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Vascular rinsing and chilling effects on meat quality attributes from cull dairy cows associated with the two lowest-valued marketing classes

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ABSTRACT

Commercially harvested cull dairy cow carcasses ($n = 64$) from the two lowest-valued marketing classes (MC: Lean, LE; Light, LI) were conventionally chilled (CN) or vascularly rinsed with a chilled isotonic substrate solution (Rinse & Chill®; RC). *Longissimus lumborum* (LL) and *Triceps brachii* (TB) muscles were processed (steaks, ground). Early postmortem (first 24 h), RC resulted in a lower pH at each time measured. RC steaks had longer sarcomeres and lower shear force than CN. RC produced greater redness associated with blooming and display times. RC LE beef resulted in greater oxymyoglobin during display times. RC ground TB had greater moisture fat-free than CN. RC Lean LL had less purge loss compared to CN LE. RC had greater total pigments than CN. RC ground TB had greater oxygen consumption and lower thiobarbituric acid reactive substances compared to CN. RC has the potential to improve tenderness and color as well as limit lipid oxidation with similar benefits across the two marketing classes.

1. Introduction

In 2019, the United States harvested more than 3.2 million dairy cows in federally inspected plants (USDA, 2020). The predominant culling reasons of dairy cows are reproductive failure, low milk production, and udder disease (Hadley, Wolf, & Harsh, 2006; Weigel, Palmer, & Caraviello, 2003). The majority of the cows are marketed through a livestock market or auction, and only 37% of the cows are sent directly to slaughter (USDA, 2018), which could minimize transport time and, consequently, exposure to stress factors (Edwards-Callaway, Walker, & Tucker, 2019).

Cow carcasses are not classified as the higher USDA quality grades primarily because of maturity of the bone structure. Instead, these animals are classified based on the amount of lean meat they are expected to yield which are described by the categories of Breaker, Boner, Lean, and Light (Moreira, Rosa, & Schaefer, 2020; Peel & Doye, 2017). In terms of meat to bone ratios, Lean cows are thin (thinly fleshed) and Light cows are extremely thin (Moreira et al., 2020). Cow carcasses are processed into primal cuts and further processed into whole muscle cuts and marketed as steaks and roasts for the foodservice, as well as lean sources for ground beef (Nicholson, 2008; Woerner, 2010). However, due to the maturity and large live animal variability (parity, stage of lactation, body condition score) observed in cull dairy cows, the meat from these animals is often classified as tougher with prevalence of undesirable flavors (Stelzleni, Patten, Johnson, Calkins, & Gwartney, 2007; Therkildsen, Stolzenbach, & Byrne, 2011). In addition, the lean color of the *Longissimus thoracis* allowed to oxygenate from cows is typically less red and darker (Couvreur, Le Bec, Micol, & Picard, 2019) in comparison to younger beef animals (Li et al., 2020).

Dairy cows can experience significant stress, with milk production associated with heat stress, reproduction events, and termination of milking, as well as during livestock hauling to a packing plant and lairage (Alende et al., 2014; Becker, Collier, & Stone, 2020; Fabris et al., 2020). Extended periods of stress can limit the available glycogen at the time of harvest which would result in limited pH decline leading to darker color lean (Aberle, Forrest, Gerrard, & Mills, 2012).

The post-exsanguination vascular rinsing process, referred to as Rinse & Chill® was developed by MPSC, Inc. (Hudson, WI, USA). Part of the efficacy associated with Rinse & Chill® is based on the fact that the solution that is infused through the vasculature provides a source of

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metabolizable substrates that supports postmortem glycolytic activity. Rinse & Chill® has been reported to improve meat yields, increase meat tenderness, and reduce dark-cutting beef carcasses (Yancey, Dikeman, Addis, Katsanidis, & Pullen, 2002). In addition, Rinse & Chill® has been shown to decrease microbial contamination on beef carcasses (Feirtag & Pullen, 2003). Rinse & Chill® has also been shown to have a positive effect on the tenderness and color of bison and lamb (Fowler, Claus, & Hopkins, 2017; Mickelson & Claus, 2020). In ground pork, Rinse & Chill® resulted in greater oxygen consumption, and limited lipid oxidation (Kethavath et al., 2021). The objective of this study was to evaluate the effects of Rinse & Chill® application on commercially harvested cull dairy cows from the two lowest-valued marketing classes that represent animals with rather low body conditions.

2. Materials and methods

2.1. Cull dairy cow selection and carcass treatment

The experimental design included two chilling treatments (TRT: Control, CN; Rinse & Chill®, RC) and two marketing classes of cull dairy cows. Cull dairy cows (Holsteins) representing the two lowest-valued marketing classes (MC) were initially selected based on the criteria described by Peel and Doye (2017) by using body condition scores. To facilitate the distinction between the LE and LI categories, animals having a body condition score of 3 were not selected. After the TRT were implemented and the carcasses entered the cooler, carcasses were subsequently evaluated for bone maturity. Bone maturity was used for the sole purpose of excluding carcasses indicative of animals that were younger than 42 months. Bone maturity was visually assessed by evaluating the degree of fusion and ossification of the vertebrae (*sacral, lumbar, thoracic*) as described by (Romans, Costello, Jones, Carlson, & Ziegler, 1985). The same extensively experienced beef evaluator (~20 years) performed the bone maturity assessment at each harvest day. Bone maturity (C, D, E) was determined only for the purpose of describing the carcasses used in this study. Descriptive details on the animals selected, average carcass weights, and number of animals within the MC, bone maturity levels, and carcass TRT were collected (Table 1). Animal selection and therefore qualification for this research was primarily based on being categorized into the two MC. Within each marketing class as well as TRT, effort was made to have similar animal numbers relative to bone maturity.

Carcasses ($n = 64$) from two different cull dairy cow MC (Lean, LE; Light, LI) were conventionally chilled (CN) or RC processed immediately after exsanguination at a commercial plant in North Carolina. Carcasses were not electrically stimulated. Animals were harvested on three separate weeks. Exsanguination was performed by the packing plant personnel. CN animals were exsanguinated by cutting the carotid arteries. RC animals were exsanguinated by cutting the jugular veins followed by inserting the RC nozzle into a carotid artery to rinse the vasculature with a chilled isotonic solution (3 °C; 98.5% water; balance: glucose, polyphosphates, and maltose). The solution was infused at 10% based on the body weight. Carcasses designated for conventional chilling (RC not applied) were placed in a cooler (24 h) that averaged 2.6 °C (minimum of 1.3 °C, maximum of 5.3 °C). After RC processed, these carcasses were placed in the same cooler as CN. At 24 h postmortem, the *Longissimus lumborum* (LL) and *Triceps brachii* (TB) muscles were excised from the right side of the carcasses. Specifically, for the TB, an anatomically center section (~8 cm wide) was excised by two parallel cuts through the muscle perpendicular to the humerus bone followed by a lateral cut across the top of the bone. The muscles were vacuum (VAC) packaged and were shipped overnight with freezer ice packs to the University of Wisconsin-Madison (averaged 3.1 °C internal temperature upon delivery).

Table 1
Average hot carcass weights (kg)[a] and the number of cull dairy cows (Holsteins) within each chilling treatment[b], marketing class[b], and skeletal maturity.

Maturity	CN		RC	
	LE	LI	LE	LI
	Ave. (std. dev.)	Ave. (std. dev.)	Ave. (std. dev.)	Ave. (std. dev.)
C	560.4 (74.0)	542.8 (59.9)	659.1 (84.4)	532.9 (70.0)
	$n = 8$	$n = 9$	$n = 8$	$n = 8$
D	611.3 (68.4)	543.8 (56.4)	641.0 (29.3)	600.2 (46.3)
	$n = 7$	$n = 6$	$n = 8$	$n = 5$
E	693.0	513.0	566.0 (9.9)	478.0
	$n = 1$	$n = 1$	$n = 2$	$n = 1$

[a] Weights: ave. = average; std. dev. = standard deviation.
[b] Chilling treatment (CN=Control, RC = Rinse & Chill®); Marketing class (LE = Lean, LI = Light).

2.2. Temperature and pH decline

Temperature (model 11,040; DeltaTrak; Pleasanton, CA, USA) and pH decline (model MPI pH-Meter; Meat Probes Inc., Topeka, KS, USA) were recorded in the LL and *semimembranosus* (SM). Measurements were obtained at 1, 4, 8, 12 and 24 h postmortem. The pH meter and the standards (pH 4.0 and 7.0; ~2 °C) were stored in the cooler for at least 6 h prior to calibration and the start of measurements.

