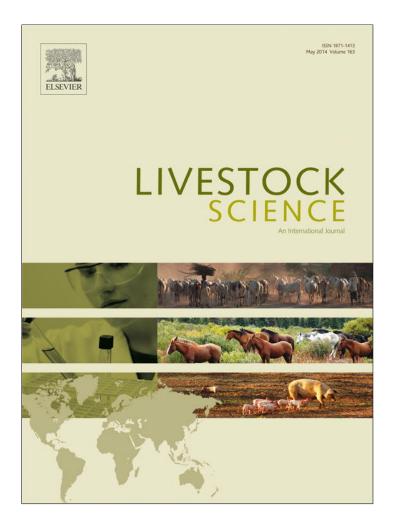
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Livestock Science 163 (2014) 140-149

Contents lists available at ScienceDirect

Livestock Science

journal homepage: www.elsevier.com/locate/livsci

Slaughter weight, sex and age effects on beef shear force and tenderness



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A R T I C L E I N F O

Article history: Received 15 January 2013 Received in revised form 5 February 2014 Accepted 8 February 2014

Keywords: Shear force Sensory panel Aging Chilling rate Sarcomere length

ABSTRACT

In Argentina, beef cattle prices decrease as slaughter weight increases regardless of animal age or carcass maturity, and this decrease is significantly greater in heifers (15%) than in steers (5%). The objectives of the present study were to (1) determine whether shear force and tenderness differ between heifers of different slaughter weight, and between heifers, steers and cull cows of similar slaughter weight; (2) evaluate whether such differences, if any, could be overcome by extending beef aging period; and (3) understand the main causes of such differences in beef shear force and tenderness. Meat from heavy heifers (H-HEIFER, 381-420 kg BW) was compared to meat from light heifers (L-HEIFER, 300-340 kg BW), steers (STEER, 391–450 kg BW) or cull cows of similar weight (COW). At slaughter, carcass characteristics were determined and Longissimus thoracis (LM) and Gluteus medius (GM) muscle samples collected for shear force determination. Total and insoluble collagen, sarcomere length and intact troponin-T content, and sensory panel scores, were evaluated for LM. Aging effect was evaluated for shear force and sensory panel scores. Irrespective of muscle or aging period, increasing heifer slaughter weight did not impact negatively on beef shear force or tenderness levels; heifers and steers of similar slaughter weight had similar beef shear force and tenderness levels; whereas heifer beef was more tender (P < 0.05) than that from cow of similar slaughter weight. Tenderness differences were not overcome by extending the aging period. Based on a principal component analysis, shear force and tenderness differences appear to be associated to the variation in chilling rate and sarcomere length.

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http://dx.doi.org/10.1016/j.livsci.2014.02.003

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1. Introduction

Despite a need to increase beef production, Argentine domestic market favors low slaughter weights regardless of animal age or carcass maturity. As heifers' and steers' slaughter weight increases from 300 to 400 kg, price (\$/BW kg) is reduced in both cattle groups, but to a greater



extent in heifers (15% and 5%, respectively) (Mercado de Liniers, 2012). As a result of these negative relationships, steers commercialized through Mercado de Liniers² are evenly distributed through the different slaughter weight categories, whereas 58% of the heifers are commercialized at light weights (317 kg BW on average).

Tenderness is one of the main attributes used by consumers to define beef quality (Grunert et al., 2004) and, in general, consumers are willing to pay more for more tender beef (Boleman et al., 1997; Destefanis et al., 2008; Killinger et al., 2004a, 2004b; Lusk et al., 2001; Miller et al., 2001; Platter et al., 2005). Although tenderness is negatively correlated to animal age when evaluated using a wide range of ages (Shorthose and Harris, 1990; Schönfeldt and Strydom, 2011), a less clear association is observed when comparing smaller age ranges. Some studies (Shackelford et al., 1995; Wulf et al., 1996) showed negative effects of animal age on tenderness, whereas no such effects were observed by other authors (Field et al., 1996; Lawrence et al., 2001). Furthermore, increasing slaughter weight within a given ossification or maturity score has shown to have no effect or to improve the Meat Quality score (MQ) from the Meat Standard Australia prediction model (Watson et al., 2008). Increasing slaughter weight by 100 kg in Argentinean pasture fattening systems would increase animal age at slaughter by no more than five month.

When comparing beef tenderness from samples aged for 14 d, neither Zinn et al. (1970) nor Choat et al. (2006) observed differences between heifers and steers, but when aged for only 7 d, lower tenderness in heifers were observed by Choat et al. (2006). This suggests that initial differences in tenderness could be overcome if most of the post-rigor tenderization phase effect is allow to proceed (Koohmaraie and Geesink, 2006). It is worth noting that in all the above studies where animal age effect on tenderness was evaluated (Field et al., 1996; Lawrence et al., 2001; Shackelford et al., 1995; Wulf et al., 1996) samples were aged for 14 d; thus, initial differences in tenderness could have been minimized. Given that, in Argentina, beef is consumed within seven days after slaughter, tenderness differences could be observed when comparing beef from cattle of slaughter groups differing in weight, age or sex.

