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Effects of postmortem calcium chloride injection on meat palatability traits of strip loin steaks from cattle supplemented with or without zilpaterol hydrochloride

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ABSTRACT: An experiment was conducted to determine the effects of zilpaterol hydrochloride m*M* supplementation (ZH; 8.3 mg/kg on a DM basis for 20 d) and calcium chloride injection [$CaCl_2$, 200 at 5% (wt/wt) at 72 h postmortem] on palatability traits of beef (*Bos taurus*) strip loin steaks. Select (USDA) strip loins were obtained from control (no ZH = 19) and ZH-supplemented carcasses (n = 20). Right and left sides were selected alternatively to serve as a control (no INJ) or $CaCl_2$–injected (INJ) and stored at 4°C. Before injecting the subprimals (72 h postmortem), 2 steaks were cut for proximate, sarcomere length, and myofibrillar fragmentation index (MFI) analyses. At 7 d postmortem each strip loin was portioned into steaks, vacuum packaged, and aged for the appropriate period for Warner-Bratzler shear force (WBSF; 7, 14, 21, and 28 d postmortem), trained sensory analysis (14 and 21 d postmortem), purge loss (7 d), and MFI (3, 7, 14, 21, and 28 d postmortem). Results indicated steaks from both ZH supplementation and INJ had reduced WBSF values as days of postmortem aging increased. The WBSF values of ZH steaks were greater ($P < 0.05$) than no ZH steaks at each postmortem aging period. The INJ steaks had lower WBSF values ($P < 0.05$) than non-injected steaks. A greater percentage (91 vs. 71%) of steaks had WBSF values < 4.6 kg from steers with no ZH supplementation at 7 d postmortem, but the percentage did not differ ($P > 0.05$) due to ZH at 14, 21, or 28 d or due to INJ at any aging period. Trained panelists rated tenderness less in ZH steaks than steaks with no ZH at 14 d and 21 d. However, INJ improved ($P < 0.05$) the tenderness ratings and flavor intensity of the trained panelists, compared with their non-injected cohorts at 21 d. Zilpaterol hydrochloride supplementation reduced ($P < 0.05$) MFI values, but INJ resulted in greater ($P < 0.05$) MFI values compared with no INJ. Subprimals from ZH and INJ showed greater purge loss ($P < 0.05$). Although no interactions were found with ZH and $CaCl_2$, injecting USDA Select strip loins from ZH-fed cattle can help reduce the normal WBSF variation as it does in steaks from non-ZH-fed cattle.

Key words: beef, calcium chloride, myofibrillar fragmentation index, shear force, tenderness, zilpaterol hydrochloride

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INTRODUCTION

Extensive research has been conducted to characterize the effects of zilpaterol hydrochloride [**ZH**; oral synthetic β-adrenergic receptor agonist (**β-AA**)] on feedlot cattle performance, carcass characteristics and retail yield, obtaining positive results on those traits (Vasconcelos et al., 2008; Beckett et al., 2009; Elam et al., 2009; Montgomery et al., 2009; Garmyn et al., 2010; Hilton et al., 2010). However, a slight reduction in tenderness score and increased shear force values resulting from ZH supplementation have also been documented (Brooks et al., 2009; Hilton et al., 2009; Leheska et al., 2009; Garmyn et al., 2010, 2011). Consequently, postmortem technologies have been applied to carcasses or steaks from animals fed with ZH (i.e., aging, enhancement, blade tenderization, electric stimulation) to improve the

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inherent problems of increased shear force on strip steaks, and the results, especially in short-aged product, have not always demonstrated similar WBSF values (Brooks et al., 2009, 2010; Leheska et al., 2009; Hope-Jones et al., 2010).

Despite the application of postmortem technologies, it is important to continue to explore other options, such calcium chloride (**$CaCl_2$**) injection, to test its ability and efficacy in reducing the variation of Warner-Bratzler shear force (**WBSF**) of steaks from cattle supplemented with ZH, as well as cattle that have not been exposed to β-AA. Limited data exist examining $CaCl_2$ infusion or injection in conjunction with the supplementation of β-AA. However, Koohmaraie and Shackelford (1991) reported $CaCl_2$ infusion was effective in overcoming the toughness of meat associated with dietary administration of $L_{644,969}$ in lambs.

Based on the aforementioned, the objective of this study was to determine the effects of ZH supplementation and $CaCl_2$ injection at 200 m*M* at 5% (wt/wt) on palatability attributes and WBSF of beef strip loin steaks.

MATERIALS AND METHODS

All procedures involving the use of animals followed the guidelines described in the Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching (FASS, 1999).

Cattle

The experiment was conducted at a research facility in Wellington, CO. British crossbred steers (n = 547) were processed within 7 d of arrival at the research facility. One day before the start of the trial (d –1), initial processing included i) measurement of individual BW, ii) vaccination with a modified-live 5-way viral vaccine, iii) treatment for parasites with Safe-Guard oral suspension (Merck Animal Health; DeSoto, KS) and Ivomec injectable (Merial; Duluth, GA), and iv) implantation with Revalor XS (200 mg TBA and 40 mg E_2; Merck Animal Health). Steers were fed a 66% concentrate receiving diet. Steers with extremes in BW, clinical signs of disease, lameness, behavioral problems, or poor body condition were removed from the experiment. Remaining steers (n = 480; initial BW = 310.8 ± 4.5 kg) were stratified by BW into 10 weight strata. On d 0, 1 steer from each strata was assigned randomly to each of 48 pens. Steers were weighed as a pen on d 1 and assigned a pen tag. Four treatments arranged in a 2 × 2 factorial were assigned to pens in a completely randomized design (48 pens total; 12 pens/treatment; 10 steers/pen). Factors included supplementation with ZH for 0 or 20 d (8.3 mg/kg, on a DM basis) followed by a 3-d withdrawal period before shipping with or without supplemental vitamin D_3. Vitamin D_3 supplementation began 10 d before harvest to provide 500,000 IU vitamin D_3/(steer · d). It should be noted that steers receiving vitamin D_3 were not eligible for carcass or subprimal selection.

Management and Feeding Procedures

Steers were adapted from the receiving diet to the final diet using 5 step-up diets (66, 76, 85, 86, and 89% concentrate diets). Steers were permitted ad libitum consumption according to the standard operating procedures of the research facility. The final 89% concentrate diet is shown in Table 1. Diets were formulated to meet or exceed NRC (1996) recommendations for nutrients. Once steers reached the final diet, feed delivery occurred once daily with a commercial feed delivery truck according to the standard operating procedures of the facility. Feed truck diet carryover was addressed by loading the feed truck and feeding in the following order: i) flush (corn-silage), ii) negative control diet, iii) vitamin D_3, iv) vitamin D_3 with ZH, and v) ZH. Feeding order of diets was documented daily to ensure that carryover was not a potential confounding factor. Zilpaterol hydrochloride was supplied as a Type A medicated article affixed to a ground corncob carrier and was dispensed through a micro-ingredient machine. Based on mid-study pen weights, pens were divided into light and heavy groups (24 pens total/group; 6 pens/treatment).