2.3. Muscle processing

On 4 d postmortem percentage purge associated with the LL muscles was determined. After removing the TB and LL from the VAC packages, all visible fat was trimmed before the muscles were further processed (steaks, ground). For determining Warner-Bratzler shear force, sarcomere length, and cooking loss, LL muscles were cut perpendicular to the length into steaks ($n = 3$, each 2.5 cm thick) and individually VAC packaged in plastic bags and stored (2 °C) in the dark (until aged 7 d postmortem). The remaining portion of each individual muscle was ground (model 548 J11; Biro Manufacturing Co., Marblehead, OH, USA) sequentially through two plates (19 mm, 3 mm). The ground beef samples were overwrapped with oxygen-permeable polyvinyl chloride film using a single-roll over-wrapper on a Styrofoam tray (Kethavath et al., 2021). Before displaying the ground beef samples, they were placed in a walk-in cooler (2 °C) and allowed to bloom for 1 h prior to taking measurements. PVC wrapped ground beef was continuously displayed (1, 4, 7 d postmortem) in an open-topped refrigerated (3 °C) display case under fluorescent lighting (40 watt, that provided approximately 1615 lx (Kethavath et al., 2021). A separate set of LI ground beef samples (LL, TB) were PVC wrapped and stored in the dark (1–2 °C) for 2 d prior to oxygen consumption determination.

The remaining ground beef samples were individually VAC packaged (vacuum setting, 10/10; model 2100-C; UltraVac-dual-chamber; Koch Equipment LLC., Kansas City, MO) in plastic pouches (2.7 mil thick, OTR 3-6 cc/m^2/24 h atm @ 4 °C, 0% relative humidity, blend of very-low-density polyethylene and ethylene vinyl acetate copolymer, 18 × 30 cm, product code 9KN81, Sealed Air Corporation) and stored in the dark (2 °C, 3 d) for subsequent water holding capacity determined as expressible moisture (EM). Samples used to analyze total pigments, pH and moisture fat-free (MFF) were stored in the freezer (-20 °C, 4 d) and were thawed (2 °C, 1 d), before the day of analysis. For hexanal and TBARS analyses (thiobarbituric acid reactive substances), only LE ground TB samples were collected, VAC packaged and stored in the freezer (-20 °C).

2.4. Warner-Bratzler shear force (WBS), sarcomere length, and cooking loss

Duplicate LL steaks from each treatment and marketing class combination were used for WBS determination. The steaks for WBS were weighed before being cooked and after the cooked steaks cooled (2 h) at

room temperature. Internal temperature during cooking was determined by a thermocouple needle (Type K; Electronic Temperature Instruments LTD West Sussex BN14 8NW UK, model 920,000–00; Digi-Sense; Cole-Parmer Instrument Company, Vernon Hills, IL, USA) inserted into the geometric center of each steak (American Meat Science Association, 2015). The steaks (four at a time, each TRT x MC included) were cooked on an electrical grill (model GGR50; George Foreman Grill). Once the steaks were placed on the grill, the metal dome cover was position on top of the grill. Steaks were turned initially when the internal temperature reached 41 °C and removed at 68 °C. After steaks were weighed to determine cooking loss, four strips (1 cm wide by 1 cm thick) were removed from each steak parallel to the muscle fibers. A Warner-Bratzler Shear Apparatus was used as described by Kethavath et al. (2021).

One steak was used to determine the sarcomere length by a laser diffraction method (Cross, West, & Dutson, 1981; Kethavath et al., 2021). The steak was cut into roughly 2.5 cm^3 duplicate samples and chemically processed after which four muscle fiber samples were removed. Eight sarcomere lengths were determined and the sample average was used for the statistical analysis.

2.5. Instrumental color and myoglobin determinations

Meat color bloom (allowed to oxygenate for 1 h post-grinding) and color during display (1, 4, and 7 d) were determined using colorimeter and scanning reflectance spectrophotometer. Six colorimeter measurements and two spectrophotometric measurements were obtained on each ground sample of LL and TB. Meat surface color was measured using a colorimeter as described by Kethavath et al. (2021). Measurements were collected for a* (redness) and L* (lightness).

To estimate the chemical states of myoglobin the instrument parameters described by Kethavath et al. (2021) for using a reflectance spectrophotometer and an integrating sphere were followed. The chemical states of myoglobin were estimated using percentage reflectance (R) ratios (deoxymyoglobin, DMb, %R at 474 nm/%R at 525 nm; metmyoglobin (MMb, %R at 572 nm/%R at 525 nm; oxymyoglobin, OMb, %R at 610 nm/%R at 525 nm) recommended by American Meat Science Association (2012). Averages were calculated from duplicate readings from each sample and used for the statistical analysis.

Total pigments were analyzed using 5 g of ground meat (4 d post-mortem) as described by American Meat Science Association (2012) for determining myoglobin concentration. It should be noted that the mg/g units represent Mb and hemoglobin in the meat as the millimolar extinction 139 coefficient of hemoglobin also is 7.6 on a mass basis.

2.6. Ultimate pH, water holding capacity, and oxygen consumption determinations

For ultimate pH of ground beef, samples (in duplicate, approx. 10 g each) were homogenized in a mini container in 30 ml distilled water. The homogenate was filtered using Whatman #1 filter paper (110 mm) before readings were determined using a calibrated (4, 7 buffer standards) pH meter (model PB-11-P11–1, Sartorius pH Basic with a glass electrode). Moisture fat-free (MFF) was determined on 4-g ground meat samples using an integrated rapid moisture and fat analyzer (moisture: Smart Turbo model 907,993; fat: Smart Trac model meat trac magnet 158,990; CEM Corp.). Water Holding Capacity (WHC) was reported as expressible moisture percentage (Kethavath et al., 2021) determined in duplicate on approximately 100 g samples of ground TB (3 d postmortem).

Oxygen consumption was determined as described by Kethavath et al. (2021). On 2 d postmortem the PVC on the LI ground beef (LL and TB packages) was removed before the product was VAC packaged and evaluated for oxygen consumption. Oxygen consumption data were reported as residual oxygen percentage left in the VAC packaged sample at different intervals of time (0, 20, 40, 60, 80 min).

2.7. Hexanal and thiobarbituric acid reactive substances determinations

Hexanal and thiobarbituric acid reactive substances (TBARS) were determined on cooked LE ground beef patties. Raw ground beef (125 g) was formed into 1.3-cm thick patties (Progressive® patty set). Patties were placed on broiler pans and cooked in a preheated oven to 163 °C (fan speed-high; model Master 450; Garland; Mississauga, Ontario, Canada) until their internal temperature reached 71 °C. Cooked patties were individually VAC packaged in plastic pouches and stored for 1 and 10 d in a cooler (3 °C). After being stored in the vacuum package, patties were finely ground (model DLC-1SS; Cuisinart mini-prep processor; Windsor, NJ, USA) and stored overnight before analysis. At each display time a 1 g sample for hexanal analysis from each patty was frozen (-20 °C) in a capped 20 ml vial. The remaining ground patty was stored (3 °C) in a sealed vacuum bag before TBARS determination (1 d after fine ground).

TBARS were determined according to the procedure described by Witte, Krause, and Bailey (1970) on duplicate 10 g samples of the cooked ground beef. Hexanal was extracted using a solid phase micro-extraction (SPME) technique following a procedure described by Park, Undeland, Sannaveerappa, and Richards (2013) and reported as μmol/kg meat.

2.8. Preparation of myofibrils

The purification of myofibrils from muscle tissues was performed at 4 °C according to the myofibril preparation procedure described by Huang, Huang, Xu, and Zhou (2009) with minor revisions. The meat samples were homogenized in the ratio 1:7.5 (meat: solution; *w/v*) of pyrophosphate relaxing buffer (PRB: 100 mM KCl; 2 mM $MgCl_2$; 2 mM egtazic acid, EGTA; 1 mM dithiothreitol; 2 mM $Na_4P_2O_7$; 10 mM Tris-maleate; pH 6.8). The homogenate was centrifuged (1000 ×*g*, 10 min), the pellet was washed eight times with 7.5 ml of a low-salt buffer (same as PRB except pyrophosphate omitted), the myofibrils were suspended in 7.5 ml of Tris–EDTA buffer (15 mM Tris; 5 mM ethylenediaminetetraacetic acid, EDTA; pH 8.0). The pure myofibrils were mixed with 2 ml of treatment buffer (125 mM Tris; 4% sodium dodecyl sulfate, SDS). The protein concentration of the samples was determined with a BCA Protein Assay Kit (BCA, bicinchoninic acid; Thermo Fisher Scientific, Waltham, MA, USA).

