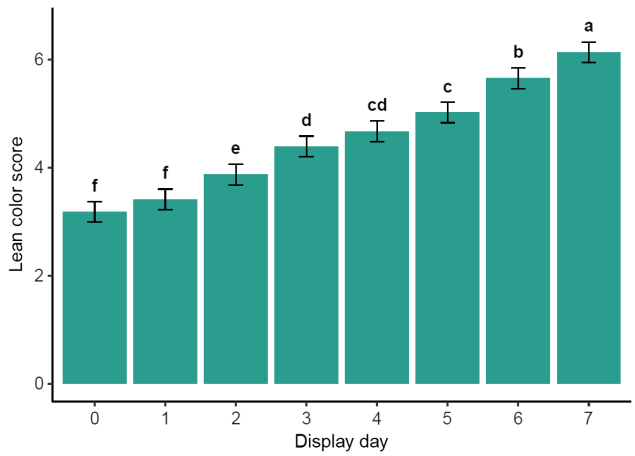

Figure 4. Effect of low-temperature (−2.7 ± 0.3°C) storage on visual lean color evaluation, by a trained panel ($n = 6$), of striploin steaks ($n = 10$) during retail display (3°C) for 7 d under aerobic packaging. Different letters (a–f) indicate significant differences ($P < 0.05$). Error bars represent the standard error of the mean. Panelists scored each steak to assess lean color using a continuous 8-point scale (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).

($P < 0.05$) from moderately bright cherry red to slightly bright cherry red. The percentage discoloration of SL steaks from all LT storage times increased ($P < 0.05$) during retail display, with no differences ($P \geq 0.05$) observed between the LT storage times up to day 6 of retail display, where the LT90 steaks had less ($P < 0.05$) percentage of discoloration than the LT60 and LT75 samples. Moreover, the discoloration percentage of SL steaks on day 4 was more than double their corresponding scores from day 3, regardless of the LT storage time.

Microbiological analyses

Microbial populations (APC, LAB counts, and *Pseudomonas* spp. counts) recovered from the IR, RE, and SL steaks on days 0, 2, 4, and 7 of retail display are presented in Tables 4, 5, and 6 and in Figure 5. For IR samples (Table 4), an interaction between LT storage time and display day was observed ($P < 0.05$) for the APC, whereas for the LAB and *Pseudomonas* spp.

Table 4. Mean ($n = 10$) bacterial counts (log CFU/cm^2 ± standard deviation) of inside round (IR) steaks in retail display (3°C, 7 d)

Bacterial count	Days of LT storage	Days of retail display			
		0	2	4	7
Aerobic bacterial populations	60	1.8 ± 0.3^{F}	2.7 ± 0.4^{F}	4.1 ± 0.6^{E}	6.3 ± 0.8^{C}
	75	4.6 ± 1.1^{E}	6.0 ± 0.8^{CD}	6.5 ± 0.6^{BC}	7.7 ± 0.5^{A}
	90	5.1 ± 0.8^{DE}	6.3 ± 0.5^{CD}	6.7 ± 1.0^{ABC}	7.6 ± 0.6^{AB}
Lactic acid bacteria	60	3.0 ± 1.0^{cy}	3.7 ± 1.2^{cy}	5.1 ± 0.7^{by}	6.4 ± 0.8^{ay}
	75	4.8 ± 0.9^{cx}	5.8 ± 0.7^{bx}	6.0 ± 0.5^{bx}	7.3 ± 0.8^{ax}
	90	4.9 ± 0.7^{cx}	5.8 ± 0.3^{bcx}	6.6 ± 1.0^{abx}	7.1 ± 0.7^{axy}
***Pseudomonas* spp.**	60	$<1.4 \pm 0.3^{cy1}$	1.6 ± 0.7^{cy}	3.2 ± 0.6^{by}	6.1 ± 0.9^{ay}
	75	2.0 ± 0.5^{dxy}	2.8 ± 0.5^{cx}	4.1 ± 0.5^{bx}	7.4 ± 0.4^{ax}
	90	2.2 ± 0.8^{dx}	3.4 ± 0.5^{cx}	4.7 ± 0.5^{bx}	7.4 ± 0.8^{ax}

Steaks were fabricated from subprimals that were previously held under vacuum-packaged, low-temperature (LT; −2.7 ± 0.3°C) storage conditions for 60, 75, or 90 d.

[1]Four of the 10 samples analyzed had a *Pseudomonas* spp. count that was below the microbial analysis detection limit of 1.2 log CFU/cm^2 (15 CFU/cm^2); therefore, the mean is reported as < (less than).