Carcass Evaluation and Selection

At 143 d, the heavy group (n = 234) was shipped approximately 595 km to a commercial plant for carcass data and subprimal collection, and at 163 d, the light group (n = 232) was shipped approximately 503 km to another commercial plant. During the study, 3 steers

Table 1. Composition and analyzed nutrient content (DM basis) of the finishing diets

Item	Diet, %
Ingredient	
Flaked corn	61.23
Wet distillers grain	25.03
Corn silage	5.11
Corn stalks	3.45
Micro premix[1]	5.16
Supplement	0.03
Analyzed Composition	
DM	54.9
CP	16.06
Ether extract	5.33
Calcium	0.68
Phosphorus	0.40
NDF	12.62

[1]Micro premix supplied (90% DM basis) 33 mg/kg of Rumensin (Elanco Animal Health, Indianapolis, IN) and 10 mg/kg of Tylan (Elanco Animal Health).

died and 11 steers were removed from the trial due to disease, appetency problems, or other conditions deemed unacceptable or inconsistent with the study objectives. Animals were identified at slaughter, and their identities were maintained throughout the grading and fabrication process. At 36 h postmortem, carcasses were graded. Carcass data were collected by trained personnel from Texas Tech University. Carcass measurements included HCW, 12th-rib fat thickness, LM area, KPH, and marbling score. Yield grade was calculated from HCW, LM area, 12th-rib fat thickness, and KPH (USDA, 1997). A subset of USDA Select carcasses (USDA, 1997) were chosen from the negative control and the group of steers supplemented with ZH only, and their carcass data are reported in Table 2 (non-ZH-fed = 19; ZH-fed = 20). As previously noted, carcasses from steers receiving supplemental vitamin D_3 were not selected. At 48 h postmortem, paired strip loins (Institutional Meat Purchase Specifications #180; NAMP, 2007) were obtained from their carcasses, placed in combos (non-vacuum packaged), and immediately shipped under refrigeration at 4°C to the Texas Tech Gordon W. Davis Meat Science Laboratory (Lubbock, TX).

Fabrication and Injection

The shipment arrived at the meat laboratory approximately 72 h postmortem for further processing. Strip loins from the right or left sides from both feeding groups were selected alternatively to serve as a control (**no INJ**) or injected with 200 m*M* food grade $CaCl_2$ (**INJ**) at 5% (wt/wt; Tetra Technologies, The Woodlands, TX). Previous work has shown injection at 24 or 48 h with 200 m*M* $CaCl_2$ at 5% could be applied without detrimental effects on palatability (Wheeler et al., 1993; Diles et al., 1994; Kerth et al., 1995, Lansdell et al., 1995). Before injecting the subprimals, 2 initial steaks (anterior and posterior portions) were cut for proximate (anterior portion), sarcomere length and myofibrillar fragmentation index (**MFI**) analyses (posterior portion), and stored at –20°C (for proximate) or –84°C (for sarcomere length and MFI) in Whirl-pak bags (Nasco, Fort Atkinson, WI). The control strip loins were not injected with water, because previous work has shown that injection of water has little, if any, effect on muscle tenderness (Wheeler et al., 1993) and because water injection is not used commercially (Diles et al., 1994). Distilled and deionized water (5°C) was used to formulate the $CaCl_2$ solution. Injection was performed using a multi-needle pickle injector (Wolf-Tec, Inc., Model Schroder/Imax 350, Kingston, NY). Strip loins were placed fat side down before introduction into the injector. Strip loins were allowed to equilibrate for 5 min after injection, and were then weighed to determine final percentage of injection (4.99% ± 1.07). After injection, each strip loin was relabeled by animal number and treatment, vacuum packaged (Barrier bag BH620T, Sealed Air Inc., Cryovac division, Duncan, SC), and stored at 4°C until 7 d postmortem (4 d after injection).

Table 2. Effects of supplementing crossbred steers with zilpaterol hydrochloride (ZH) on carcass traits of USDA Select carcasses (no ZH = 19; ZH = 20)[1]

	Feeding group			
Trait	No ZH	ZH	*P*-value	SEM
HCW, kg	375.77	402.72	0.0017	7.37
Marbling score[2]	356.32	361.50	0.5837	9.37
Fat thickness, mm	11.99	12.14	0.9022	1.18
LM area, cm^2	88.97	101.05	0.0001	2.54
KPH, %	3.31	3.40	0.7152	0.22
USDA yield grade	2.36	1.80	0.0259	0.24

[1]Zilpaterol hydrochloride (8.3 mg/kg on DM basis) was fed for 20 d with a 3-d withdrawal (Merck Animal Health, DeSoto, KS).

[2]Marbling scores: 300 = slight, 400 = small.

Procurement of Samples and Postmortem Aging

At 7 d postmortem (4 d after injection), strip loins were removed from their packages, weighed (for purge loss calculation), and eight 2.54-cm thick steaks were obtained from the anterior end using a manual meat slicer (Berkel, Model X13E, 13 inch, South Bend, IN) to ensure uniform thickness.

The steaks obtained at 7 d postmortem from each strip loin were assigned to these analyses and aging treatment: 1 steak was cut into 4 equally sized pieces and assigned to MFI at 7, 14, 21, and 28 d postmortem; 1 steak for proximate analysis at 7 d; 2 steaks for trained sensory analysis at 14 or 21 d postmortem; 4 steaks for WBSF and slice shear force (**SSF**) at 7, 14, 21, and 28 d postmortem. Steaks for proximate, sensory, WBSF, and SSF evaluation were vacuum packaged in a multilaminar, thermo-shrinkable (BHT 620 Sealed Air Inc., Cryovac division, Duncan, SC) using a Koch Ultravac vacuum packaging machine (Model UV-250, Kansas City, MO).

Steaks for proximate analysis were frozen immediately at –20°C and stored until analysis. Steaks assigned to MFI were frozen at –84°C after the appropriate aging period (7, 14, 21, or 28 d) had elapsed. All shear force and sensory steaks were stored at 4°C and frozen on the appropriate postmortem aging day (d 7, 14, 21, or 28). Bias due to anatomical position was avoided by alternating designated analyses (i.e., sensory tests, shear force test) and by rotating postmortem aging period treatment (7, 14, 21 or 28 d) among steak locations for each subsequent subprimal.

Purge Loss

At 7 d postmortem, each strip loin was removed from the vacuum package, blotted with a towel to remove surface moisture, and weighed to acquire the actual boneless strip loin weight. Percentage purge was calculated using this formula: [(weight of subprimal before package, g – weight of drained and blotted subprimal after removing from its package, g)/weight of subprimal before package, g] × 100.