2.9. SDS-PAGE

Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) was performed on 12% Mini-PROTEAN® TGX™ Precast Protein Gels (Bio-Rad Laboratories, Hercules, CA, USA) in a discontinuous buffer system as described by Huang et al. (2009). A subset of the LE experimental samples was selected to be represented of each harvest period and chilling treatment based on bone maturity (CN: C = 4, D = 3, E = 1; RC: C = 4, D = 4, E = 1) and WBS averages (excluded samples with large standard deviations) for the LL steaks.

The target proteins were visualized with the ChemiDoc XRS gel documentation system (Bio-Rad Laboratories). The intensities of the SDS-PAGE bands were quantified using Image Lab software (Bio-Rad Laboratories) within the calibration range.

2.10. Statistical analysis

For all statistical analyses, animal served as the experimental unit (random effect) and week was included in the model as a co-variant to account for potential variation associated with the different weeks required for animal harvest. A randomized block design with the fixed effects of carcass chilling treatment (TRT: CN, RC) and marketing class (MC: LE, LI) was used to test the main effects and interactions (TRT x MC) for purge percentage, expressible moisture, sarcomere length, WBS, and cooking loss percentage. A split plot design (TRT, MC, and muscle

main effects; time, split plot factor effect; and their interactions) was used for pH decline. A split plot design (TRT, main effect; DAY, split plot factor effect; and interactions) was used for TBARS and hexanal. A split plot design (TRT and MUSCLE, main effects; time, split plot factor effect; and interactions) was used for oxygen percentage. A split plot design (TRT and MC, main effects; muscle, split plot factor effect; and interactions) was used for moisture fat-free, total pigment, ultimate pH, and bloom (a* L*). A split plot design (TRT and MC main effects; MUSCLE, split plot factor effect; and interactions) was used for colorimeter values (a*, L*) and reflectance estimators of the chemical states of myoglobin (OMb, MMb, DMb). In a stepwise manner, non-significant interactions ($P > 0.05$) greater than 2-way interactions were removed from the statistical model.

The SAS MIXED procedure (SAS 9.1.3 Service Pack 3, SAS Institute Inc., Cary, NC, USA) was used to determine significance ($P < 0.05$) in the models and when significance was found Adjusted Tukey-Kramer was used for mean separation. Letter assignment to individual means to enable statistical comparisons was achieved using the pdmix800 macro (Saxton, 1998).

Gel electrophoresis data were statistically analyzed (SPSS statistical package; SPPS Inc., Chicago, IL, USA) as a one-way ANOVA and difference between the carcass chilling treatment means were compared by Duncan's multiple range tests with $P < 0.05$ used as the level for significance.

3. Results and discussion

Details on the animals used and their assignment to TRT and MC can be found in Table 1. The main objective of this research was on the effects of RC application on commercially harvested cull dairy cows from the two lowest-valued MC. As such, the main effect results of carcass chilling treatment (TRT: CN, RC) and the interactions of TRT with other independent variables (MC, MUSCLE, DAY, TIME) have been discussed within this section. Type 3 tests of fixed effects can be found in Tables 2-4.

3.1. pH and temperature

Interactions were found ($P < 0.05$) for TRT x MC (Table 2), and TRT x MUSCLE (Table 2) associated with pH at various times postmortem. Despite a TRT x TIME interaction, RC resulted in a lower pH at each time postmortem (Fig. 1). Our results support those of Hunt et al. (2003) who found that vascular rinsed Hereford x Angus cross breed steers had a greater decline in pH during the first 4 h postmortem. In addition, Farouk, Price, and Salih (1992) reported lamb carcasses infused at a 10% solution volume (body wt.) containing 0.10% maltose, 0.21% glycerin, 0.14% phosphate and 0.23% dextrose had a faster rate of pH decline than non-infused carcasses. Yancey et al. (2002) suggested that the glucose in the rinse solution likely serves as an accessory source for anaerobic metabolism after glycogen reserves are depleted thereby facilitating pH decline. Interestingly, the LE cull cow carcasses had a lower ($P < 0.05$) pH than LI cow carcasses in the CN (Table 5). It would be anticipated that LE cows potentially would be less stress susceptible based on successfully maintaining a higher body condition (more muscle mass). As such the LE cows may have had greater glycogen reserves compared to LI cull cows at the time of harvest available for postmortem glycolysis. In the CN, the SM had a lower ($P < 0.05$) pH than LL, whereas there was no difference found in those from RC (Table 5). Perhaps differences exist in the vasculature between the SM and LL such that the SM was more effectively rinsed by RC thereby providing access to additional substrates for metabolism.

The carcass temperature decline graph of RC compared to CN is shown in Fig. 2. A TRT x MUSCLE interaction was found ($P < 0.05$, Table 2). RC resulted in a lower ($P < 0.05$, S.E. = 0.26) temperature in the SM (23.3 °C) compared to the SM in CN (24.6 °C) whereas there was not a temperature difference in the LL (13.1 °C). The ability to more effectively chill the SM using RC has commercial implications as the outer portion of the SM is known to be paler in color and is less color stable than the inner portion in part because slower chilling negatively affects metmyoglobin reducing ability and NAD levels (Sammel, Hunt, Kropf, Hachmeister, & Johnson, 2006). However, in a study by Hunt et al. (2003) with Hereford x Angus steers they indicated the rate of temperature decline was not affected by vascular chilling. Similarly, Murphy and Zerby (2004) did not report a temperature effect on lamb carcasses when infused (12.5 °C solution) with different added substrates to improve tenderness. Since our study used a colder rinse solution that might partially explain the difference observed.

3.2. Cooling loss, Warner-Bratzler shear force and sarcomere length

MC did influence cooking loss ($P < 0.05$), but no difference ($P > 0.05$) was observed between TRT (Table 6). CN LE cows had greater ($P < 0.05$) cooking losses compared to CN LI cows. Meat having a pH closer to the isoelectric point of the proteins would be expected to have a lower water holding capacity and as indicated the meat pH of CN LE was lower than CN LI.

Similar to other published research RC did not ($P > 0.05$) influence cooking loss. Fowler et al. (2017) reported that no treatment differences in cooking losses from lamb carcasses infused with RC. Similarly, Farouk and Price (1994) found there was no treatment effect on drip and cooking losses in samples refrigerated for 7 days. Kethavath et al. (2021) found that in pork, RC had a lower pH at 4 h postmortem than controls but did not affect cooking loss. In contrast, Mickelson and Claus (2020) reported that RC resulted in greater cooking loss (+1.7%) in the

Table 2
Type 3 tests of fixed effects associated with cull dairy cow carcass chilling treatment, time, and muscle on dependent variables (pH decline, temperature decline, percentage oxygen, TBARS, hexanal)[a].

Independent variables[b]	pH decline	Temperature decline	Independent variables[b]	Oxygen (%)	Independent variables[b]	TBARS	Hexanal
trt	<0.0001	0.0035	trt	0.3381	trt	0.0063	0.1665
time	<0.0001	<0.0001	musc	0.0091	day	0.7123	0.0344
trt*time	<0.0001	0.2911	trt*musc	0.0221	trt*day	0.1032	0.4281
mc	0.5489	<0.0001	time	<0.0001			
trt*mc	0.0008	0.3365					
musc	<0.0001	<0.0001					
trt*musc	0.0002	0.0014					
time*musc	0.0296	<0.0001					

[a] Dependent variables: pH and temperature decline early postmortem (1–24 h); percentage oxygen refers the oxygen level within the vacuum packaged meat, and measures of lipid oxidation (TBARS, Hexanal).

[b] Independent variables: trt = carcass chilling treatment (control, CN; Rinse & Chill®, RC), time = time postmortem, mc = marketing class (Lean, LE; Light, LI), musc = muscles (*Longissimus lumborum* = LL, *Semimembranosus* = SM, *Triceps brachii* = TB), day = storage days. pH and temperature decline used LL and SM. Percentage oxygen used Light LL and TB. TBARS and hexanal used Lean TB. In a stepwise manner, non-significant interactions ($P > 0.05$) greater than 2-way interactions were removed from the statistical model.

Table 3
Type 3 tests of fixed effects associated with cull dairy cow marketing class, carcass chilling treatment, and muscle for various physical and chemical dependent variables[a].