The objectives of the present study were to (1) determine whether beef shear force and tenderness differ between heifers of different slaughter weight, and between heifers, steers and cull cows of similar slaughter weight; (2) evaluate whether such differences, if any, could be overcome by extending beef aging period; and (3) understand the main causes of such differences in beef shear force and tenderness between slaughter groups.

2. Material and methods

2.1. Animals and treatments

A total of forty Aberdeen Angus cattle from the same cow-calf herd were used. At weaning (April–May) 20 female calves were selected and randomly assigned to one of two treatments defined by the slaughter weight: light heifers (L-HEIFER; 300-340 kg BW) and heavy heifers (H-HEIFER, 381-420 kg BW). At the same time, 10 male calves were selected to be slaughtered when they reached the weight to be classified as heavy steers (STEER, 391-430 kg BW). In the next production cycle (March of the following year), after rectal palpation of the cows from the original herd, ten non-pregnant cows that had at least two lactations were selected (**COW**). Animals (n=10 per)slaughter group) corresponding to a given treatment (slaughter group) were slaughtered on the same date. Heifers and steers from each treatment were slaughtered when the group's average and median BW were within its slaughter weight range; in turn, cows were slaughtered after two months of fattening.

2.2. Animal management

Animals were assigned to a 10 ha consociate temperate pasture, comprised by Medicago sativa, Lolium perenne, Bromus unioloides, and Festuca arundinacea as the main forage species. Animals grazed under a rotational system with the objective to optimize body weight gain and forage utilization. Animal initial and final weight were determined as the average of its individual weight determined in two consecutive days at the beginning and end of their fattening period. During the fattening period, animals were weighed every 21 d. When the average BW of the animals assigned to a given slaughter group reached the defined slaughter weight, or after two months of fattening for COW, animals were shipped to a commercial slaughter house. Following regular practices in Argentina, animals were rounded up at 03:00 pm and, after an overnight feed withdrawal, they were shipped at 01:00 pm, and arrived in the slaughter house 1.5 h later. At 7:00 am of the following day, animals were slaughtered. They were stunned with a captive bolt without being electrically immobilized or stimulated. Carcasses were moved into the chiller within one hour after being stunned.

2.3. Measurements and sampling at harvest

At harvest, hot carcass and kidney fat weight (kg) were registered within 45 min postmortem. At 3 h postmortem, *Longissimus thoracis* muscle pH (pH@3 h) and temperature (Temp@3 h) were measured between the 12th and 13th ribs using a portable pH-meter (Sper Scientific model 850081) with a temperature penetration probe and a pH penetration probe type 13 (Testo). At 24 h, postmortem pH determination was repeated (ultimate pH).

Twenty-four hour postmortem, the rib section was removed from the left side of each carcass by cutting between the 6th and 7th thoracic vertebrae and 13th thoracic and 1st lumbar vertebrae. At the same time, the top sirloin was removed from the left carcass side. Both primals were then transported to the meat laboratory at the Instituto Nacional de Tecnología Agropecuaria-Estación Experimental Agropecuaria Balcarce (INTA-EEA Balcarce; Balcarce, Buenos Aires, Argentina) and stored at 2 °C till the following day for fabrication.

² Buenos Aires live cattle market that commercializes approximately 6000 heads per day (**2006–2011**). Daily prices for the different categories are used as reference prices for other cattle transactions in Argentina.

2.4. Sample preparation and analysis

The whole rib section (7-13th thoracic vertebrae) was cut between the joints of the 8-9th and 11-12th vertebrae to obtain the 9-11th rib section. On the cut face corresponding to the 11th rib, subcutaneous fat thickness and LM area were determined. A 2.5 cm-thick steak was obtained from the 12–13th rib section (from cranial to caudal), for Longissimus thoracis muscle total fat analysis, after removing all external fat and connective tissue. Two additional, 0.5 cm-thick steaks were obtained from the remainder of the 12-13 rib section; one was stored for sarcomere length determinations, and the other one was stored for intact troponin-T (Tn-T) quantification after being aged for 7 d at 2 °C. The Longissimus thoracis muscle from the 9-11 rib section was cut into four 2.5 cm-thick steaks for Warner-Bratzler shear force (WBSF) evaluation. Steaks were randomly assigned to one of four aging periods (3, 7, 14 and 28 d). Three 2.5 cm-thick steaks were obtained from the 7-8 rib section for sensory panel evaluation; steaks were randomly assigned to one of three aging periods (3, 7, and 14 d).