[A–F]Least-squares means with different superscripts are different ($P < 0.05$).

[a–d]Least-squares means with different superscripts within a row are different ($P < 0.05$).

[x,y]Least-squares means with different superscripts within a column and bacterial count type are different ($P < 0.05$).

Table 5. Mean ($n = 10$) bacterial counts (log CFU/cm^2 ± standard deviation) of bone-in ribeye (RE) steaks in retail display (3°C, 7 d)

Bacterial count	Days of LT storage	Days of retail display			
		0	2	4	7
Aerobic bacterial populations	60	4.1 ± 0.6^{G}	4.6 ± 0.6^{FG}	6.2 ± 0.5^{DE}	8.3 ± 0.3^{AB}
	75	5.3 ± 0.3^{EF}	6.2 ± 0.7^{D}	7.4 ± 0.9^{BC}	8.6 ± 0.3^{A}
	90	5.4 ± 0.4^{DEF}	5.9 ± 0.3^{DE}	7.3 ± 0.5^{C}	8.5 ± 0.3^{A}
Lactic acid bacteria	60	4.2 ± 0.6^{cy}	4.6 ± 0.6^{cy}	6.1 ± 0.6^{by}	7.8 ± 0.5^{ax}
	75	5.3 ± 0.3^{dx}	6.2 ± 0.8^{cx}	6.9 ± 0.7^{bx}	8.2 ± 0.4^{ax}
	90	5.4 ± 0.3^{cx}	6.0 ± 0.3^{cx}	7.3 ± 0.4^{bx}	8.3 ± 0.3^{ax}

Steaks were fabricated from subprimals that were previously held under vacuum-packaged, low-temperature (LT; −2.7 ± 0.3°C) storage conditions for 60, 75, or 90 d.

[A–G]Least-squares means with different superscripts are different ($P < 0.05$).

[a–d]Least-squares means with different superscripts within a row are different ($P < 0.05$).

[x,y]Least-squares means with different superscripts within a column and bacterial count type are different ($P < 0.05$).

counts, only the main effects were significant ($P < 0.05$). For LT60, LT75, and LT90 IR samples, APC increased ($P < 0.05$) from 1.8 to 6.3, 4.6 to 7.7, and 5.1 to 7.6 log CFU/cm^2, respectively, during the retail display period (Table 4). Regardless of the retail display day, the APC of LT60 IR steaks were less ($P < 0.05$) than those of LT75 and LT90 IR samples. Likewise, LAB counts of LT60 IR samples were less ($P < 0.05$) than those of LT75 and LT90 IR steaks but only until day 4 of retail display. On day 7, LAB counts of LT60 steaks were less ($P < 0.05$) than those of LT75 steaks but similar ($P \geq 0.05$) to those of LT90 steaks. Counts of LAB recovered from IR samples were initially (day 0) numerically similar or greater than the recovered APC. Regardless of the retail display day, *Pseudomonas* spp. counts of LT60 IR samples were less ($P < 0.05$) than those of LT75 and LT90 IR steaks. Initially recovered *Pseudomonas* spp. counts from all IR samples were numerically less than the recovered APC and LAB; however, they were numerically similar by day 7 of the retail display.

For RE samples (Table 5 and Figure 5), LT storage time × display day was significant ($P < 0.05$) for the APC but not for the LAB and *Pseudomonas* spp. counts. Both LT storage time and display day affected ($P < 0.05$) the LAB counts, but only display day ($P < 0.05$) affected the *Pseudomonas* spp. counts (Figure 5). Aerobic bacterial populations (APC) of LT60, LT75, and LT90 RE steaks increased ($P < 0.05$) from 4.1 to

Table 6. Mean ($n = 10$) bacterial counts (log CFU/cm^2 ± standard deviation) of striploin (SL) steaks in retail display (3°C, 7 d)