Independent variables[b]	Moisture fat free (%)	Total pigment (mg/g meat)	Ultimate pH	Purge (%)	EM (%)	Sarcomere length	WBS (N)	Cooking loss (%)	Bloom (1 h)	
									a*	L*
trt	<0.0001	0.0119	0.3051	0.2289	0.0003	<0.0001	<0.0001	0.8572	<0.0001	0.3617
mc	0.048	0.3153	0.0051	0.8299	0.0521	0.9620	0.1755	0.0474	0.0810	0.4756
trt*mc	0.4278	0.3001	0.6775	0.0215	0.2312	0.8524	0.3746	0.3754	0.1745	0.0582
musc	<0.0001	0.0815	<0.0001						0.7859	0.8108
trt*musc	<0.0001									

[a] Dependent variables: Moisture fat-free percentage (MFF); Total pigment; Ultimate pH; EM = expressible moisture percentage; sarcomere length, WBS (Warner Bratzler Shear); Cooking loss percentage; and Bloom (color after exposed to air for 1 h), a* (colorimeter redness); L* (colorimeter lightness).
[b] Independent variables: mc = marketing class (Lean, LE; Light, LI), trt = carcass chilling treatment (control, CN; Rinse & Chill®, RC), and musc = muscles (*Longissimus lumborum* = LL, *Triceps brachii* = TB). MFF, Total pigment, Ultimate pH, and Bloom used LL and TB. EM used TB.

Table 4
Type 3 tests of fixed effects associated with cull dairy cow marketing class, carcass chilling treatment, muscle, and display days on various meat color related dependent variables[a].

Independent variables[b]	a*	L*	OMb	MMb	DMb
trt	<0.0001	0.0414	0.0029	0.5011	0.4384
mc	0.2280	0.7160	0.8841	0.3204	0.8824
trt*mc	0.0015	0.0267	0.0016	0.1723	0.0500
musc	0.0458	<0.0001	0.8979	0.2343	0.0028
mc*musc	0.7439	0.9479	0.1794	0.0079	0.7803
trt*musc	0.5500	0.0003	0.0029	0.6544	0.4837
day	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
musc*day	0.0397	0.2800	0.0074	0.8590	0.0017

[a] Dependent variables: a* (colorimeter redness); L* (colorimeter lightness); reflectance spectrophotometric estimators of the chemical states of myoglobin: OMb = oxymyoglobin, MMb = metmyoglobin, and DMb = deoxymyoglobin.
[b] Independent variables: mc = marketing class (Lean, LE; Light, LI), trt = carcass chilling treatment (control, CN; Rinse & Chill®, RC), and musc = muscles (*Longissimus lumborum* = LL, *Triceps brachii* = TB). In a stepwise manner, non-significant interactions ($P > 0.05$) greater than 2-way interactions were removed from the statistical model.

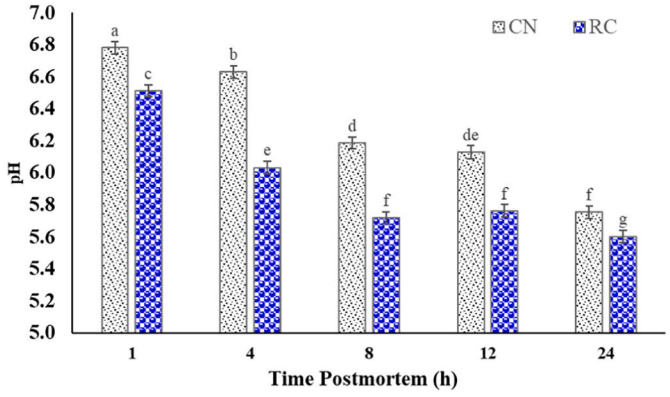

Fig. 1. Rinse & Chill® (RC) effect on pH decline (means pooled across marketing class: Lean- LE, Light- LI; muscle: *Longissimus lumborum* and *Semimembranosus*) of cull dairy cow carcasses compared to the control (CN). [a-g]Means with unlike letters are different ($P < 0.05$).

Longissimus lumborum of bison compared to the control.

After 7 days of aging, meat from both cow MC subjected to the RC treatment had lower shear force ($P < 0.05$) than CN (Fig. 3). RC infusion was able to reduce shear force by 58% and 55% for Lean and Light cows, respectively. These results support previous research on the RC effect on tenderness of various species. Farouk, Price, Salih, and Burnett (1992) observed that tenderness was improved by 13% using a similar solution on cull dairy cow carcasses. Using the same solution as the present study,

Table 5
Least square means for pH values associated with the pH decline period as affected by cull dairy cow carcass chilling treatment, marketing class, and muscle[1].

TRT	Marketing Class		Muscle	
	LE	LI	LL	SM
CN	6.25[b]	6.34[a]	6.40[a]	6.19[b]
RC	5.96[c]	5.89[c]	5.94[c]	5.91[c]
S.E.	0.027		0.027	

[a–c]Means within a marketing class or muscle with unlike superscripts letters are different ($P < 0.05$). Means represent the average value calculated over the five postmortem measurement times (1, 4, 8, 12, 24 h).
[1] Independent variables: Carcass chilling treatment (TRT): CN = control, RC = Rinse & Chill®; Marketing class: LE = Lean, LI = Light; Muscles: LL = *Longissimus lumborum*, SM = *Semimembranosus*.

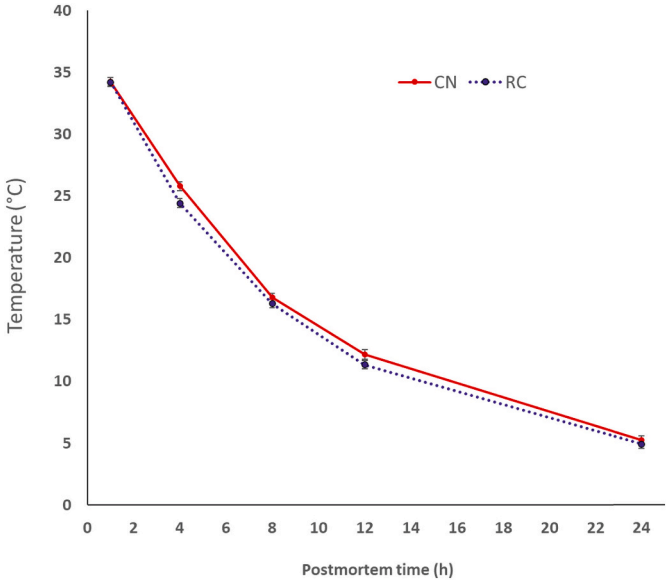

Fig. 2. Rinse & Chill® effect on muscle temperature decline (means pooled across marketing class, MC: Lean- LE, Light- LI; muscle: *Longissimus lumborum* and *Semimembranosus)* of cull dairy cow carcasses. Treatments: CN, Control; RC, Rinse & Chill®.

Mickelson and Claus (2020) found a 24% reduction in toughness in bison *Longissimus lumborum* steaks. Fowler et al. (2017) using the same solution composition but at 14 °C, demonstrated a 34% shear force reduction compared to the control (32.4 N) for lamb *Longissimus lumborum* chops.

According to Farouk, Price, and Salih (1992) the lower shear force

Table 6
Least square means for ultimate pH, sarcomere length, moisture-fat free percentage, expressible moisture percentage, purge percentage, and cook loss affected by marketing class, carcass chilling treatment, and muscles from cull dairy cows[1].

Independent variable	Ultimate pH	Sarcomere length in LL (μm)	Moisture-fat free (%)		Expressible moisture (%) in TB	Purge (%) in LL		Cook loss (%) in LL
			LL	TB		LE	LI	
CN	5.63[a]	1.42[b]	76.67[b]	76.87[b]	5.75[b]	1.04[a]	0.74[ab]	23.49[a]
RC	5.68[a]	1.80[a]	76.60[b]	78.54[a]	8.65[a]	0.62[b]	0.87[ab]	23.66[a]
S.E.	0.0344	0.0363	0.179		0.679	0.118		0.650
Lean	5.60[b]							25.51[a]
Light	5.72[a]							22.64[b]
S.E.	0.0343							0.650
LL	5.57[b]							
TB	5.75[a]							
S.E.	0.0342							

[a,b]Means within an independent variable with unlike superscript letters are different.
[1] Independent variables: Marketing class (LE = Lean, LI = Light); Carcass chilling treatment (TRT): CN = control, RC = Rinse & Chill®; Muscles: LL = *Longissimus lumborum*, TB = *Triceps brachii*.