The *Gluteus medius* muscle was obtained from the top sirloin (rump) and four 2.5 cm-thick steaks were obtained and randomly assigned to one of four aging periods (3, 7, 14 and 28 d) for WBSF evaluation.

All steaks were vacuum packaged and stored at -20 °C after its corresponding aging period at 2 ± 2 °C. Steaks assigned to *Longissimus thoracis* muscle total fat analysis or sarcomere length evaluation were immediately stored at -20 °C.

2.5. Longissimus thoracis muscle total fat content

Prior to analysis, steaks for ether extract analysis were pulverized while frozen using a grinder for hard materials (Arcano). Ether extract content was determined using a XT15 Extractor (ANKOM Technology, Macedon, NY, USA).

2.6. Warner-Bratzler shear force

Warner-Bratzler shear force analysis was conducted according to AMSA (1995)'s guidelines. Steaks were thawed overnight at 4 °C, and broiled to an internal temperature of 71 °C on Farberware Open Hearth grills (Farberware, Bronx, NY). Internal temperature was controlled using a multiscan digital thermometer (Scanning Thermometer, Digi-Sense, ColePalmer). Steaks were weighed before and after broiling in order to calculate cooking loss percentages. Steaks were cooled to room temperature, six 1.27 cm-diameter cores were removed in parallel to the muscle fiber, and cores were sheared perpendicular to the fibers' longitudinal axis. Peak shear force was measured using a digital force gauge (BFG 500N, QuantrolTM, Dillon/Quality Plus, Inc., Kansas City, MO, USA), equipped with a WBSF attachment at a crosshead speed of 200 mm/min (Warner-Bratzler meat shear, G-R Manufacturing CO., Manthattan, KS, US).

2.7. Trained sensory panel

Steaks were thawed for 24 h at 5 °C, cooked to an internal temperature of 71 °C on a preheated double contact plate electric grill to 200 °C, trimmed of all external fat and major connective tissue, and cut into $1 \times 1 \times 2.54$ -cm³ samples. Samples were served to a fivemember trained sensory panel seated in individual booths under fluorescent lighting. Each panel member, trained according to AMSA (1995), received two subsamples per sample according to the general AMSA (1995)'s guidelines and general rules of IRAM (Instituto Argentino de Normalización y Certificación; http://www.iram.org.ar) for sensory analysis. Five or six samples were served in randomized order at approximately 3-min intervals in each session. A total of twenty three sessions were performed (one a day). Steak samples were evaluated for initial tenderness, overall tenderness, juiciness, beef flavor and beef odor intensity, and amount of connective tissue using a 9-point scale for each independent attribute where 1=extremely tough, dry, bland (beef flavor and beef odor), no off-flavor, and none to 9=extremely tender, juicy, intense (beef flavor and beef odor), intense off-flavor, and abundant. Panel members were also provided with a salt-free cracker and water for cleansing the palate between samples.

2.8. Intact troponin-T (tn-T)

Longissimus thoracis muscle samples from all animals (n=40) aged for 7 days were used for Tn-T degradation analysis. Intact Tn-T was determined using western blotting procedures according to Huff-Lonergan et al. (1996) with minor changes. Briefly, whole muscle extracts were prepared homogenizing 1 g of muscle in 10 ml of deionized water. Homogenate protein content was determined using Microplate Spectrophotometer equipped with reader type Epoch (No. 257878; Biotek). Samples were adjusted to constant protein level (3 mg/ml) using distilled deionized water. Thirty micrograms protein from the whole muscle preparation were loaded onto 12% polyacrylamide slab separating gels with a 5% polyacrylamide stacking gel (BioRad). Gels were run at room temperature (approximately 20 °C) for 45 min at a constant voltage of 200 and 60 mA per gel. Gels were transferred to Immuno-Blot PVDF Transfer membranes (No. 55518; Thermo Scientific) using a Mini Trans-Blot Cell (Bio-Rad) for 1 h at 100 V and 340 mA. Complete transfer of proteins in the 30-kDa molecular weight range was verified using a Kaleidoscope prestained molecular weight marker (BioRad) and subsequent staining of membranes after transfer. The Western blotting procedure used anti-troponin-T JLT-12 (Sigma, St. Louis, MO; 1:7000) as the primary antibody, anti-mouse IgG labeled with peroxidase (Sigma; 1:7000) as the secondary antibody, and a chemiluminescent detection system (Pierce Super Signal Substrate; Pierce, Rockford, IL). Each blot included a sample as internal standard for reference. Protein bands were quantified using the ImageQuant 400 digital analysis system (GE). Bands of Tn-T within molecular weights of 41.7 and 39.9 kDa were considered to be intact Tn-T and the sum of their intensities was quantified (Weaver et al., 2008). The intact Tn-T content was estimated by expressing the density

of the intact Tn-T band from each sample in a given blot relative to the density of the intact Tn-T band in the same blot of a sample used as internal standard for reference.

