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Effect of early postmortem enhancement of calcium lactate/phosphate on quality attributes of beef round muscles under different packaging systems

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ABSTRACT

The objective was to determine the influence of calcium lactate/phosphate enhancement on quality of beef round cuts in high-oxygen modified atmosphere (HiOx-MAP; 80% O_2/20% CO_2). *Mm. semimembranosus* (SM), *semitendinosus* (ST), and *adductor* (AD) were divided and assigned to water-injected control (CON), 3 mM phosphate (STP), or 200 mM calcium lactate/3 mM phosphate (CAL/STP) treatments at 24 h postmortem. Steaks ($n = 10$) were vacuum packaged (VAC) and stored for 9 days, then displayed for 7 days in VAC or HiOx-MAP. Lipid oxidation, pH, surface color, star probe, and sensory characteristics were evaluated. HiOx-MAP resulted in greater lipid oxidation, more discoloration, and decreased sensory quality of steaks ($P < 0.05$) compared to VAC. However, CAL/STP enhancement significantly reduced lipid oxidation of all steaks, decreased ST and SM star probe values, and improved tenderness of HiOx-MAP packaged AD and SM ($P < 0.05$). Results suggest that CAL/STP enhancement has beneficial effects on lipid stability and sensory attributes of beef round cuts under HiOx-MAP.

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1. Introduction

Overall appearance of retail meat cuts is the primary factor that consumers consider when determining meat freshness and making purchasing decisions. In combination with meat color, tenderness and flavor are considered to be among the most important quality attributes by consumers (Faustman & Cassens, 1991; Platter et al., 2003). Thus, maintaining a bright-red color, improving tenderness, and enhancing flavor and juiciness are critical factors that need to be addressed as they significantly affect consumers' repeat purchases of fresh meat.

Modified atmosphere packaging (MAP) systems with a high oxygen (80%; HiOx-MAP) level are used in retail meat markets because the bright red color of meat in this packaging system attracts consumers. However, there is compelling evidence that high oxygen levels are likely to increase incidence of oxidative changes in the meat and consequently accelerate surface discoloration and decrease desirable flavors of the meat (Grobbel, Dikeman, Hunt, & Milliken, 2008b; Kim, Huff-Lonergan, Sebranek, & Lonergan, 2010b; Kim, Stuart, Rosenvold, & Maclennan, 2013; Lund, Lametsch, Hviid, Jensen, & Skibsted, 2007; Seyfert et al., 2005). Moreover, decreased meat tenderness under HiOx-MAP storage has also been reported (Kim, Huff-Lonergan, Sebranek, & Lonergan, 2010b; Kim, Stuart, Black, & Rosenvold, 2012; Lund et al., 2007). This negative effect of HiOx-MAP can be attributed to the intermolecular cross-linking and aggregation of myosin with other high molecular weight proteins such as titin (Kim, Huff-Lonergan, Sebranek, & Lonergan, 2010b), since myosin is susceptible to oxidation (Lund, Heinonen, Baron, & Estévez, 2011; Morzel, Gatellier, Sayd, Renerre, & Laville, 2006).

The addition of calcium to steaks is known to result in increased protein degradation and subsequent increased tenderness as a result of activation of endogenous calpain enzymes (Busch et al., 1972). This effect was shown over 20 years ago with the use of calcium chloride (Whipple & Koohmaraie, 1993). However, the caveat to calcium chloride injection is that it results in greater oxidation and discoloration of the steak (Harris, Huff-Lonergan, Lonergan, Jones, & Rankins, 2001; Wheeler, Koohmaraie, & Shackelford, 1996). An alternative to calcium chloride could be calcium lactate. Lactate enhancement (commonly sodium or potassium lactate) is extensively practiced in fresh meat because it increases juiciness, flavor, tenderness, and color stability (Kim et al., 2006; Lawrence, Dikeman, Hunt, Kastner, & Johnson, 2004; Maca, Miller, Bigner, Lucia, & Acuff, 1999; Ramanathan, Mancini, & Dady, 2011). A strong antioxidant capacity of calcium lactate in fresh meat has been reported (Kim, Huff-Lonergan, & Lonergan, 2012; Kim et al., 2009). However, some studies have found that calcium lactate alone does not improve color stability (Kim, Huff-Lonergan, & Lonergan,