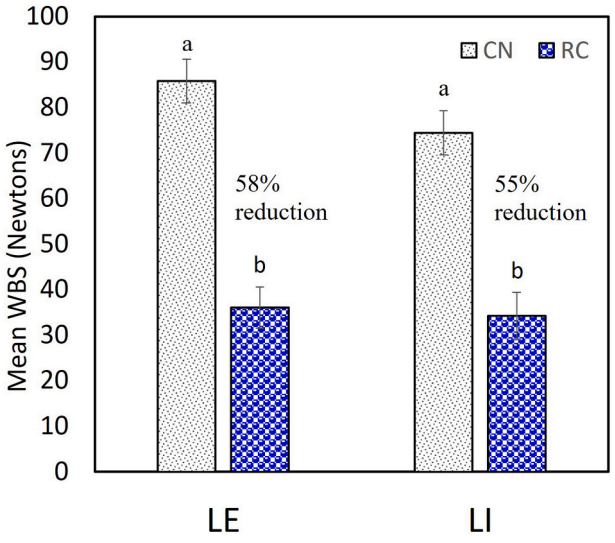

Fig. 3. Rinse & Chill® effect on Warner-Bratzler shear force (WBS) in cull dairy cow carcass *Longissimus lumborum* steaks aged 7 d postmortem; Treatments (TRT): CN, Control; RC, Rinse & Chill®. Marketing Class (MC) represented by LE (Lean) and LI (Light). Means with unlike letters are different ($P < 0.05$, S.E. 5.0; TRT x MC, $P > 0.05$).

can be explained by the enhancement of proteolytic activity caused by the ingredients present in the solution. Additionally, the phosphate blend in the solution could contribute to the dissociation of actomyosin, thus decreasing toughness (Dalrymple & Hamm, 1973; Trout & Schmidt, 1984). Moreover, the rapid pH decline previously reported was likely associated with accelerated glycolysis rate, thus completing rigor mortis earlier and enhancing the aging process (Farouk & Price, 1994; Farouk, Price, & Salih, 1992).

The application of RC on cull dairy cow carcass did demonstrate its ability to improve tenderness by decreasing shear force below an unacceptable/acceptable threshold of 48 N (Hopkins, Hegarty, Walker, & Pethick, 2006) regardless of live animal variability, which often has been observed in these animals.

TRT did affect sarcomere length (Table 6) of the LL. Sarcomeres for RC samples were 26.7% longer ($P < 0.05$) than those from CN, which likely helps to explain the differences in shear force. The CN or RC carcasses were not electrically stimulated. Electrical stimulation is known to accelerate postmortem glycolysis and prevent cold shortening (Adeyemi & Sazili, 2014). Depending on the temperature and pH early postmortem can affect muscle shortening which affects tenderness. During rigor, both warm- and cold-shortening can occur (Tornberg, 1996). Differences in the temperature range that results in minimum shortening have been reported. Locker and Hagyard (1963) reported minimum muscle shortening between 14 and 19 °C. Tornberg (1996) reported minimum muscle shortening between 10 and 15 °C. In general, when the pH is above 6 and the temperature is below 15 °C creates the conditions likely to result in cold shortening (Aberle et al., 2012). At 12 h postmortem the pH in the LL of the CN was 6.3 and RC was 5.8 while the temperature was less than 10 °C. As such, the CN likely exhibited cold-induced shortening whereas the more rapid glycolytic activity induced by RC enabled the formation of permanent crossbridges between actin and myosin before cold-induced shortening could occur. Similar results were reported for bison (1.77 control vs. 1.80 μm RC; Mickelson & Claus, 2020). In contrast, Fowler et al. (2017) did not observe a RC effect on sarcomere length on lamb *Longissimus lumborum* samples.

3.3. Ultimate pH, expressible moisture (EM) and purge

Ultimate pH was not affected ($P > 0.05$, Table 6) by TRT. Unlike the difference found associated with the pH decline, the lack of a difference here likely was associated with the pH of the CN declining further after the muscles were excised associated with the carcasses not being electrically stimulated whereas RC was able to accelerate glycolysis. Our results support Mickelson and Claus (2020) who reported chilling method did not alter the ultimate pH in bison meat. Ultimate pH of the LE was lower ($P < 0.05$) than LI. In addition, the LL had a lower ($P < 0.05$) ultimate pH than TB.

For WHC, RC had greater ($P < 0.05$) expressible moisture than CN (Table 6). Farouk and Price (1994) reported that the shoulder of infused beef carcasses had greater percentage moisture than non-infused. Since they did not determine WHC it is not known if the greater moisture level was indicative of more expressible moisture. A MC x TRT interaction was found with purge loss (Tables 3 and 6) such that the LE *Longissimus lumborum* CN had greater purge loss than LE RC. The lower pH of CN may have contributed to this effect. In contrast, no difference in purge was observed in LI *Longissimus lumborum*.

3.4. Moisture fat-free (MFF), and total pigments

No differences were found in fat % ($P > 0.05$) between chilling treatments (CN = 2.97%, RC = 2.79%; S.E., 0.205). The overall carcass chilling treatment mean for fat was 2.87% (S.E., 0.521). There was a difference in fat % related to MC (LE: 3.13 and LI: 2.63; $P < 0.05$; S.E., 0.158). Muscles differed in fat % (LL: 3.20; TB: 2.50; $P < 0.05$; S.E., 0.158). The results of MFF showed a TRT x MUSCLE interaction (Table 3) in that RC TB resulted in a greater MFF in the TB compared to CN (Table 6). However, TRT did not affect MFF in the LL. Farouk and Price (1994) reported that infused carcasses retained more moisture in the following order: shoulder > loin > leg. The higher retained moisture

in the shoulder may be due to the infusion site being closer to the shoulder muscle or due to the downward movement of the solution during the rinsing process (Farouk, Price, Salih, & Burnett, 1992). The position of the carcass during infusion process could explain differences in moisture within the animal, as the beef carcass infused when hanging vertically on the rail, might impact the ability of the solution to equally reach all muscles especially in the hind leg due to gravitation forces on the solution.

The amount of total pigments was affected by TRT (RC: 9.08; CN: 8.38 mg/g meat; $P < 0.05$; S.E., 0.247). Perhaps the greater MFF partially supports more myoglobin as this pigment is associated with the sarcoplasmic fluid (Aberle et al., 2012). In contrast to our study, Hunt et al. (2003) cited a thesis research (Schoenbeck, 1998) that found no difference in the pigment concentration (myoglobin and hemoglobin) between infused and noninfused beef carcasses. There was not a difference ($P > 0.05$) in pigment concentration between the LL and TB. Typically muscles that contain a higher percentage of red muscle fibers would be expected to contain more myoglobin (Aberle et al., 2012). Since older animals are expected to also have more myoglobin, this might partially explain the lack of difference between these two muscles (Aberle et al., 2012).

3.5. Bloom and display color

After 1 h of bloom, RC was more red (CIE a*, 22.10) than CN (19.92; $P < 0.05$; S.E., 0.2663). Associated with bloom, lightness (L*) was not affected ($P > 0.05$, data not shown) by TRT or MC. For display there was a TRT*MC interaction found for CIE a* (Table 4). Nevertheless, RC was redder than CN in both MC (Table 7). In terms of MC, no difference was found in CN, but the LE class was redder than the LI class in the RC group. Associated with CIE L* there were interactions in TRT*MC and TRT*MUSCLE (Table 4). In LE, RC was lighter ($P < 0.05$) than CN, but no difference ($P > 0.05$) in lightness was found between TRT in the LI class (Table 7). RC resulted in lighter TB than in the CN, but no carcass chilling treatment difference was found in the LL (Table 7).

Associated with oxymyoglobin, interactions were found with TRT*MC and TRT*MUSCLE (Table 4). No differences ($P > 0.05$) associated with TRT or their interactions were found for MMb and DMb (Table 4). RC resulted in more ($P < 0.05$) oxymyoglobin in the LE than in the CN LE, but TRT did not affect oxymyoglobin in the LI class (Table 7). RC had greater oxymyoglobin in the TB than in the CN TB, but treatment did not affect oxymyoglobin in LL (Table 7).