2.9. Sarcomere length

The helium-neon laser diffraction method described by Cross et al. (1981) was used for sarcomere length evaluation on *Longissimus thoracis* muscle samples. The sarcomere length value from each sample was the average of 10 readings.

2.10. Total and insoluble collagen content

Total collagen content on *Longissimus thoracis* muscle was estimated by determining hydroxyproline concentration using the rapid procedure described by Bergman and Loxley (1963). *Longissimus thoracis* muscle insoluble collagen content was determined according to a procedure adapted from Hill (1966). Shortly, it consisted in heating samples in a water bath for 70 min at 77 °C, and centrifugation at 6000g for 10 min at 2 °C. Collagen content of the pellet was estimated through the determination of hydroxyproline concentration as described above.

2.11. Statistical analysis

All data were analyzed using the MIXED procedure of SAS (SAS Inst. Inc., Cary, NC, USA). All, but sensory panel and shear force, data were analyzed as a complete randomized model with slaughter group as fixed effect and animal nested within slaughter category as random effect. A similar model was used for sensory panel data analysis in addition to aging period (postmortem days) and slaughter group by aging period interaction as fixed effects; animal nested within slaughter category was the subject on which repeated measures across aging periods were taken. Shear force data were analyzed as a split-plot design with repeated measures; the model included slaughter group (main plot), muscle (sub-plot), aging period and their interaction as fixed effects. The random effect of animal nested within slaughter group was used to test slaughter group effect and as the subject on which repeated measures across aging periods were taken. When significant main or interaction effects were detected, leastsquare means were separated using the PDIFF option adjusted by Tukey from PROC MIXED LSMEAN statement. Statistical significance was set at $P \le 0.05$.

The relationship between main variables was determined with principal component analysis (Destefanis et al., 2000) using ONES as prior communality estimates with the FACTOR procedure of SAS, and using the principal axis method to extract the components. To determine which independent carcass trait variables better explained the observed variation in *Longissimus thoracis* muscle Temp@3 h, multiple regression analysis was performed applying the stepwise method to choose the independent variables to be maintained in the model (P < 0.10).

3. Results

Heavy heifers reached the average slaughter weight after 465 d on pasture, whereas L-HEIFER and STEER reached it in 317 and 402 d, respectively; COW were slaughtered after 86 d on the trial pasture. Final live weight for H-HEIFER was 76 kg greater than for L-HEIFER, similar to STEER and 33 kg lighter than COW (Table 1; P < 0.001); final live weight was similar for STEER and COW. Average daily gain was similar between H-HEIFER, L-HEIFER and STEER and lowest in COW (P < 0.01).

Slaughter groups differed in all evaluated carcass traits (P < 0.05). Hot carcass weight did not differ between H-HEIFER, STEER and COW, but was 49 kg lighter for L-HEIFER. Dressing percentage was highest for H-HEIFER and lowest in L-HEIFER and COW. Variables associated to carcass fatness (subcutaneous fat thickness, kidney fat and Longissimus thoracis muscle total fat content) were greater in H-HEIFER than in L-HEIFER and STEER. Subcutaneous fat thickness in COW was similar as in H-HEIFER and greater than in L-HEIFER and STEER; in contrast, kidney fat content in COW was similar as in L-HEIFER and STEER and lower than in H-HEIFER. Longissimus thoracis muscle total fat content in COW was intermediate, not differing from any of the other slaughter groups evaluated. Longissimus thoracis muscle area in H-HEIFER and L-HEIFER did not differ from the largest LM area observed in STEER neither from the smallest one observed in COW. Heavy heifers, light heifers and steers had the highest Temp@3 h, but heavy heifers had the lowest pH@3 h and the other two slaughter groups, the highest pH@3 h. Cull cows had the lowest Temp@3 h, (5 °C lower) and intermediate pH@3 h value. Heavy heifers presented the lowest ultimate pH of all four slaughter categories, and steers, the highest one. Ultimate pH from light heifers and cull cows were intermediate, not differing from that in H-HEIFER or STEER.

None of the variables (total and insoluble collagen content, insoluble to total collagen content, sarcomere length, intact Tn-T content) associated with the main factors that would define meat tenderness (basal tenderness, shortening phase, and tenderization phase) differed between slaughter groups (P > 0.05; Table 2).