Proximate Analysis

Steaks for proximate analysis were thawed, trimmed of external fat, and homogenized in a food processor (KitchenAid with grinder adaptor, Model KP26M1XER Professional 600; St. Joseph, MI) to obtain approximately 200 g of sample. Compositional analysis of moisture, fat, protein, and collagen was conducted using an AOAC-approved (Official Method 2007.04; Anderson, 2007) near-infrared spectrophotometer (FOSS Food ScanTM 78800; Laurel, MD).

Sarcomere Length

Sarcomere length was determined on fresh strip loin samples, using the neon laser diffraction method described by Cross et al. (1981). The neon laser (Spectra Physics Inc., Model 117A, CA, Irvine, CA) was operated at 632.8 nm wavelength.

Samples were cut into small pieces (3.0 × 3.0 × 2.0 cm^3) with the fibers running longitudinally and placed in scintillation vials. Glutaraldehyde solution (5%; Fisher Scientific; Fair Lawn, NJ) was added and fixed for 4 h at 4°C. The glutaraldehyde solution was poured off and replaced with a 0.2 M sucrose solution. Samples were then stored overnight at 4°C. A small bundle of muscle fiber was obtained with dissecting forceps and placed on microscope slides. Individual fibers were gently spread and covered with glass cover slips. A drop of sucrose solution was used to keep samples moist. The lengths of at least 5 diffraction patterns from each sample were measured, and sarcomere length (μm) was determined by averaging the measurements.

Myofibrillar Fragmentation Index

Myofibril fragmentation index was determined in frozen muscle at 3, 7, 14, 21, and 28 d postmortem, according to the procedure described by Culler et al. (1978). Analysis was performed in duplicate (acceptable when intra-sample variation ≤ 10%). Optical density was read using a spectrophotometer (DU-640, Beckman Instruments, Inc., Fullerton, CA) at a wavelength of 540 nm.

Cooking Procedures

Steaks were thawed at 4°C for 24 h before shear force or sensory evaluation. Sample preparation and cooking procedures were followed according to guidelines described by the American Meat Science Association (AMSA, 1995). Before cooking, steaks were trimmed to remove external fat. Thawed weight and initial temperature (Cooper Instruments digital meat thermometer Model SH66A, Middlefield, CT) were recorded. Steaks were cooked on a Magi-grill belt grill (Model TBG-60, Magigrill, Magi-Kitch'n Inc., Quakertown, PA) with a grill-plate temperature set at 163°C to achieve a final internal temperature of 71°C.

Once steaks exited the belt grill, final internal temperature (°C) and cooked weight (g) were recorded immediately. Cooking loss (%) was calculated using the following formula: [(thawed weight, g – cooked weight, g)/thawed weight, g] × 100.

Slice Shear Force Analysis

Immediately after cooking, a 1- to 2-cm slice was removed across the width of the steak from both the lateral and medial end to square off the steak and expose the muscle fibers. Using a cutting guide, a 5-cm long × 1-cm thick section was obtained from the lateral end by cutting across the section at a 45° angle parallel to the muscle fiber orientation. The sample was center sheared perpendicular to the muscle fiber using a United force analyzer (Model SSTM-500 with a tension attachment, United Calibration Corp. Huntington Beach, CA) with a cross head speed of 500 mm/min with a load cell of 50 kg. Shear force values were recorded in kilograms.

Warner-Bratzler Shear Force Analysis

After each steak was slice sheared, the portions of the remaining steak were placed on metal trays, covered with polyvinyl chloride film and chilled at approximately 2°C for 24 h. Warner-Bratzler shear values were obtained by removing six 1.3-cm-diameter cores parallel to the muscle fiber orientation from the remaining portions of the steak. Cores were then sheared perpendicular to the muscle fibers using a WBSF analyzer (G-R Elec. Mfg., Manhattan, KS). Shear force values were recorded in kg, and the values from the 6 cores of each steak were averaged for statistical analysis.

Trained Sensory Evaluation

Steaks for sensory analysis were cooked as described above. Cooked steaks were trimmed of visible fat and connective tissue. Each steak was cut into cubes (1 × 1 × 2.5 cm^3), which were placed in a preheated pan to keep

samples warm (at 50°C) until serving. Samples were served warm and accompanied with a glass of distilled, deionized water, apple juice, and unsalted crackers to allow panelists to cleanse their palates between samples.

The sensory panel was composed of 6 to 8 trained panelists recruited from the Animal and Food Sciences department at Texas Tech University. Sensory sessions were conducted twice a day, with a 1-h break between sessions. Ten samples were evaluated in each session. During sessions, panelists were seated randomly in individual booths in a temperature controlled room with red lighting following recommendations of AMSA (1995). Panelists scored samples using a 8-point verbally anchored numerical scale for initial and sustained juiciness (8 = extremely juicy, 1 = extremely dry), initial and overall tenderness (8 = extremely tender, 1 = extremely tough), flavor intensity (8 = extremely intense, 1 = extremely bland), beef flavor (8 = extremely characteristic beef flavor, 1 = extremely uncharacteristic beef flavor), and overall beef mouthfeel (8 = extremely beef-like mouthfeel, 1 = extremely non-beef-like mouthfeel). The off flavor was scored using a 5-point scale (1 = none, 5 = extremely off flavor).

Statistical Analysis

All data were analyzed using SAS (SAS Inst. Inc., Cary, NC). Harvest facility was included in the model as a random effect. Pen was not included in analyses because quality grade was used as selection criterion for inclusion in the trial, and carcasses were selected randomly to meet this criterion. When applicable, ZH supplementation, $CaCl_2$ injection, postmortem aging, and their interactions were treated as fixed effects. The degrees of freedom in the denominator were adjusted using the Satterthwaite procedure. Least squares means were separated (F-test, $P < 0.05$) by using least significant differences generated by the PDIFF option. Special considerations are explained in the next paragraphs.

Proximate Analysis. Two models were used because samples were processed separately for each postmortem day (3 or 7 d). At 3 d postmortem, ZH supplementation was the only fixed effect because samples were obtained before injection. Analysis of samples from d 7 included ZH supplementation, injection, and the 2-way interaction as fixed effects.

Myofibrillar Fragmentation Index. Because some confounding effects of injection treatment and postmortem aging were expected, data were divided into 2 sub-sets for conducting analyses. The first data sub-set corresponded only to the non-injected samples from both ZH supplementation groups at 3, 7, 14, 21, and 28 d postmortem. Fixed effects included ZH supplementation and postmortem aging. The second data sub-set represented injected and non-injected steaks from both ZH supplementation groups aged at 7, 14, 21, and 28 d postmortem. Fixed effects included ZH supplementation, $CaCl_2$ injection, and postmortem aging.