In general, our results confirmed those of Mickelson and Claus (2020) who found that with PVC and VAC packaged ground bison RC resulted in redder and lighter product than CN. However, in PVC ground bison no differences were found in the chemical states of myoglobin. In contrast, in vacuum packaged ground bison they reported more deoxymyoglobin, and less metmyoglobin. Numerous factors can affect meat color including myoglobin concentration, enzymatic reducing ability, and presence of antimicrobials. Farouk and Price (1994) suggested that dilution of pigments in the muscles would affect color by resulting in lighter colored muscles in infused lamb carcasses associated with greater light scattering. Ledward (1985) reported that at a lower temperature during post-mortem glycolysis influences red color (higher CIE a* values) by preserving the activity of metmyoglobin-reducing enzymes. Inclusion of polyphosphates in the RC solution also might have provided some anti-microbial activity and improved color found in the dairy cow samples. Moreira, Connolly, and Claus (2018) reported that lean cows which are infused with RC solution showed a decline in aerobic plate counts on the carcass surface compared to controls.

Table 7
Least square means for colorimeter values (CIE a*, CIE L*), reflectance estimator of oxymyoglobin, and residual oxygen percentage as affected by carcass chilling treatment, marketing class, and muscles[1].

TRT	CIE a*(Redness)		CIE L*(Lightness)		CIE L*(Lightness)	
	LE	LI	LE	LI	LL	TB
CN	13.62^{c}	14.07^{c}	40.65^{b}	41.00^{ab}	40.76^{b}	40.89^{b}
RC	16.87^{a}	15.86^{b}	41.62^{a}	40.97^{ab}	40.46^{b}	41.98^{a}
S.E.	0.266		0.220		0.219	

TRT	Oxymyoglobin		Oxymyoglobin		Residual oxygen (%)	
	LE	LI	LL	TB	LL	TB
CN	1.76^{c}	1.81^{b}	1.81^{b}	1.75^{c}	2.58^{a}	2.50^{a}
RC	1.86^{a}	1.81^{b}	1.81^{b}	1.86^{a}	2.90^{a}	1.69^{b}
S.E.	0.207		0.206		0.324	

[a-c]Means within a dependent variable with unlike superscript letters are different ($P < 0.05$).

[1] Independent variables: Carcass chilling treatment: CN = control, RC = Rinse & Chill®; MC: marketing class (LE = Lean; LI = Light); Muscles: LL = *Longissimus lumborum*, TB = *Triceps brachii*. Dependent variables: CIE a* (redness), CIE L* (lightness), and oxymyoglobin (percentage reflectance at 610 nm divided by percentage reflectance at 525 nm); larger values indicate greater redness, lightness, and oxymyoglobin, respectively.

3.6. Oxygen consumption

Oxygen consumption was greater ($P < 0.05$) in the RC TB than CN as the amount of residual oxygen was lower (Table 7). The percentage of residual oxygen decreased ($P < 0.05$) with time (Fig. 4). These results suggest that RC preserved mitochondrial respiration in the TB. However, treatment did not affect ($P > 0.05$) oxygen consumption in the LL (Table 7). The TB did have greater ($P < 0.05$) oxygen consumption than LL in the RC treatment. This difference likely is related to the expectation that the TB muscle would have more of Type I fibers and greater mitochondria concentration than in the LL. Ramanathan, Mancini, and Konda (2010) reported the mitochondria have the ability to consume oxygen in postmortem muscle.

The greater oxygen consumption measured in the VAC package first appears contradictory to the greater oxymyoglobin content in the PVC packaged during display. This is understandable because oxygen at very low partial pressure is known to promote metmyoglobin formation (Mancini & Hunt, 2005). In addition, deoxygenation under adequate reducing conditions would lead to deoxymyoglobin formation. Formation of oxymyoglobin on the surface involves complex reactions as influenced by numerous factors including pH, film permeability to oxygen, mitochondria respiration, and presence of bacteria. Greater oxygen consumption likely infers increased aerobic metabolism through the TCA cycle which would be expected to generate more NADH that helps maintain the heme iron in the ferrous state. The ferrous state enables the

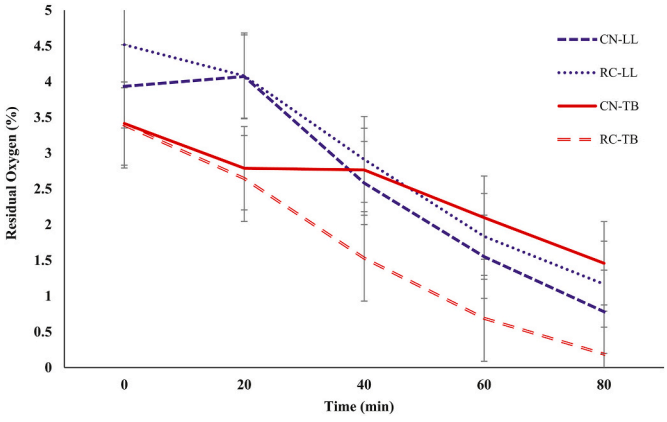

Fig. 4. Rinse & Chill® effect on oxygen consumption (residual oxygen %) in cull dairy cow carcasses (Light marketing class); Treatments: CN, Control; RC, Rinse & Chill®. Muscles: LL = *Longissimus lumborum*, TB = *Triceps brachii*. S.E. = 0.592.

formation of deoxymyoglobin and oxymyoglobin (Aberle et al., 2012). With sufficient penetration depth of oxygen and adequate reducing ability, increased levels of oxymyoglobin would be expected.

3.7. Lipid oxidation

TBARS and hexanal samples were analyzed only in the LE marketing class (Table 2). RC had lower ($P < 0.05$) TBARS than CN (Table 8). TRT did not influence ($P > 0.05$) hexanal content. The decreased lipid oxidation as found in TBARS by RC perhaps was associated with the removal of more blood thereby lowering the heme-iron content in the muscle. Erazo-Castrejón, Zhang, Mickelson, Claus, and Yin (2019) reported the RC method removed 40% more blood from the muscle compared to the conventional method. Aisen and Listowsky (1980) reported that hemoglobin and smaller amounts in myoglobin accounts for about two-thirds of body iron. The remainder comprises components in various iron-containing enzymes and in the form of transport protein transferrin. Kanner (1994) suggested that the stability of a muscle food product will depend on the involvement of metal ions (iron) in the process which acts as proxidant. Cheng and Ockerman (2003) indicated inclusion of sodium tripolyphosphate (an iron chelator) in precooked roast beef, could be an effective strategy to reduce the formation of TBARS. TBARS did not ($P > 0.05$) increase with storage time (Table 8). Hexanal content was lower ($P < 0.05$) on day 10 than on day 1. Aldehydes produced during lipid oxidation can form adducts with proteins and potentially cause lower hexanal values.

3.8. Myofibril proteins

Analyzing the myofibril samples using 12% gels provided visualization of the Troponin-T degradation and confirmed the presence of a 30-kDa fragment. Troponin-T, one of the myofibrillar proteins, is well known to degrade during postmortem aging of meat (Ho, Stromer, & Robson, 1994). The degradation of troponin-T and accumulation of the 30-kDa fragment has often been used an indicator of the overall postmortem proteolysis associated with aging animal muscles (Huff Lonergan, Zhang, & Lonergan, 2010; Lawrence, Dikeman, Stephens, Obuz, & Davis, 2004). SDS-PAGE results demonstrated that the abundance of the 30-kDa troponin-T fragment in six of the nine samples from RC processed carcasses (RC; 3, 4, 6, 7, 8, 9; Fig. 5a and b). In contrast, the amount of 30-kDa troponin-T fragment was only readily apparent in two CN carcasses (5 and 7). However, overall the level of proteolysis based on the accumulation of the 30-kDa troponin-T fragment was not affected by TRT ($P = 0.199$, Fig. 5c).

Although meat tenderness was increased by RC based on shear force, using the accumulation of a 30-kDa troponin-T fragment was not useful as an indicator of meat tenderization (proteolysis). Fowler et al. (2017) concluded from their lamb study that the mechanism by which RC increases tenderness was still unknown. What likely was the most significant tenderization mechanism, specific to our research, was the apparent inhibition of muscle shortening by RC. The CN samples had much shorter sarcomeres than RC samples. Carcasses were not electrical stimulated which if used would have accelerated postmortem pH decline and reduced the likelihood of cold shortening occurring. In addition, muscles were excised at 24 h which may not have provided enough time for rigor to be complete prior to their removal associated with the controls.

Table 8
Least square means for thiobarbituric acid reactive substances (TBARS) and hexanal on ground, cooked, vacuum packaged beef *Triceps brachii* from lean (LE) marketing class cull dairy cows as affected by carcass chilling treatment and storage time (4 °C)[1].