No main effects (slaughter group, muscle, and aging period) interactions or muscle effects were significant on shear force (P > 0.05; Table 3). Shear force improved (P < 0.001) when extending aging period. Extending the aging period from 3 d to 7 d and from 7 d to 28 d reduced shear force from 3.87 kg to 3.47 kg and to 3.09 kg, respectively. Shear force obtained in samples aged for 14 d (3.22 kg) did not differ from that obtained at 7 or 28 d. Among slaughter groups, shear force was lower (P < 0.001) in H-HEIFER than in COW and intermediate in L-HEIFER and STEER, not differing from either H-HEIFER or COW.

Sensory panel scores for *Longissimus thoracis* muscle connective tissue amount where lower in H-HEIFER than in COW (P=0.044), whereas scores in L-HEIFER and STEER did not differ from either one. Extending aging period reduced (P=0.007) connective tissue scores. Beef flavor intensity was not affected (P > 0.05) by slaughter group nor by aging period or their interaction. Juiciness score was greater (P=0.041) in STEER than in COW, but none of them

144

J. Lucero-Borja et al. / Livestock Science 163 (2014) 140-149

Table 1

Animal and carcass traits from heifers of different slaughter weight, steers and cull cows from grazing systems.

	Slaughter grou	MSE ^e	P-value			
	H-HEIFER	L-HEIFER	STEER	COW		
Animal traits						
Initial body weight, kg	164 ^a	159 ^a	170 ^a	408 ^b	6.4	< 0.0001
Final body weight, kg	385 ^b	309 ^c	389 ^{a,b}	426 ^a	10.3	< 0.0001
Pre-shipping body weight, kg	379 ^b	318 ^c	384 ^{a,b}	418 ^a	10.03	< 0.0001
Body weight gain, kg/d	0.47 ^a	0.47 ^a	0.55 ^a	0.21 ^b	0.04	< 0.0001
Carcass traits						
HCW ^f , kg	211 ^a	160 ^b	205 ^a	213 ^a	5.54	< 0.0001
Dressing percentage ^g , %	55.74 ^a	50.50 ^c	53.40 ^b	50.91 ^c	0.49	< 0.0001
Subcutaneous fat thickness, mm	7.5 ^a	4.2^{b}	5.1 ^b	7.9 ^a	0.70	< 0.0001
Kidney fat, %	2.76 ^a	1.32 ^b	1.40 ^b	1.51 ^b	0.15	< 0.0001
LM total fat ^h , %	11.60 ^a	7.74 ^b	7.13 ^b	9.70 ^{a,b}	0.89	0.0042
LM area ⁱ , cm ²	53.82 ^{a,b}	50.75 ^{a,b}	57.68 ^a	48.73 ^b	1.89	0.0114
Ultimate pH	5.66 ^b	5.70 ^{a,b}	5.86 ^a	5.80 ^{a,b}	0.05	0.0230
pH@3 h ^j	6.06 ^c	6.60 ^a	6.68 ^a	6.34 ^b	0.06	< 0.0001
Temp@3 h ^k , °C	24.71 ^a	24.50 ^a	24.28 ^a	19.56 ^b	0.69	< 0.0001

^{,b,c} Lsmeans with uncommon superscript within a row differ (P < 0.05).

^d H-HEIFER, Heavy Heifers; L-HEIFER, Light Heifers; STEER, Steers; COW, Cull Cows.

^e MSE, mean standard error.

^f HCW=hot carcass weight.

^g Dressing percentage=HCW/Pre-shipping body weight \times 100.

^h LM total fat=Longissimus thoracis muscle ether extract content, DM basis.

ⁱ LM area=Longissimus thoracis muscle area.

^j pH@3 h=Longissimus thoracis muscle pH determined at 3 h postmortem.

^k Temp@3 h=Longissimus thoracis muscle temperature determined at 3 h postmortem.

Table 2

Longissimus thoracis muscle sarcomere length, content of intact Troponin-T (Tn-T), total and insoluble collagen content and insoluble to total collagen ratio from heifers of different slaughter weight, steers and cull cows from grazing systems.

	Slaughter catego	ory ^a			MSE ^b	P-value
	H-HEIFER	L-HEIFER	STEER	COW		
Sarcomere length, µm	2.02	2.02	2.00	1.98	0.018	0.4527
Intact Tn-T, %	1.45	1.84	1.63	2.27	0.73	0.8712
Collagen, mg/g fresh tissue						
Total	4.27	4.35	5.97	4.58	0.525	0.0935
Insoluble	2.41	2.76	3.21	3.34	0.394	0.3271
Insoluble/Total ratio	0.59	0.63	0.60	0.74	0.066	0.3863

^a H-HEIFER, Heavy Heifers; L-HEIFER, Light Heifers; STEER, Steers; COW, Cull Cows.