WBSF, SSF, and Cooking Loss. Internal cooked temperature affected SSF and cooking loss ($P < 0.05$), so it was included in those models as a covariate. For WBSF and cooking loss, no 2-way or 3-way interactions were observed ($P > 0.05$). Data were subsequently analyzed and reported by postmortem aging period. For SSF, an interaction between $CaCl_2$ injection and postmortem aging was observed ($P < 0.05$); however, no other interactions were observed for SSF ($P > 0.05$). Additionally, WBSF threshold levels were analyzed as binomial proportions using the GLIMMIX procedure with the ILINK option of the LSMEANS statement used to calculate the least squares means for the proportions. Injection, ZH supplementation, and their interaction were analyzed by postmortem aging period.

Trained Sensory Evaluation. Data across postmortem aging period (14 or 21 d) were analyzed separately because trained panelists were served within aging treatment. The internal cooked temperature did not affect treatments involved ($P > 0.05$) so it was not included in the model as a covariate.

RESULTS AND DISCUSSION

Carcass Data

Supplementation with ZH affected ($P < 0.05$) HCW, LM area, and USDA yield grade for the carcasses selected (Table 2). The ZH carcasses were heavier (>26.95 kg) and had larger LM area (>12.08 cm^2), translating to a lower numerical USDA yield grade than no-ZH carcasses. In the carcasses chosen for this trial, fat thickness, KPH, and marbling score were not affected ($P > 0.05$) by ZH supplementation.

Generally, previous results have shown ZH supplementation increased HCW and LM area (Avendano-Reyes et al., 2006; Vasconcelos et al., 2008; Beckett et al., 2009; Elam et al., 2009), whereas KPH, fat thickness (Beckett et al., 2009; Montgomery et al., 2009) and marbling were not affected (Casey et al., 1997; Plascencia et al., 1999). In contrast, other researchers (Vasconcelos et al., 2008; Elam et al., 2009; Hilton et al., 2009) have reported a decrease in KPH, as well as fat thickness with ZH supplementation.

There were no differences in marbling because all carcasses selected for this study were targeted within the USDA Select quality grade. However, in previous reports marbling score has decreased when ZH was fed to steers for more than 20 d (Beckett et al., 2009; Elam et al., 2009; Hilton et al., 2009; Montgomery et al., 2009). In alignment with previous studies, yield grade decreased with ZH supplementation compared with

Table 3. Effects of zilpaterol hydrochloride (ZH) on proximate composition, collagen content, and sarcomere length of strip loin steaks at 3 d postmortem (no ZH = 19; ZH = 20)[1]

Item	Feeding group		*P*-value	SEM
	No ZH	ZH		
Protein content, %	23.64	24.13	0.002	0.10
Fat content, %	2.98	2.88	0.710	0.27
Moisture content, %	71.80	71.39	0.092	0.16
Collagen content, %	1.61	1.60	0.685	0.02
Sarcomere length, μm	1.88	1.81	0.290	0.04

[1]Zilpaterol hydrochloride (8.3 mg/kg on DM basis) was fed for 20 d with a 3-d withdrawal (Merck Animal Health, DeSoto, KS).

controls (Beckett et al., 2009; Elam et al., 2009; Hilton et al., 2009; Montgomery et al., 2009).

Proximate Analysis, Collagen Content, and Sarcomere Length

As seen in Table 3, ZH increased ($P < 0.01$) the percentage of protein by 0.49% in samples taken at 3 d postmortem before injection, but it did not affect fat, moisture, and collagen contents ($P > 0.05$). Carcasses were selected at the same USDA Select quality grade, so ZH did not affect the percentage of fat ($P = 0.71$). Leheska et al. (2009) did not observe any effect of ZH treatment on fat, moisture, and ash; however, protein content increased with ZH supplementation. In contrast, Hilton et al. (2009) reported feeding ZH for 30 d decreased fat percentage, whereas protein and moisture percentage were not affected by ZH supplementation. On the other hand, Kellermeier et al. (2009) and Rathmann et al. (2009) found cattle fed ZH had a decreased percentage of fat and a greater percentage of protein and moisture.

In the current study, there was no difference in collagen due to ZH supplementation ($P = 0.69$). In contrast, supplementation of other β-AA have resulted in a reduction of total collagen (Fiems et al., 1990; Vestergaard et al., 1994; Kellermeier et al., 2009; Strydom et al., 2009) due to the increased muscle hypertrophy, thus creating a dilution effect that decreased the concentration of collagen in muscle. In the current study, ZH did not have this effect on collagen.

Table 3 shows sarcomere length was not affected at 3 d postmortem by ZH supplementation ($P = 0.29$). Results from Hope-Jones et al. (2010) were in agreement with the current study in that ZH had no effect on sarcomere length. Cold shortening may be induced in animals treated with β-AA because they generally produce leaner carcasses. This could result in tougher meat; however, the carcasses from the current study had similar back fat thickness. According to Jeremiah (1996), carcasses should have between 2 and 13 mm of external fat to avoid cold shortening. Carcasses in the current study were within this optimum range, with at least 11 mm.

Table 4. Effects of zilpaterol hydrochloride (ZH) and $CaCl_2$ injection on proximate composition in strip loin steaks at 7 d postmortem

Item	Protein content, %	Fat content, %	Moisture content, %
ZH supplementation[1]			
No ZH	23.65	3.91	71.29
ZH	24.63	3.67	70.94
SEM[2]	0.11	0.26	0.18
P-value[3]	<0.01	0.52	0.17
Injection[4]			
No $CaCl_2$	24.19	3.81	70.86
$CaCl_2$	28.09	3.78	71.37
SEM[2]	0.10	0.20	0.16
P-value[3]	0.43	0.87	0.01

[1]Zilpaterol hydrochloride (8.3 mg/kg on DM basis) was fed for 20 d with a 3-d withdrawal (Merck Animal Health, DeSoto, KS).

[2]Pooled (largest) SE of least squares means. Sample numbers: No ZH = 38; ZH = 40; No $CaCl_2$ = 39; $CaCl_2$ = 39.

[3]Observed significance levels for main effects of ZH supplementation and $CaCl_2$ injection. No significant 2-way interactions were observed ($P > 0.05$).

[4]Calcium chloride food grade at 200 m*M* at 5% (wt/wt); Tetra Technologies, The Woodlands, TX.