Bacterial count	Days of LT storage	Days of retail display			
		0	2	4	7
Aerobic bacterial populations	60	3.1 ± 0.6^{G}	3.7 ± 0.7^{G}	5.5 ± 0.5^{DEF}	7.3 ± 0.4^{AB}
	75	4.8 ± 0.6^{F}	5.7 ± 0.6^{CD}	6.5 ± 0.5^{BC}	8.1 ± 0.6^{A}
	90	4.8 ± 0.6^{EF}	5.7 ± 0.6^{CDE}	6.4 ± 0.7^{BC}	7.6 ± 0.3^{A}
Lactic acid bacteria	60	3.8 ± 0.7^{cy}	4.3 ± 0.9^{cy}	5.6 ± 0.6^{bx}	6.4 ± 0.4^{ay}
	75	4.7 ± 0.6^{cx}	5.8 ± 0.7^{bx}	6.1 ± 0.4^{bx}	7.4 ± 0.4^{ax}
	90	4.7 ± 0.5^{cx}	5.7 ± 0.7^{bx}	6.3 ± 0.7^{abx}	7.0 ± 0.6^{axy}
***Pseudomonas* spp.**	60	2.0 ± 0.6^{cy}	2.8 ± 0.5^{cy}	5.0 ± 0.6^{bx}	7.3 ± 0.5^{ax}
	75	2.7 ± 0.6^{dxy}	3.8 ± 0.9^{cx}	5.4 ± 0.9^{bx}	7.8 ± 0.5^{ax}
	90	2.8 ± 0.8^{cx}	3.4 ± 0.7^{cxy}	5.0 ± 0.6^{bx}	7.5 ± 0.4^{ax}

Steaks were fabricated from subprimals that were previously held under vacuum-packaged, low-temperature (LT; −2.7 ± 0.3°C) storage conditions for 60, 75, or 90 d.

$^{A–G}$Least-squares means with different superscripts are different ($P < 0.05$).

$^{a–d}$Least-squares means with different superscripts within a row are different ($P < 0.05$).

^{x,y}Least-squares means with different superscripts within a column and bacterial count type are different ($P < 0.05$).

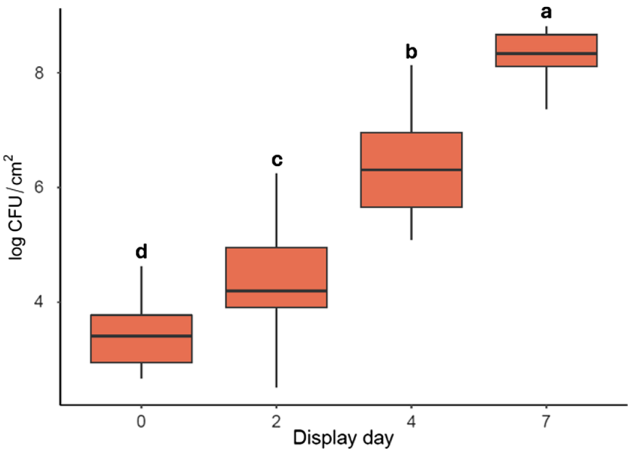

Figure 5. Effect of low-temperature (−2.7 ± 0.3°C) storage on *Pseudomonas* spp. counts (log CFU/cm^2 ± standard deviation) of bone-in ribeye ($n = 10$) during retail display (3°C) for 7 d under aerobic packaging. Different letters (a–d) indicate significant differences ($P < 0.05$).

8.3, 5.3 to 8.6, and 5.4 to 8.5 log CFU/cm^2, respectively, from day 0 to day 7 of retail display. The APC of LT60 RE steaks were less ($P < 0.05$) than those of the LT75 and LT90 samples until day 4 of retail display, and similar by day 7. Similarly, even when the interaction was not significant, LAB counts of LT75 and LT90 RE steaks were greater ($P < 0.05$) than the LAB counts of LT60 steaks until day 4. *Pseudomonas* spp. counts of RE samples increased ($P < 0.05$) from 3.4 log CFU/cm^2 on day 0 to 8.3 log CFU/cm^2 by the end of the display period.

Similar to the other subprimals, there was an LT storage time and display day interaction ($P < 0.05$) for the APC of SL steaks (Table 6). Initial APC of LT60, LT75, and LT90 SL samples were 3.1, 4.8, and 4.8 log CFU/cm^2, respectively, and these increased ($P < 0.05$) to 7.3, 8.1, and 7.6 log CFU/cm^2, respectively, by the end of retail display. The APC of LT60 SL steaks on days 0, 2, and 4 were less ($P < 0.05$) than the APC of corresponding LT75 and LT90 samples and similar ($P \geq 0.05$) by day 7. LT storage time and display day influenced ($P < 0.05$) the LAB and *Pseudomonas* spp. counts of SL steaks, but there was no interaction ($P \geq 0.05$) between display day and storage time. On days 0 and 2, LAB counts from LT60 SL steaks were less ($P < 0.05$) than LAB counts of LT75 and LT90 samples. For *Pseudomonas* spp. counts, samples of LT60 were less ($P < 0.05$) than those of LT90 steaks only on day 0. The *Pseudomonas* spp. counts were similar ($P \geq 0.05$) on days 4 and 7, regardless of the LT storage time.