^b MSE, mean standard error.

differed from heavy or light heifers. Juiciness scores were also affected by aging period (P < 0.001). Samples aged for 7 d (6.17 ± 0.12) were juicer than when aged for 14 d (5.67 \pm 0.12) and these from those aged for 3 d (5.03 \pm 0.12). Slaughter group effects on initial tenderness, overall tenderness and odor scores were affected by aging period (P=0.015, P < 0.001, and P < 0.001, respectively). When samples were aged for 3 d, initial and overall tenderness scores did not differ between slaughter groups, but when aged for 7 d, they were greater in H-HEIFER than in COW and intermediate for L-HEIFER and STEER. When aging period was extended to 14 d, initial tenderness scores were greater in H-HEIFER and STEER than in COW, and overall tenderness scores were greater in H-HEIFER, L-HEIFER and STEER than in COW. Within slaughter groups, extending the aging period only increased initial tenderness in H-HEIFER and overall tenderness in H-HEIFER and STEER when extended from 3 to 7 d.

Loading scores for the six principal components that explained 75% of total variance in the 20 variables included in the PCA are presented in Table 4. Loading and score vectors for the first and second principal components (CP1 and CP2), that explained 26 and 14% of total variance, are plotted in Fig. 1. Shear force and overall tenderness at all evaluated aging periods were the variables best represented on CP1 in conjunction with Longissimus thoracis muscle Temp@3 h. Overall tenderness variables and Temp@3 h were grouped together in the loading plot and opposed to shear force variables. Although closer to the axis origin than the above mentioned variables, sarcomere length, LM area, and kidney fat content were also well represented in CP1. Of these variables, LM area was positively associated with overall tenderness, while LM area and sarcomere length were negatively associated to shear force (Table 5). The second principal component was composed by carcass weight and fatness, and pH@3 h. Positive associations were

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J. Lucero-Borja et al. / Livestock Science 163 (2014) 140-149

Table 3
Shear force and sensory panel evaluation from heifers of different slaughter weight, steers and cull cows from grazing systems.

	Slaughte	r group ^d			MSE ^e	P-value ^f						
	H- HEIFER	L- HEIFER	STEER	COW		SG	MUSC	AP	$SG \times MUSC$	$SG \times AP$	$MUSC \times AP$	$SG \times AP \times MUSC$
Shear force, kg	2.96 ^b	3.23 ^b	3.30 ^b	4.16 ^a	0.17	< 0.001	0.161	< 0.001	0.084	0.458	0.091	0.879
Sensory panel evaluation Connective tissue	1 ^g 2.94 ^b	3.77 ^{a,b}	3.65 ^{a,b}	3.93 ^a	0.25	0.044		0.007		0.343		
amount	2.54	5.77	5.05	J.3J	0.25	0.044		0.007		0.545		
Beef flavor	5.42	5.08	5.10	5.32	0.15	0.338		0.527		0.482		
Juiciness	5.57 ^{a,b}	5.74 ^{a,b}	5.97 ^a	5.25 ^b	0.17	0.041		< 0.001		0.089		
Beef odor												
3 d	5.80 ^a	4.58 ^{z,b}	4.81 ^b	5.56 ^a	0.19	0.001		0.851		< 0.001		
7 d	5.28 ^a	5.41 ^{y,a}	4.49 ^b	5.48 ^a								
14 d	5.10	5.44 ^y	4.88	5.51								
Initial tenderness												
3 d	5.97 ^y	5.74	6.09	5.29	0.22	< 0.001		< 0.001		0.009		
7 d	7.05 ^{z,a}	6.32 ^{a,b}	6.16 ^{a,b}	5.89 ^b								
14 d	7.20 ^{z,a}	6.45 ^{ab}	6.70 ^a	5.56 ^b								
Overall tenderness												
3 d	6.44 ^y	6.31	5.72 ^y	5.95	0.22	< 0.001		< 0.001		< 0.001		
7 d	7.56 ^{z,a}	6.74 ^{a,b}	6.75 ^{z,a,b}	6.53 ^b								
14 d	7.55 ^{z,a}	6.93 ^a	7.20 ^{z,a}	5.86 ^b								

 $^{\rm a,b,c}$ Lsmeans with uncommon superscript within a row differ (P < 0.05).

^{y, z} Lsmeans with uncommon superscript within a column differ (P < 0.05).

^d H-HEIFER, Heavy Heifers; L-HEIFER, Light Heifers; STEER, Steers; COW, Cull Cows.

^e MSE, mean standard error.

^f SG, slaughter group; MUSC, muscle (*Longissimus thoracis vs. Gluteus medius*); AP, aging period.

^g 9-point scale where 1=extremely tough, dry, bland (beef flavor and beef odor), no off-flavor, and none to 9=extremely tender, juicy, intense (beef flavor and beef odor), intense off-flavor, and abundant.