At 7 d postmortem, ZH samples had a greater ($P < 0.01$) percentage protein compared with no ZH (Table 4). In addition, $CaCl_2$ steaks had greater ($P < 0.01$) moisture compared with no $CaCl_2$ steaks. No other differences in proximate composition were detected ($P > 0.05$) due to ZH supplementation or $CaCl_2$ injection at 7 d postmortem. In addition, no significant 2-way

Table 5. The effects of zilpaterol hydrochloride (ZH)[1] and postmortem aging period on myofibrillar fragmentation index (MFI)

Item	MFI
ZH supplementation	
No ZH	68.58
ZH	58.36
SEM[2]	3.35
P-value[3]	<0.01
Postmortem age, d	
3	40.17[a]
7	67.75[b]
14	68.73[b]
21	69.40[b]
28	71.32[b]
SEM[2]	3.66
P-value[3]	<0.01

[a,b]Least squares means within a column lacking a common superscript letter differ ($P < 0.05$).

[1]Zilpaterol hydrochloride (8.3 mg/kg on DM basis) was fed for 20 d with a 3-d withdrawal (Merck Animal Health, DeSoto, KS).

[2]Pooled (largest) SE of least squares means. Sample numbers/day: No ZH = 19; ZH = 20.

[3]Observed significance levels for main effects of ZH and postmortem aging. No significant interaction was observed ($P = 0.86$).

Table 6. Effects of zilpaterol hydrochloride (ZH)[1] and $CaCl_2$ injection (INJ)[2] on myofibrillar fragmentation index (MFI) at 7, 14, 21, and 28 d postmortem (no ZH = 19; ZH = 20)

	No ZH		ZH			*P*-values[4]		
Day	No INJ	INJ	No INJ	INJ	SEM[3]	ZH	INJ	Z × I
7	74.53	86.61	61.22	72.48	3.25	<0.01	<0.01	0.90
14	75.39	88.26	62.88	75.03	3.38	<0.01	<0.01	0.91
21	74.56	93.78	64.43	79.09	3.37	<0.01	<0.01	0.44
28	74.39	96.94	68.35	85.28	4.46	0.02	<0.01	0.47

[1]Zilpaterol hydrochloride (8.3 mg/kg on DM basis) was fed for 20 d with a 3-d withdrawal (Merck Animal Health, DeSoto, KS).

[2]Calcium chloride food grade at 200 m*M* at 5% (wt/wt); Tetra Technologies, The Woodlands, TX.

[3]Pooled (largest) SE of least squares means. Sample numbers/day: No ZH = 38; ZH = 40; No INJ = 39; INJ = 39.

[4]Observed significance levels for main effects of zilpaterol hydrZHochloride, $CaCl^2$ injection, and the ZH × $CaCl^2$ injection interaction by aging period. No 2-way or 3-way interactions were observed ($P > 0.05$).

interactions were observed ($P > 0.05$). Due to the added water, $CaCl_2$–injected samples had greater moisture content (0.51%) compared with non-injected samples, without any effects on the other chemical components.

Myofibrillar Fragmentation Index

Table 5 shows the effects of ZH supplementation and postmortem aging on MFI of non-injected samples only. Both ZH supplementation ($P < 0.01$) and postmortem aging ($P < 0.01$) affected MFI; however, no interaction was observed ($P = 0.76$). Steaks from non-ZH supplemented steers showed greater MFI compared with ZH steers. The effect of postmortem aging on MFI had the most dramatic change in the first 4 d postmortem, when there was a significant increase from d 3 to d 7; however, little change occurred from d 7 to d 28 in MFI values.

The effects of ZH and $CaCl_2$ injection on MFI at 7, 14, 21, and 28 d postmortem can be seen in Table 6. No 2-way or 3-way interactions were observed ($P > 0.05$) for ZH, INJ, or postmortem aging. At each aging period, ZH supplementation resulted in steaks with lower MFI values compared with non-supplemented controls. In addition, INJ had greater MFI values at every postmortem aging period compared with non-injected steaks. Moreover, extended postmortem aging resulted in greater MFI values ($P < 0.01$; data not shown in tabular form). Regardless of ZH or INJ, steaks aged 7 d had the lowest MFI values (73.60), but the average MFI value reached 81.24 by 28 d postmortem.

Fiems et al. (1990) reported that feeding diets with cimaterol (60 μg or 4 mg/kg kg/d in the diet, until d 246) reduced myofibrillar protein degradation by reducing the activity of the proteolytic enzymes in young bulls. Likewise, Kretchmar et al. (1990) reported that MFI values from control lambs steadily increased over the 6-d postmortem aging period (50%); in contrast, the index values for the β-AA treatment group increased only slightly over 6 d (8.7%), and MFI values obtained on d 2, 4, and 6 were not significantly different from those obtained on d 1. Similar results have been obtained in lambs and steers fed with $L_{644,969}$ (Koohmaraie et al., 1991; Wheeler and Koohmaraie, 1992) so this study did not appear to follow those trends as MFI increased from 3 to 7 d from steaks that came from cattle fed ZH.

Table 7. Effects of $CaCl_2$ injection (INJ)[1] and postmortem aging (7, 14, 21, or 28 d) on cooking loss (%)

Item	Cooking loss, %
Injection	
No $CaCl_2$	18.60[b]
$CaCl_2$	19.46[a]
SEM[2]	0.20
P-value	<0.01
Postmortem age, d	
7	17.62[c]
14	18.50[b]
21	19.16[b]
28	20.83[a]
SEM[2]	0.29
P-value	<0.01

[a,b]Least squares means within a column and main effect lacking a common superscript letter differ ($P < 0.05$).

[1]Calcium chloride food grade at 200 m*M* at 5% (wt/wt); Tetra Technologies, The Woodlands, TX.

[2]Pooled (largest) SE of least squares means. Sample numbers/day: No ZH = 19; ZH = 20.

[3]Observed significancelevels for injection and postmortem aging. No significant interaction was observed ($P > 0.05$).

The processes of enzymatic tenderization in meat from β-AA treated animals are reduced, due mainly to the action of enzymatic inhibitor (i.e., calpastatin), which decreases muscle protein degradation (Wang and Beermann, 1988; Parr et al., 1992). However, Hilton et al. (2009) showed ZH supplementation had no effect on calpastatin, μ- or m-calpain activities. In meat with excess variation in tenderness (Koohmaraie and Shackelford, 1991; Morgan et al., 1991; Wheeler et al., 1991; Diles et al., 1994; Lansdell et al., 1995), $CaCl_2$ has been used to accelerate the tenderization process through activation of m-calpain activity rather than μ-calpain (Koohmaraie et al., 1989).