Discussion

Temperatures lower than regular chilling have been used to extend the storage shelf-life of meat (Chen et al., 2019; Lu et al., 2019; Chen et al., 2020; Zhang et al., 2023). However, most of these studies only evaluated microbial population levels and meat quality characteristics immediately following the storage times. In the current study, we evaluated shelf-life during retail display following extended storage for 3 economically important beef subprimals (IR, RE, and SL).

An increase in purge is considered an economic loss to the meat industry and is unappealing to consumers (Kim et al., 2014; Van Rooyen et al., 2018). The PL

is likely an accumulative effect of changes in the water-holding capacity (Zhu et al., 2017), where an increased storage time could increase PL (Colle et al., 2015; Gagaoua et al., 2024). In the current study, the effect of LT storage time impacted PL ($P < 0.05$) of IR only (Figure 1), which could be due to the differences in muscle composition compared to RE and SL (Kirchofer et al., 2002). Similarly, Hur et al. (2009) evaluated PL in SL and IR stored at 0°C for 35 d and observed greater PL in IR than in SL. Moreover, Lu et al. (2019) reported an increase in PL from week 4 to week 8 in beef SL stored at 2°C and −4°C. The samples stored at −4°C were also evaluated for extended storage for up to 24 wk, but the PL did not further increase after week 8 (Lu et al., 2019).

Meat color is commonly associated with freshness and wholesomeness by consumers, which makes it a critical quality attribute (Tomasevic et al., 2021). In this study, both instrumental and visual color attributes were evaluated. In general, retail display day was the critical component in the color stability of the 3 muscles. While LT storage time played a role, it was not as significant as display day, probably because all LT storage times could be considered extended storage. The L^* values of LT60 steaks were less than those of LT75 and LT90 steaks from day 0 through day 4 of retail display for RE steaks and days 0 and 3 for SL samples. With more time in storage, greater proteolysis is expected, which could increase the amount of surface water and, consequently, the lightness (Hughes et al., 2020). Likewise, English et al. (2016b) reported a greater L^* value in beef SL aged 62 d compared to 21 d. Additionally, Colle et al. (2015) assessed the color of aged (2, 14, 21, 42, 63 d) SL steaks during 4 d of retail display and found an interaction between aging time and display day. These researchers observed lesser L^* values in SL steaks aged for 2 d as compared to the rest of the aging periods during the first 2 d of retail display.

The LT60 steaks for all subprimals had less initial (day 0) surface redness (a^*) than their corresponding LT75 and LT90 samples. As postmortem age increases, there is decreased competition from mitochondria for oxygen, consequently improving myoglobin oxygenation and initial color; however, color stability typically decreases due to a decrease in mitochondria functionality (Mancini and Ramanathan, 2014; Suman et al., 2014; Nair et al., 2018; Ramanathan and Mancini, 2018). In contrast, English et al. (2016a) reported a lesser initial a^* value in beef SL aged 62 d compared with 21 d. Also, Karney et al. (2022) observed lesser initial a^* values for beef SL steaks aged for 63 d compared with 14, 21, 28, and 49 d of aging. On the other hand, a recent study (Zhang et al., 2023) evaluating the retail shelf-life of beef SL after storage at −1°C reported that samples stored for 4 wk had greater initial a^* values (day 1) than those held for 8, 12, and 16 wk, but were similar to those stored for 20 wk. However, the a^* values decreased faster during the display days as storage time increased, indicating poor color stability during retail display (Zhang et al., 2023). Moreover, in another study that evaluated beef SL stored at −1°C up to 20 wk, initial a^* values gradually increased with an increase in storage time (Chen et al., 2020).

The LT storage did not affect the visual lean color of the steaks from the muscles we examined (Figures 2, 3 and 4). On day 2 of the retail display, the lean color scores of samples were greater compared with their corresponding day 0 scores. These results are similar to the instrumental redness values, where differences were observed between the LT storage times on day 0 of retail display, but a^* values declined over the display period. Similarly, English et al. (2016a) also observed a rapid increase in muscle color scores for SL aged 62 d, where in 24 h, score averages changed from bright cherry red to slightly bright cherry red. Moreover, lean muscle color scores from 62 d aged products were greater than 21 d but were similar to 42 d aged samples (English et al., 2016a).