Days	TBARS (mg MDA/kg meat)			Hexanal (μmol/kg meat)		
	1	10	Overall	1	10	Overall
CN	1.09	1.37	1.23[a]	11.48	7.38	8.72[a]
RC	0.92	0.74	0.83[b]	5.98	4.38	6.10[a]
Overall	1.01[x]	1.06[x]		9.43[x]	5.40[y]	

[xy]Overall day means within a row and dependent variable with unlike letters are different ($P < 0.05$). S.E.: TBARS (0.103); Hexanal (1.516).
[ab]Overall TRT: means within a column and dependent variable with unlike letters are different ($P < 0.05$). S.E.: TBARS (0.103); Hexanal (1.510).
[1] Independent variables: Carcass chilling treatment: CN = control, RC = Rinse & Chill®.

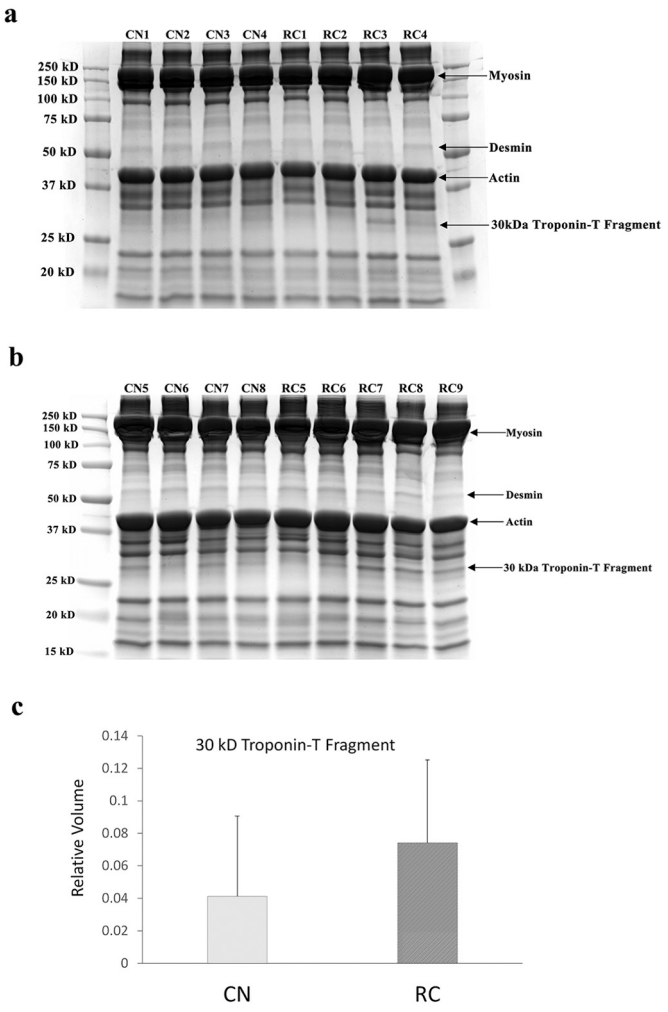

Fig. 5. Gel electrophoresis of cull cow samples (5a, 5b) from two different chilling treatments (CN, control; RC, Rinse & Chill®) presented to visualize Troponin-T degradation relative to the 30-kDa fragment accumulation. Only samples from the Lean marketing class were used. Each gel lane represents an individual beef animal from a corresponding chilling treatment. Troponin-T degradation 30-kDa fragment relative volume (5c) between CN and RC was not different ($P > 0.05$; S.E. = 0.024).

4. Conclusions

Rinse & Chill® technology has commercial potential for its ability to accelerate the pH decline and improve meat tenderness. The accelerated drop in pH may avoid the risk of cold-induced shortening (toughening) that produces tenderness issues, especially in cull dairy cow carcass that have limited external fat. For packing plants that do not have electrical stimulation and would find value in fabricating carcasses as soon as 24 h postmortem, Rinse & Chill® appears to accelerate rigor mortis development thereby limiting muscle shortening. Rinse & Chill® has the ability to improve color by enhancing mitochondrial activity and

oxymyoglobin formation. Muscles or areas of a muscle that are deep within the carcass may realize a color benefit from Rinse & Chill® relative to more effective chilling compared to conventional chilling. This technology also has potential to decrease lipid oxidation by removing prooxidant metal ions associated with heme pigments. Rinse & Chill® can be used on beef from Lean and Light cull dairy cow marketing classes and packers can expect similar meat quality benefits from the process.

Declaration of competing interest

None.

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References

Aberle, E. D., Forrest, J. C., Gerrard, D. E., & Mills, E. W. (2012). *Principles of meat science* (5th ed.). Dubuque, Iowa: Kendall/Hunt Publishing Co.

Adeyemi, K. D., & Sazili, A. Q. (2014). Efficacy of carcass electrical stimulation in meat quality enhancement: A review. *Asian-Australasian Journal of Animal Sciences, 27*(3), 447–456. https://doi.org/10.5713/ajas.2013.13463

Aisen, P., & Listowsky, I. (1980). Iron transport and storage proteins. *Annual Review of Biochemistry, 49*, 357–393.

Alende, M., Lagreca, V., Pordomingo, A. J., Pighin, D., Grigioni, G., Carduza, F., … Sancho, A. M. (2014). Effects of transport, lairage and ageing time on stress indicators and on instrumental and sensory quality of beef from steers. *Archivos de Medicina Veterinaria, 46*(2), 217–227. https://doi.org/10.4067/S0301-732X2014000200007

American Meat Science Association. (2012). *Meat color measurement guidelines*. M.C. Hunt & A. King (Eds.). American Meat Science Association, Publisher http://www.meatscience.org.

American Meat Science Association. (2015). *Research guidelines for cookery, sensory evaluation, and instrumental tenderness measurements of meat* (2nd ed.). American Meat Science Association, Publisher (version 1.0) (T. L. Wheeler, L. S. Papadopoulos, R. K. Miller, Eds.).

Becker, C. A., Collier, R. J., & Stone, A. E. (2020). Invited review: Physiological and behavioral effects of heat stress in dairy cows. *Journal of Dairy Science, 103*(8), 6751–6770. https://doi.org/10.3168/jds.2019-17929

Cheng, J.-H., & Ockerman, H. W. (2003). Effect of phosphate with tumbling on lipid oxidation of precooked roast beef. *Meat Science, 65*, 1353–1359.

Couvreur, S., Le Bec, G., Micol, D., & Picard, B. (2019). Relationships between cull beef cow characteristics, finishing practices and meat quality traits of longissimus thoracis and rectus abdominis. *Foods, 8*(4), 1–16. https://doi.org/10.3390/foods8040141

Cross, H. R., West, R. L., & Dutson, T. R. (1981). Comparison of methods for measuring sarcomere length in beef semitendinosus muscle. *Meat Science, 5*, 261–266.

Dalrymple, R. H., & Hamm, R. (1973). A method for the extraction of glycogen and metabolites from a single muscle sample. *International Journal of Food Science & Technology, 8*(4), 439–444.

Edwards-Callaway, L. N., Walker, J., & Tucker, C. B. (2019). Culling decisions and dairy cattle welfare during transport to slaughter in the United States. *Frontiers in Veterinary Science, 5*, 1–5.

Erazo-Castrejón, S. V., Zhang, W., Mickelson, M. A., Claus, J. R., Yin, J., & Richards, M. P. (2019). Quantification of hemoglobin and myoglobin in pork muscle: Effect of rinse & chill technology on blood removal. *Meat and Muscle Biology, 1*(3), 110. https://doi.org/10.221751/rmc2017.105

Fabris, T. F., Laporta, J., Skibiel, A. L., Dado-Senn, B., Wohlgemuth, S. E., & Dahl, G. E. (2020). Effect of heat stress during the early and late dry period on mammary gland development of Holstein dairy cattle. *Journal of Dairy Science, 103*(9), 8576–8586. https://doi.org/10.3168/jds.2019-17911

Farouk, M. M., & Price, J. F. (1994). The effect of post-exsanguination infusion on the composition, exudation, color and post-mortem metabolic changes in lamb. *Meat Science, 38*, 477–496.

Farouk, M. M., Price, J. F., & Salih, A. M. (1992). Post-exsanguination infusion of ovine carcasses: Effect on tenderness indicators and muscle microstructure. *Journal of Food Science, 57*, 1311–1315.

Farouk, M. M., Price, J. F., Salih, A. M., & Burnett, R. J. (1992). The effect of postexsanguination infusion of beef on composition, tenderness, and functional properties. *Journal of Animal Science, 70*, 2773–2778.