Moisture Loss

Zilpaterol hydrchloride supplementation and $CaCl_2$ affected ($P < 0.01$) purge loss at 7 d postmortem (data not shown in tabular form). Steers supplemented with ZH (1.71%) had strip loins with 0.4% greater purge loss compared with no ZH (1.32%). Strip loins injected with $CaCl_2$ (2.39%) had 1.75% greater purge loss compared with non-injected strip loins (0.64%). No interaction was observed ($P = 0.08$) between ZH supplementation

Table 8. Effects of zilpaterol hydrochloride (ZH)[1] and $CaCl_2$ injection (INJ)[2] on Warner-Bratzler shear force (WBSF) at 7,14, 21, and 28 d postmortem (no ZH = 19; ZH = 20)

Days post-mortem	No ZH		ZH			*P*-values[4]		
	No INJ	INJ	No INJ	INJ	SEM[3]	ZH	INJ	Z × I
7	3.65	3.19	4.45	4.12	0.20	<0.01	0.06	0.75
14	3.53	3.06	4.08	3.76	0.14	<0.01	<0.01	0.61
21	3.29	3.00	3.71	3.59	0.15	<0.01	0.17	0.59
28	2.95	2.78	3.50	3.28	0.11	<0.01	0.07	0.83

[1]Zilpaterol hydrochloride (8.3 mg/kg on DM basis) was fed for 20 d with a 3-d withdrawal (Merck Animal Health, DeSoto, KS).

[2]Calcium chloride food grade at 200 m*M* at 5% (wt/wt); Tetra Technologies, The Woodlands, TX.

[3]Pooled (largest) SE of least squares means. Sample numbers/day: No ZH = 38; ZH = 40; No INJ = 39; INJ = 39.

[4]Observed significance levels for main effects of ZH, $CaCl^2$ injection, and ZH × $CaCl^2$ injection interaction by aging period.

and injection. As seen in Table 7, cooking loss was affected by $CaCl_2$ injection ($P < 0.01$) and postmortem aging ($P < 0.01$). Steaks aged 28 d postmortem had the greatest cooking loss. Steaks aged 14 and 21 d were intermediate and did not differ from each other. Steaks aged 7 d had the least cooking loss. Steaks from strip loins injected with $CaCl_2$ had greater ($P < 0.01$) cooking loss when compared with non-injected strip loins; however, ZH supplementation had no effect on cooking loss ($P > 0.05$). Cooking loss was not affected ($P > 0.05$) by any interactions between ZH supplementation, $CaCl_2$ injection, and postmortem aging period.

Greater purge loss was reported by Kellermeier et al. (2009) when evaluating the use of terminal implants (estrogen + trenbolone acetate) and feeding ZH (8.38 mg/kg DM basis) for 30 d. Rathmann et al. (2009) reported similar results for the evaluation of feeding ZH (8.33 mg/kg, DM basis) for 0, 20, 30, or 40 d before slaughter and days on the finishing diet (DOF; 136, 157, 177, and 198 d). Avendano-Reyes et al. (2006) evaluated the effects of ZH and RH supplied for 33 d and observed that drip loss by press for ZH and RH steers increased with time. Similarly, Strydom (2009) found greater drip loss (> 1.79%) in ZH-fed steaks than control steaks; however, RH and clenbuterol groups had intermediate drip loss. In contrast, other studies have indicated no or little variation in drip loss using cimaterol (Fiems et al., 1990; Boucqué et al., 1994).

Most studies have indicated $CaCl_2$ injection reduces water holding capacity (greater drip loss). Wheeler et al. (1993) determined how concentration (175, 200, and 250 m*M*) and injection amount (5 and 10%) of $CaCl_2$ affected drip loss of LM. Injection amount did not affect drip loss, but increasing the concentration resulted in greater drip loss. Boleman et al. (1995) indicated drip loss was greater in samples injected at different times (300 m*M* of $CaCl_2$ at 1, 12, and 24 h postmortem); Jaturasitha et al. (2004) reported the injection of $CaCl_2$ [200, 300, and 400 m*M* at 10% (wt/wt)] increased drip loss by over twofold.

In alignment with the current results, studies have indicated no variation in cooking loss of steaks from ZH-treated animals (Hilton et al., 2009; Leheska et al., 2009; Garmyn et al., 2010). However, Garmyn et al. (2011) saw increased cooking loss due to ZH supplementation when steaks were aged up to 28 d postmortem. Researchers have observed that $CaCl_2$ injection increased cooking loss (≥5%) when injected at various concentrations (200, 250, 300 m*M* $CaCl_2$) at post-rigor or freezing (Wheeler et al., 1992, 1993, 1997; Boleman et al., 1995). However, other researchers have reported no change or reduction in cooking loss (Morgan et al., 1991; Diles et al., 1994; Kerth et al., 1995; Lansdell et al., 1995; Jaturasitha et al., 2004), which may be attributed to greater purge loss during postmortem storage of injected subprimals or steaks.

Shear Force

The effects of ZH and $CaCl_2$ injection on WBSF are shown in Table 8. There were no interactions between ZH, $CaCl_2$ injection, and postmortem aging for WBSF ($P > 0.05$). Postmortem aging affected ($P < 0.01$) WBSF values. Significantly less shear force was required for each additional 7 d of postmortem aging. Shear force values for steaks aged 7, 14, 21, and 28 d were 3.86, 3.61, 3.40, and 3.13 kg, respectively (data not shown in tabular form). Data were subsequently analyzed by postmortem aging, least squares means are reported in Table 8. Supplementation with ZH resulted in steaks with greater ($P < 0.01$) WBSF values at 7, 14, 21, and 28 d postmortem when compared with no ZH

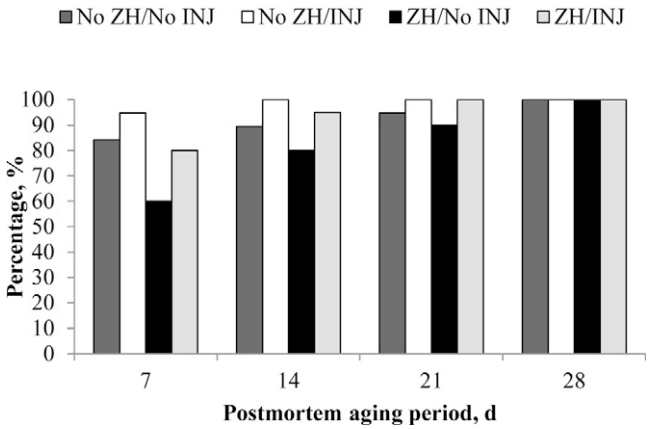

Figure 1. Effects of feeding 8.3 mg/kg (DM basis) zilpaterol hydrocloride (ZH; Merck Animal Health, DeSoto, KS) for 0 or 20 d and postmortem $CaCl_2$ injection [INJ; 200 m*M* at 5% (wt/wt) at 72 h postmortem] on the percentage of USDA Select strip loin steaks with Warner-Bratzler shear force values < 4.6 kg at 7, 14, 21, and 28 d postmortem. (no ZH = 19, ZH = 20; d 7: SEM = 10.1; *P*-values: ZH = 0.05, INJ = 0.12, ZH × INJ = 0.87; d 14: SEM = 8.9; *P*-values: ZH = 0.97, INJ = 0.97, ZH × INJ = 0.97; d 21: SEM = 6.7; *P*-values: ZH = 0.99, INJ = 0.97, ZH × INJ = 0.99); d 28: SEM = 0.0; *P*-values: ZH = 1.00, INJ = 1.00, ZH × INJ = 1.00).

supplementation. Injection reduced ($P < 0.01$) shear values on d 14 postmortem compared with steaks from non-injected strip loins. There were no interactions between ZH and $CaCl_2$ for WBSF at any postmortem aging period ($P > 0.05$). A greater percentage (91 vs. 71%) of steaks had WBSF values < 4.6 kg from steers with no ZH supplementation at 7 d postmortem, but the percentage did not differ ($P > 0.05$) due to ZH at 14, 21, or 28 d or due to INJ at any aging period (Figure 1). As seen in Figure 1, greater than 90% of all steaks would be classified as tender based on WBSF values <4.6 kg by d 21 postmortem and 100% of steaks required less 4.6 kg of force to shear at 28 d postmortem.