In the current study, steaks from the 3 LT storage times discolored similarly within each subprimal evaluated. Slight discoloration of beef can generate consumer discrimination or an initial discount by the retailer, and extensive beef discoloration could be rejected by the consumer, causing a significant loss of resources and value for the meat industry (Mancini and Hunt, 2005; Suman et al., 2014; Ramanathan et al., 2022). With increased postmortem time, mitochondrial functionality decreases, leading to a decline in metmyoglobin reducing activity and faster discoloration (Ramanathan and Mancini, 2018; Ramanathan et al., 2019). In agreement, Colle et al. (2015) reported that SL steaks aged 21, 42, and 63 d had more surface discoloration than 2 and 14 d aged samples. According to Hood and Riordan (1973), consumers are 50% less likely to purchase beef steaks when the surface discoloration reaches 20%. In this study, the discoloration percentages stayed below 20% for RE and SL until days 3 and 4 of retail display, respectively, regardless of LT storage time. Although IR steaks discolored faster, with more than 20% discoloration present by day 2 of retail display, this was expected because it is comprised of muscles that are less color stable (*adductor* and *semimembranosus*; McKenna et al., 2005).

Steaks from the different muscles and LT storage times in the current study were analyzed for APC, LAB counts, and *Pseudomonas* spp. counts on days 0, 2, 4, and 7 of retail display. Day 0 APC of LT60, LT75, and LT90 RE steaks were numerically greater than those recovered from day 0 samples of IR and SL steaks. This could be due to the additional handling involved in processing bone-in products. Overall, the APC of LT60 steaks were consistently lower than the APC of LT75 and LT90 samples during the first 4 d of retail display across all subprimals. Interestingly, there were no differences between the APC of samples from LT75 and LT90, irrespective of the evaluated subprimal or display day. Chen et al. (2019) evaluated the initial (day 0) APC of SL steaks stored at −1 ± 0.5°C for up to 20 wk and, similarly, reported an increase in APC of samples up to week 5 of storage, but there were no differences between APC of samples stored for 9, 12, 15, and 20 wk. Colle et al. (2015) analyzed the APC of SL steaks on days 0 and 4 of retail display after storage at 0°C and reported an increase in the APC of SL steaks as aging time (2, 14, 21, 42, and 63 d) increased.

In general, recovered initial (day 0 of retail display) LAB counts of samples, regardless of subprimal, were numerically similar or greater than the recovered APC of their corresponding LT storage time. After 7 d of retail display, counts of LAB were similar to or lower than the APC. With a prolonged storage period under vacuum-packaged conditions, as in the case of LT storage, it is anticipated that bacterial populations will be dominated by LAB at the start of retail display. After exposing the samples to oxygen, other bacteria will grow and potentially slow down the growth rate of the LAB. Other studies have found similar results after extended vacuum storage of beef (Small et al., 2012; Luzardo et al., 2016; Chen et al., 2019). Moreover, the previously mentioned Chen et al. (2019) study also reported that LAB counts were greater than or similar to the APC. These authors observed similar APC and LAB initial counts of samples after 9 wk (63 d) of storage at −1 ± 0.5°C (Chen et al., 2019).

Pseudomonas spp. are considered important spoilage organisms of chilled meat stored under aerobic conditions (Pennacchia et al., 2011; Hilgarth et al., 2019). In the current study, initial *Pseudomonas* spp. counts on steaks were lower than those of APC and LAB counts regardless of LT storage time or subprimal. This was an expected finding because LT storage occurred under vacuum-packaged conditions, thereby inhibiting *Pseudomonas* spp. growth (Gill, 1996). Once the steaks were placed in retail display (aerobic packaging), *Pseudomonas* spp. populations increased rapidly, reaching levels of ca. 6 to 8 log CFU/cm^2 on day 7 of retail display.

Considering a threshold of more than 7 log CFU/cm^2 for APC as an indicator of microbial spoilage in beef (Ayres, 1960; Vieira et al., 2009), our study indicated that LT60 IR, RE, and SL steaks had a longer retail shelf-life than the LT75 and LT90 steaks. This outcome was expected because the LT60 samples had a lower initial microbial load than the LT75 and LT90 samples. However, the retail microbial shelf-life of LT75 and LT90 steaks was similar.

Conclusions

Retail shelf-life performance of beef products can affect consumer purchase decisions. Therefore, evaluating shelf-life performance (color and microbiological) after extended storage periods is essential. Among the subprimals evaluated, only the PL of the IR steaks was affected by the different LT storage times. The initial redness (*a** value) of LT60 steaks was less than that of LT75 and LT90 steaks for all the subprimals. In general, trained panelists did not find differences in lean color and discoloration percentage with the different LT storage times, irrespective of the subprimal evaluated. Additionally, microbial retail shelf-life depended on initial contamination levels on steaks fabricated from subprimals after LT storage, with a longer retail shelf-life for samples for LT60 and a similar retail shelf-life for LT75 and LT90 samples. The findings of this study can be useful for the meat industry when considering extending the storage shelf-life of boxed beef subprimals by storing them in a controlled low-temperature environment.

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