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2012; Seyfert, Hunt, Lundesjo Ahnstrom, & Johnson, 2007), suggesting that another antioxidant, such as phosphate, may be required for maximum effectiveness and improved quality. Beef loins enhanced with calcium lactate and phosphate maintained the most stable red color with substantially minimized lipid oxidation, even under highly oxidizing conditions (HiOx-MAP with 2.4 kGy irradiation) when compared with control loins or loins enhanced with phosphate alone (HiOx-MAP with no irradiation treatment) (Kim et al., 2009). Finally, there is additional evidence that calcium lactate addition to early postmortem (24 h) beef loins results in an increase in myofibrillar protein degradation under HiOx-MAP (Kim, Huff-Lonergan, & Lonergan, 2012).

Beef round cuts are generally considered to be low quality (mainly due to inadequate tenderness) and are thus often underutilized (Anderson et al., 2012). Value added processing techniques such as enhancement combined with HiOx-MAP could enhance meat quality attributes of these underutilized beef round muscles. Taken together, it can be hypothesized that calcium lactate and phosphate enhancement to early postmortem beef round muscles will improve meat quality characteristics by minimizing oxidation related quality defects and accelerating the meat tenderization process under HiOx-MAP. Thus, the objective of the present study was to determine the influence of early postmortem calcium lactate/phosphate enhancement on meat quality and chemical/biochemical characteristics of beef round muscles under different packaging systems.

2. Materials and methods

2.1. Raw materials and processing

Ten market weight *Bos taurus* crossbred beef steers (547 ± 27 kg; 78 ± 2 weeks of age; A-maturity; USDA (1997) Low Choice grade) were harvested at the Iowa State University Meat Laboratory over 5 days. At 24 h postmortem, the *Mm. semimembranosus* (SM), *semitendinosus* (ST), and *adductor* (AD) were removed from each carcass. Within SM, the steaks were separated into the deep portion (DSM; the medial 1/3 closest to the femur) and superficial portion (SSM; the lateral 1/3 closest to the surface of the carcass) (Kim, Lonergan, & Huff-Lonergan, 2010), and were analyzed as a separate muscle for each trait. Each muscle was divided into 3 equal sections and randomly assigned one of three enhancement treatments: water injected control (CON), 0.3% sodium tripolyphosphate injection (Brifisol 512; BK Giulini Corp; Simi Valley, CA) (STP), and 200 mM calcium lactate (PURACAL; PURAC America, Inc., Lincolnshire, IL) followed by injection with 0.3% sodium tripolyphosphate (CAL/STP) by using a multi-needle injector (model N30, Wolftec, Inc., Werther, Germany). Each brine solution was injected to 12% of raw weight. Because of its smaller size, the AD was only cut into two sections assigned for STP and CAL/STP in order to test the effect of calcium lactate in enhanced AD. Sequential injections of calcium lactate followed by phosphate were applied with a 20 minute rest between injections to prevent a chelation of phosphates with calcium in solution if mixed together (Kim et al., 2009). The muscle weights before and after injection (30 min) were recorded to calculate actual brine pick-up (average 10% enhancement rate). The injected muscle sections were sliced into steaks (2.54 cm) and placed in preformed trays with soaker pads (polypropylene, 0.1 cm^3 of oxygen/tray/24 h at 22.7 °C/0% relative humidity, 2.0 g of water vapor/64,516 cm^2/24 h at 37.8 °C/100% relative humidity; Sealed Air Corp., Duncan, SC). Each tray was put in a shrinkable bag (B620, 20.3 by 30.48 cm/an oxygen-transmission rate of 3 cm^3 O_2/1 cm^3/m^2/24 h at 4.4 °C and 0% relative humidity and a water-vapor transmission rate of 0.5–0.6 g/254 cm^2/24 h at 37.8 °C and 100% relative humidity; Cryovac Sealed Air Corporation, Bolingbrook, IL), vacuum packaged (Multivac C500, Koch Supplies Inc., Kansas City, MO) and stored in a dark room at 1 °C.