Slice shear force values were greater ($P < 0.01$) due to ZH supplementation (14.1 kg) compared with no ZH (12.3 kg), regardless of postmortem aging period (data not shown in tabular form). Injection of $CaCl_2$ interacted ($P = 0.05$) with postmortem aging for SSF values (Figure 2). Non-injected steaks aged 7 d had greater SSF values than INJ steaks aged 7 d and all steaks aged 14, 21, or 28 d postmortem. Injected steaks aged 7 d and non-injected steaks aged 14 d had intermediate SSF values. Results from Mehaffey et al. (2009) are in agreement with the current study, where ZH caused increased SSF values with respect to the control treatment.

The effect of ZH supplementation on WBSF values in this study is in alignment with previous researchers who indicated postmortem aging decreased WBSF values of ZH-supplemented steaks and never reached the same WBSF value of non-ZH-supplemented steaks (Hilton et al., 2009; Holmer et al., 2009; Kellermeier et al., 2009; Leheska et al., 2009; Mehaffey et al., 2009; Rathmann et al., 2009; Strydom et al., 2009; Hope-Jones et al., 2010). Although the WBSF values of ZH supplemented steers do not reach values comparable with no ZH steaks, Garmyn et al. (2011) reported no differences in the percentage of steaks with WBSF < 4.6 kg at any postmortem aging period (7, 14, 21, 28, or 35 d) due ZH supplementation or implanting. By d 28 postmortem, over 99% of ZH steaks required less than 4.6 kg of force to shear (Garmyn et al., 2011). Brooks et al. (2009) showed supplementing with ZH followed by 14 d postmortem aging resulted in approximately 80% of steaks with WBSF values <4.5 kg. In alignment with previous results, greater than 90% of all steaks would be classified as tender in the current study by d 21 postmortem and 100% of steaks required less than 4.6 kg of force to shear at 28 d postmortem. Moreover, Brooks et al. (2009) did not find an interaction between feeding duration and postmortem aging for Choice or Select in all evaluated muscles, indicating the tenderness of control and ZH supplemented steaks improved with aging at a similar magnitude over time; thus postmortem aging from 7 to 21 d decreased WBSF values.

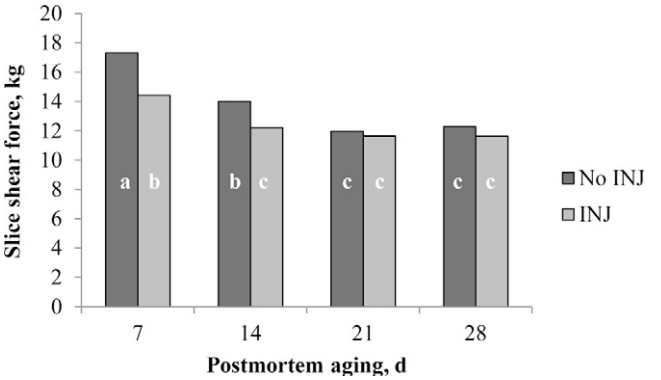

Figure 2. Effects of postmortem $CaCl_2$ injection [INJ; 200 m*M* at 5% (wt/wt) at 72 h postmortem] and postmortem aging period (7, 14, 21, and 28 d) on slice shear force values. Sample numbers/day: No ZH (zilpaterol hydrochloride; Merck Animal Health, De Soto, KS) = 38; ZH = 40; No INJ = 39; INJ = 39. SEM = 1.28; *P*-value: Aging × INJ = 0.05. Least squares means lacking a common letter differ ($P < 0.05$).

Sensory Evaluation

Table 9 shows the effects of ZH and $CaCl_2$ injection on trained sensory traits at 14 d postmortem. Zilpaterol hydrochloride supplementation resulted in lower ($P < 0.01$) scores for initial and sustained tenderness; however, panelists did not detect differences ($P > 0.05$) in any other palatability traits due to ZH supplementation in steaks aged 14 d. Injection with $CaCl_2$ resulted in greater scores for flavor intensity ($P = 0.01$) and beef flavor ($P = 0.04$) compared with steaks from non-injected strip loins aged 14 d. In addition, mean scores for off flavor were greater ($P = 0.04$) for steaks from strip loins injected with $CaCl_2$. Panelists did not detect differences ($P > 0.05$) in any other palatability traits due to $CaCl_2$ injection in steaks aged 14 d.

Table 10 shows the effects of ZH and $CaCl_2$ injection on trained sensory traits at 21 d postmortem. Panelists did not detect differences ($P > 0.05$) in any palatability traits due to ZH supplementation in steaks aged 21 d. Injection with $CaCl_2$ resulted in greater ($P < 0.01$) scores for flavor intensity and off flavor compared with steaks from non-injected strip loins aged 21 d. Panelists did not detect differences ($P > 0.05$) in any other palatability traits due to $CalCl_2$ injection in steaks aged 21 d.