Table 4

Mean, standard deviation (SD) and loadings (coefficients in the eigen vectors) for the first six principal components of the carcass traits and *Longissimus thoracis* muscle quality measurements included in the principal component analysis.

Variable	Mean	SD	Principal component									
			1	2	3	4	5	6				
Carcass traits												
HCW, kg	197.53	27.54	0.22	0.62	0.49	0.16	0.07	0.35				
LM area (LMA), cm ²	52.74	6.69	0.43	-0.14	0.46	0.37	-0.28	0.41				
Subcutaneous fat thickness (FT), mm	6.17	2.66	0.15	0.84	0.21	0.09	0.11	-0.01				
Kidney fat (KF), %	1.75	0.76	0.54	0.60	-0.11	0.09	0.06	0.09				
LM total fat (LMF), %	9.04	3.23	0.39	0.72	-0.26	-0.06	0.15	-0.24				
Temp@3 h, °C	23.26	3.02	0.63	-0.34	0.01	0.42	-0.06	0.13				
pH@3h	6.42	0.31	-0.41	-0.58	0.30	0.10	-0.19	-0.27				
Ultimate pH (pHu)	5.75	0.16	-0.24	0.12	0.72	-0.02	-0.06	-0.33				
Longissimus thoracis muscle quality mea	surements											
Intact Tn-T (Tn-T), %	1.80	2.24	0.01	0.09	0.69	-0.46	-0.07	-0.16				
Sarcomere lenght (Sl), μm	2.01	0.06	0.46	-0.11	0.00	-0.38	0.35	-0.35				
Collagen												
Total (Ct), %	4.79	1.74	-0.10	-0.22	0.03	0.56	0.73	-0.16				
Insoluble (Ci), %	2.93	1.25	-0.43	-0.25	0.15	-0.06	0.76	0.30				
Insoluble:total ratio (Cr)	0.64	0.21	-0.35	-0.09	0.27	-0.61	0.12	0.56				
Warner–Bratzler shear force, kg												
At 3 d of aging (SF3)	4.08	1.38	-0.73	0.11	-0.31	0.04	-0.31	0.09				
At 7 d of aging (SF7)	3.53	1.00	-0.76	0.19	0.09	0.20	0.11	0.07				
At 14 d of aging (SF14)	3.22	0.78	-0.71	0.17	-0.07	0.26	-0.17	0.02				
At 28 d of aging (SF28)	3.05	0.89	-0.74	0.32	0.07	0.33	-0.01	0.01				
Overall tenderness score												
At 3 d of aging (OT3)	5.77	0.88	0.52	-0.17	0.52	0.33	-0.05	-0.21				
At 7 d of aging (OT7)	6.36	0.78	0.74	-0.05	-0.10	-0.09	-0.04	0.04				
At 14 d of aging (OT14)	6.48	0.94	0.68	-0.30	-0.11	0.08	0.01	0.33				

HCW, hot carcass weight; LMA, *Longissimus thoracis* muscle area; KF, carcass kidney fat content; LMF, *Longissimus thoracis* muscle total fat content; Temp@3 h, *Longissimus thoracis* muscle temperature at 3 h postmortem; pH@3 h, *Longissimus thoracis* muscle pH at 3 h postmortem.

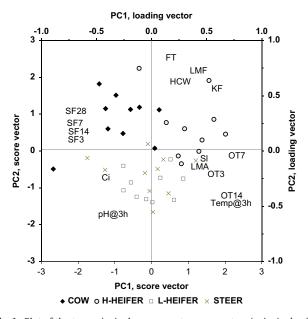


Fig. 1. Plot of the two principal components score vectors (principal axis) and loading vectors (secondary axis). Only variables with loadings with an absolute value greater than 0.4 for any of two principal components were represented. HCW, hot carcass weight; LMA, *longissimus* muscle area; FT, subcutaneous fat thickeness; KF, carcass kidney fat content; LMF, *longissimus* muscle total fat content; Temp@3 h, *longissimus* muscle temperature at 3 h postmortem; pH@3 h, *longissimus* muscle pH at 3 h postmortem; Ci, insoluble collagen content; SF3, SF7, SF14, and SF28, *longissimus* muscle shear force at 3, 7, 14 and 28 d of aging; OT3, OT7, and OT14, *longissimus* muscle overall tenderness scores at 3, 7, and 14 d of aging. H-HEIFER, Heavy heifer; L-HEIFER, Light heifer; STEER, steer; and COW, cull cow.

observed between hot carcass weight, total LM fat content, subcutaneous fat thickness and kidney fat content. In turn, negative associations were detected between these variables and pH@3 h.