Steaks assigned for MAP packaging were opened at day 9 and repackaged in HiOx-MAP (80% O_2/20% CO_2, Certified Standard within ± 2%, Praxair, Inc. Specialty Gases; Cahokia, IL) using a Multivac packaging machine (C500, Koch Supplies Inc., Kansas City, MO) by applying vacuum, and then flushing the package (shrinkable bag — B620) with the gas mixture followed by sealing. The gas headspace-to-steak ratio was approximately 2:1. VAC and HiOx-MAP packages were further displayed for 7 days (until day 16) at 1 °C under continuous fluorescent natural white light (Sylvania F40N, 3600 K, CRI = 86; Osram Sylvania, Danvers, MA) with 2150 ± 50 lux intensity. The gas composition of each package in HiOx-MAP was determined by using a headspace oxygen/carbon dioxide analyzer (PBI Dansensor, Glen Rock, NJ) at the end of display (day 16). Each carcass/muscle/enhancement treatment/packaging treatment/aging time combination represented one analytical sample.

2.2. pH

The pH of steaks at days 1, 9, and 16 of storage and display was measured with a calibrated pH glass penetration probe (Hanna 9025 pH/ORP meter; Hanna Instruments, Woonsocket, RI) at three different locations per steak.

2.3. Color stability

Surface lightness and redness (CIE L^* and a^* values, respectively) for days 1, 9, and 16 of storage and display of each steak was measured using a HunterLab LabScan® XE Spectrophotometer (Illuminant A, 2.54 cm diameter aperture, 10° standard observer; Hunter Associates Laboratory, Inc., Reston, VA). Three color readings for each steak were scanned and averaged for statistical analyses.

2.4. Star probe and sensory analyses

The steaks for star probe and sensory analyses were cooked on clamshell grills to an internal temperature of 71 °C. The temperature of individual steaks was monitored by using thermocouples inserted into the center of each steak (Omega Engineering, Inc., Stamford, CT). Star probe analysis of steak samples at days 1, 9, and 16 was conducted by following the procedure of Lonergan et al. (2007). A trained sensory panel (n = 6) evaluated sensory characteristics of steaks for day 16 samples only. Sensory traits including tenderness, chewiness, juiciness, beef flavor, and off-flavor (rancidity) were evaluated using a 15-cm line scale (1 = not tender, chewy, juicy, low beef flavor; 0 = no off-flavor; 15 = very tender, chewy, juicy, high beef flavor, high off-flavor) (Lonergan et al., 2007), and data were entered into a computerized sensory software system (Compusense five 4.6, Compusense, Inc., Guelph, Ontario, Canada).

2.5. Lipid oxidation

Lipid oxidation of steak samples from day 16 was determined using the 2-thiobarbituric acid reactive substance (TBARS) distillation method described by Tarladgis, Watts, Younathan, and Dugan (1960). The TBARS assay was conducted for each sample in duplicate.

2.6. SDS-PAGE and Western blot

Whole muscle protein samples were prepared from each sample and stored in gel sample buffer at −80 °C (Melody et al., 2004). For titin degradation, ST samples from days 1, 9, and 16 were analyzed using a 5% polyacrylamide continuous gel (acrylamide:N,N′-bis-methylene acrylamide = 100:1 [wt/wt],0.1% [wt/vol] sodium dodecyl sulfate, 0.05% [vol/vol] TEMED, 0.05% [wt/vol] ammonium persulfate, and 0.5 M Tris–HCl, pH 8.8). Gels (14 cm wide by 15 cm tall) were loaded with 160 μg protein and run for 48 h in Hoefer SE 400 electrophoresis units at 5 mA before staining with colloidal Coomassie blue dye (34% methanol, 17% ammonium sulfate (wt./vol.), 3% phosphoric

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