Similar to results to the current study, Leheska et al. (2009) indicated cattle supplemented with ZH had lower scores in overall tenderness from trained panelists than control at 28 d of aging; however, Leheska et al. (2009) also reported ZH supplementation reduced overall juiciness and flavor. In the same way, Hilton et al. (2009) found that ZH-fed steaks aged for 14 d received lower scores from trained panelists in juiciness, tenderness, flavor intensity, and beefy flavor; consumers rated ZH-fed steaks with lower tenderness scores as well. In the current study, the juiciness scores were not affected by

Table 9. Effects of zilpaterol hydrochloride (ZH)[1] and $CaCl_2$ injection (INJ)[2] on trained sensory traits at 14 d postmortem (no ZH = 19; ZH = 20)

	No ZH		ZH			*P*-values[4]		
Trait	No INJ	INJ	No INJ	INJ	SEM[3]	ZH	INJ	Z × I
Initial juiciness[5]	5.98	6.16	6.03	6.03	0.11	0.76	0.41	0.44
Sustained juiciness[5]	5.95	6.12	5.96	6.04	0.12	0.79	0.31	0.72
Initial tenderness[5]	6.19	6.38	5.71	5.96	0.15	<0.01	0.12	0.85
Overall tenderness[5]	6.26	6.46	5.77	6.05	0.15	<0.01	0.11	0.78
Flavor intensity[5]	6.44	6.55	6.36	6.58	0.07	0.62	0.01	0.34
Beef flavor[5]	6.45	6.52	6.30	6.50	0.07	0.22	0.04	0.32
Overall mouthfeel[5]	5.52	5.70	5.37	5.43	0.13	0.09	0.34	0.66
Off flavor[6]	1.07	1.10	1.03	1.12	0.03	0.65	0.04	0.29

[1]Zilpaterol hydrochloride (8.3 mg/kg on DM basis) was fed for 20 d with a 3-d withdrawal (Merck Animal Health, DeSoto, KS).

[2]Calcium chloride food grade at 200 m*M* at 5% (wt/wt); Tetra Technologies, The Woodlands, TX.

[3]Pooled (largest) SE of least squares means. Sample numbers: No ZH = 38; ZH = 40; No INJ = 39; INJ = 39.

[4]Observed significance levels for main effects of ZH, $CaCl^2$ injection, and the ZH × $CaCl^2$ injection interaction by aging period.

[5]Traits scored on an 8-point scale: 1 = extremely dry, tough, bland, uncharacteristic beef flavor, non-beef like; 8 = extremely juicy, tender, intense, characteristic beef flavor, beef-like.

[6]Scored on a 5-point scale: 1 = none; 5 = extremely off flavor.

ZH treatment. On the other hand, Garmyn et al. (2010) did not observe an interaction between ZH treatment (0 or 8.3 mg/kg ZH) and postmortem aging (14 and 21 d) on sensory traits of strip loin steaks scored by trained panelists. However, ZH supplementation resulted in lower scores for juiciness and tenderness of strip steaks, along with a greater amount of detected connective tissue by trained panelists (Garmyn et al., 2010).

Previous studies have shown injecting LM with 200 m*M* $CaCl_2$ at 5% (wt/wt) at 24 h postmortem can result in tenderization without compromising other palatability traits, such as off flavor (Wheeler et al., 1993; Diles et al., 1994; Kerth et al., 1995; Lansdell et al., 1995). In the current study, $CaCl_2$ resulted in greater scores from trained panelists for flavor intensity, but they also detected greater off flavor due to $CaCl_2$, regardless of postmortem aging period. Although direct comparisons cannot be made, the current results align with the findings from consumer trials. Consumers rated $CaC1_2$–injected steaks with greater tenderness and superior flavor intensity ratings compared with the control (Hoover et al., 1995). Additionally, consumers responded favorability to $CaC1_2$–products, indicating they thought those products were more tender, juicy, and palatable with more flavor desirability and no off flavor compared with the controls (Miller et al., 1995). Moreover, Carr et al. (2004) found consumers were willing to pay $0.95/kg more for steaks marinated with $CaCl_2$ with respect to control steaks.

In conclusion, increased postmortem aging decreased WBSF regardless of ZH supplementation; however, WBSF and SSF values from ZH steaks were greater than non-ZH steaks during all postmortem aging periods. Although a greater percentage (91 vs. 71%)

Table 10. Effects of zilpaterol hydrochloride (ZH)[1] and $CaCl_2$ injection (INJ)[2] on trained sensory traits at 21 d postmortem

	No ZH		ZH			*P*-values[4]		
Trait	No INJ	INJ	No INJ	INJ	SEM[3]	ZH	INJ	Z × I
Initial juiciness[5]	5.87	5.82	5.78	5.89	0.13	0.95	0.85	0.56
Sustained juiciness[5]	5.90	5.82	5.87	5.82	0.13	0.89	0.58	0.91
Initial tenderness[5]	6.11	6.27	5.89	6.08	0.14	0.13	0.21	0.92
Overall tenderness[5]	6.14	6.30	5.87	6.19	0.14	0.17	0.08	0.52
Flavor intensity[5]	6.42	6.55	6.32	6.59	0.08	0.65	<0.01	0.32
Beef flavor[5]	6.37	6.40	6.23	6.35	0.08	0.25	0.40	0.58
Overall mouthfeel[5]	5.51	5.47	5.37	5.45	0.10	0.43	0.89	0.56
Off flavor[6]	1.10	1.20	1.07	1.21	0.04	0.80	<0.01	0.59

[1]Zilpaterol hydrochloride (8.3 mg/kg on DM basis) was fed for 20 d with a 3-d withdrawal (Merck Animal Health, DeSoto, KS).

[2]Calcium chloride food grade at 200 m*M* at 5% (wt/wt); Tetra Technologies, The Woodlands, TX.

[3]Pooled (largest) SE of least squares means. Sample numbers: No ZH = 38; ZH = 40; No INJ = 39; INJ = 39.

[4]Observed significance levels for main effects of ZH, $CaCl^2$ injection, and the ZH × $CaCl^2$ injection interaction by aging period.

[5]Traits scored on an 8-point scale: 1 = extremely dry, tough, bland, uncharacteristic beef flavor, non-beef like; 8 = extremely juicy, tender, intense, characteristic beef flavor, beef-like.

[6]Scored on a 5-point scale: 1 = none; 5 = extremely off flavor.

of steaks were classified as tender (<4.6 kg) based on WBSF values for steaks from steers with no ZH supplementation when steaks were aged 7 d postmortem, over 90% of all steaks would be classified as tender by d 21 postmortem and 100% of steaks required less 4.6 kg of force to shear at 28 d postmortem. Conversely, $CaCl_2$–injection reduced shear force values (WBSF and SSF) in comparison with non-injected steaks. However, injection did not affect the percentage of steaks classified as tender (<4.6 kg) at any postmortem aging period.

According to trained panelists, ZH supplementation reduced tenderness of USDA Select strip loin steaks at 14 d, but they could not detect differences in any palatability traits due to ZH supplementation when steaks were aged 21 d. Injection of $CaCl_2$ resulted in improved tenderness when measured instrumentally; however, panelists only detected flavor differences compared with non-injected steaks. Although no interactions were found with ZH and $CaCl_2$, injecting USDA Select strip loins from ZH-fed cattle can help reduce the normal WBSF variation as it does in steaks from non-ZH-fed cattle. However, further research is needed to determine if any detrimental effects on shelf-life and color stability exist when injecting $CaCl_2$ into strip loins from cattle supplemented with or without ZH. This will dictate the optimum combination of postmortem technologies, like $CaCl_2$ injection with postmortem aging, to most effectively improve tenderness in cattle supplemented with ZH.

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