Feirtag, J. M., & Pullen, M. M. (2003). Novel intervention for the reduction of bacteria on beef carcasses. *Food Protection Trends, 23*(7), 558–562.

Fowler, S. M., Claus, J. M., & Hopkins, D. L. (2017). The effect of applying a rinse and chill procedure to lamb carcases immediately post-death on meat quality? *Meat Science, 134*, 124–127.

Hadley, G. L., Wolf, C. A., & Harsh, S. B. (2006). Dairy cattle culling patterns, explanations, and implications. *Journal of Dairy Science, 89*, 2286–2296.

Ho, C. Y., Stromer, M. H., & Robson, R. M. (1994). Identification of the 30 kDa polypeptide in post mortem skeletal muscle as a degradation product of troponin-T. *Biochimie, 76*, 369–375.

Hopkins, D. L., Hegarty, R. S., Walker, P. J., & Pethick, D. W. (2006). Relationship between animal age, intramuscular fat, cooking loss, pH, shear force and eating quality of aged meat from sheep. *Australian Journal of Experimental Agriculture, 46*, 879–884.

Huang, M., Huang, F., Xu, X., & Zhou, G. (2009). Influence of caspase3 selective inhibitor on proteolysis of chicken skeletal muscle proteins during post mortem aging. *Food Chemistry, 115*, 181–186.

Huff Lonergan, E., Zhang, W., & Lonergan, S. M. (2010). Biochemistry of postmortem muscle - Lessons on mechanisms of meat tenderization. *Meat Science, 86*, 184–195.

Hunt, M. C., Schoenbeck, J. J., Yancey, E. J., Dikeman, M. E., Loughin, T. M., & Addis, P. B. (2003). Effects of postexsanguination vascular infusion of carcasses with calcium chloride or a solution of saccharides, sodium chloride, and phosphates on beef display - Color stability. *Journal of Animal Science, 81*, 669–675.

Kanner, J. (1994). Oxidative processes in meat and meat products: Quality implications. *Meat Science, 36*, 169–189.

Kethavath, S. C., Hwang, K.-E., Mickelson, M. A., Campbell, R. E., Richards, M. P., & Claus, J. R. (2021). Vascular infusion with concurrent vascular rinsing on color, tenderness, and lipid oxidation of hog meat. *Meat Science, 174*, 1–7. https://doi.org/10.1016/j.meatsci.2020.108409

Lawrence, T. E., Dikeman, M. E., Stephens, J. W., Obuz, E., & Davis, J. R. (2004). In situ investigation of the calcium-induced proteolytic and salting-in mechanisms causing tenderization in calcium-enhanced muscle. *Meat Science, 66*(1), 69–75.

Ledward, D. A. (1985). Post-slaughter influences on the formation of metmyoglobin in beef muscles. *Meat Science, 15*, 149–171.

Li, X., Zhang, D., Ijaz, M., Tian, G., Chen, J., & Du, M. (2020). Colour characteristics of beef longissimus thoracis during early 72 h postmortem. *Meat Science, 170*, 108245. https://doi.org/10.1016/j.meatsci.2020.108245

Locker, R. H., & Hagyard, C. J. (1963). A cold shortening effect in beef muscles. *Journal of the Science of Food and Agriculture, 14*, 787–793.

Mancini, R. A., & Hunt, M. C. (2005). Current research in meat color. *Meat Science, 71*, 100–121.

Mickelson, M. A., & Claus, J. R. (2020). Carcass chilling method effects on color and tenderness of bison meat. *Meat Science, 161*, 1–5. https://doi.org/10.1016/j.meatsci.2019.108002

Moreira, L. C., Connolly, C., & Claus, J. R. (2018). Vascular rinse & chill effects on meat quality and shelf life of beef, 64th International Congress of Meat Science and Technology, Melbourne, Australia www.iastatedigitalpress.com/mmb/article/id/8886/.

Moreira, L. C., Rosa, J. M., & Schaefer, D. (2020). Get more value from cull cows. *Hoard's Dairyman, 165*, 220–221. April 10.

Murphy, M. A., & Zerby, H. N. (2004). Prerigor infusion of lamb with sodium chloride, phosphate, and dextrose solutions to improve tenderness. *Meat Science, 66*, 343–349.

Nicholson, J. D. W. (2008). *National market cow and bull beef quality audit-2007: A survey of producer-related defects*. Master of Science Thesis. Texas A&M University (p. 166).

Park, S. Y., Undeland, I., Sannaveerappa, T., & Richards, M. P. (2013). Novel interactions of caffeic acid with different hemoglobins: Effects on discoloration and lipid oxidation in different washed muscles. *Meat Science, 95*(1), 110–117.

Peel, D. S., & Doye, D. G. (2017). Cull cow grazing and marketing opportunities. Oklahoma Cooperative Extension Service, Extension fact sheet AGEC-613 https://extension.okstate.edu/fact-sheets/cull-cow-grazing-and-marketing-opportunities.html.

Ramanathan, R., Mancini, R. A., & Konda, M. K. R. (2010). Effect of lactate enhancement on myoglobin oxygenation of beef longissimus steaks overwrapped in pvc and stored at 4C. *Journal of Muscle Foods, 21*, 669–684.

Romans, J. R., Costello, W. J., Jones, K. W., Carlson, C. W., & Ziegler, P. T. (1985). *The meat we eat*. Danville, Illinois, USA: The Interstate Printers and Publishers, Inc.

Sammel, L. M., Hunt, M. C., Kropf, D. H., Hachmeister, K. A., & Johnson, D. E. (2006). Comparison of assays for metmyoglobin reducing ability in beef inside and outside semimembranosus muscle. *Journal of Food Science, 67*, 978–984.

Saxton, A. (1998). *A macro for converting mean separation output to letter groupings in Proc Mixed*. Proceedings of the 23rd SAS Users Group International, 22-25 March, Nashville, Tennessee, USA (pp. 1243–1246).

Schoenbeck, J. J. (1998). *Effects of cardiovascular infusion on beef carcass appearance and color and chemical traits of five muscles*. M.S. Thesis. Kansas State University.

Stelzleni, A. M., Patten, L. E., Johnson, D. D., Calkins, C. R., & Gwartney, B. L. (2007). Benchmarking carcass characteristics and muscles from commercially identified beef and dairy cull cow carcasses for Warner-Bratzler shear force and sensory attributes. *Journal of Animal Science, 85*, 2631–2638.

Therkildsen, M., Stolzenbach, S., & Byrne, D. V. (2011). Sensory profiling of textural properties of meat from dairy cows exposed to a compensatory finishing strategy. *Meat Science, 87*, 73–80.

Tornberg, E. (1996). Biophysical aspects of meat tenderness. *Meat Science, 43*, S175–S191.

Trout, G. R., & Schmidt, G. R. (1984). Utilization of phosphates in meat products. *Annual Reciprocal Meat Conference of the American Meat Science Association Proceedings, 36*, 24–27.

USDA. (2018). Health and management practices on U.S. dairy operations, 2014. In *National animal health monitoring system*. U.S. Department of Agriculture, Animal and

Plant Health Inspection Service. https://www.aphis.usda.gov/animal_health/nahms/dairy/downloads/dairy14/Dairy14_dr_PartIII.pdf.

USDA. (2020). *Livestock slaughter 2019 summary*. National Agricultural Statistics Service: U.S. Department of Agriculture. https://www.nass.usda.gov/.

Weigel, K. A., Palmer, R. W., & Caraviello, D. Z. (2003). Investigation of factors affecting voluntary and involuntary culling in expanding dairy herds in Wisconsin using survival analysis. *Journal of Dairy Science, 86*, 1482–1486.

Witte, V. C., Krause, G. F., & Bailey, M. E. (1970). A new extraction method for determining 2-thiobarbituric acid values of pork and beef during storage. *Journal of Food Science, 35*, 582–585.

Woerner, D. R. (2010). *Beef from market cows*. Centennial, CO, USA: Cattlemen's Beef Board and National Cattlemen's Beef Association. White Paper 7101.

Yancey, E. J., Dikeman, M. E., Addis, P. B., Katsanidis, E., & Pullen, M. (2002). Effects of vascular infusion with a solution of saccharides; sodium chloride; phosphates; and vitamins C, E, or both on carcass traits, Warner-Bratzler shear force, and palatability traits of steaks and ground beef. *Journal of Animal Science, 80*, 1904–1910.