4. Discussion

The main objectives of the present study were to determine shear force and tenderness differences between heifers of different slaughter weight or between heifers, steers and cows of similar slaughter weight. Therefore, in order to avoid confounded effects due to differences in growth rate or animal growth potential (Perry and Thompson, 2005), heifers were randomly assigned to one of the two slaughter categories for heifers (L-HEIFER or H-HEIFER) at the beginning of the fattening period. In addition, they were managed as a unique group, and slaughtered when the average live weight (BW) of the slaughter group reached the target slaughter weight. For the same reason, steers from the same herd were managed as a group with the heifers and slaughtered when their average BW reached the objective slaughter weight. By doing so it was possible to obtain similar ADG among these slaughter groups, but treatment effects were confounded with age at slaughter effects; therefore caution is required when discussing and concluding from results obtained in the present study.

Irrespective of the muscle considered, both shear force and trained sensory panel results showed that meat from heavy heifers was more tender than meat from cull cows of similar carcass weight and as tender as or even more tender than meat from lighter heifers or steers of similar carcass weight. Differences observed between heavy heifers and cull cows are in agreement with the negative correlation observed by Shorthose and Harris (1990) between animal age and tenderness when a broad age range was considered (1-60 months) and with the results of Stelzleni et al. (2007) when comparing shear force from cull beef and dairy cows to A-maturity, USDA select steers. The lack of differences observed between heavy and light heifers contradicts this negative correlation between age and tenderness and other previous reports where age had a negative effect on shear force, tenderness or on both (Shackelford et al., 1995; Wulf et al., 1996), but agrees with other studies (Field et al., 1996; Lawrence et al., 2001; Pflanzer and Felício, 2009). No differences between heifers and steers on meat shear force or tenderness were either reported by others (Choat et al., 2006; Lawrence et al., 2001; Zinn et al., 1970).

Both Shackelford et al. (1995) and Wulf et al. (1996) attributed differences in tenderness to a potential increase in collagen content with increasing age at slaughter and carcass maturity. This hypothesis was not confirmed by our data. Despite the age differences between cull cows and the other slaughter groups, no differences were observed for total or insoluble collagen content in the Longissimus thoracis muscle. Furthermore, the principal component analysis only suggests a minor association of insoluble collagen content with tenderness (Fig. 1; Table 5). Using a large data set (N=765) Chriki et al. (2012) observed that total and insoluble collagen content were negatively correlated to tenderness, but only with a small coefficient (r = -0.15 to -0.20). Chriki et al. (2012) suggested that one possible reason for the conflicting results observed in the literature when trying to associate collagen content to tenderness scores is that this association could only be detected when using a large data set.

Shear force and tenderness differences between heavy heifers and cull cows could not be explained by slaughter group differences in sarcomere length, intact Tn-T content or LM fat content, but they were associated to differences in Temp@3h postmortem. The relative rate of postmortem pH and temperature decline has been demonstrated to affect meat tenderness (Gardner et al., 2005). A quadratic relationship between carcass temperature at pH 6 and tenderness was observed in achilles tendon hanged sheep carcasses (Thompson et al., 2005); tenderness was maximized when temperature at pH 6 was 21 °C but drastically reduced at extreme temperatures ($< 10 \degree C$ or $> 30 \degree C$). Our data suggest that the relative rates of pH and temperature decline were close to being optimal for heavy heifers (24 °C at pH 6). As the lower Temp@3 h observed in cull cows was not associated with a lower pH, a greater muscle shortening could be expected in this slaughter group as compared with the remaining ones. The loading scores for PC1 and PC2 indicate that Temp@3 h is negatively associated with shear force values and positively associated with overall tenderness score and sarcomere length. Furthermore, based on sample distribution on the plot of the first two principal component score vectors (Fig. 1), heavy heifers had greater overall tenderness

 Table 5

 Pearson correlation coefficients between selected meat-quality related variables.

	Variables ^a	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
2. Kidney fat content 0.40° 3. Subcutaneous fat thickness 0.66°*** 0.48°* 4. Total LM fat content ^b 0.28° 0.62°*** 0.61**** 5. pH@3 h ^c -0.33° -0.60°*** 0.39° -0.54*** 6. LM area ^b 0.40° 0.12 -0.01 -0.17 0.01 7. Temp@3 h ^d -0.02 0.26 -0.14 -0.03 -0.02 0.54*** Longissimus thoracis muscle quality measurements 8. Sarcomere length -0.05 0.10 -0.04 0.27° -0.13 0.02 0.18 9. Insoluble collagen content -0.08 -0.28° -0.15 -0.31° 0.16 -0.22 -0.19 -0.04 Warner-Bratzler shear fore 10. At 3 d of aging -0.27° -0.19 -0.13 -0.21 0.17 -0.31° -0.46°* -0.43°* 0.04 11. At 7 d of aging -0.02 -0.39° 0.09 -0.22 0.17 -0.25 -0.51* -0.39° 0.13 0.46°* 0.47″* 13. At 28 d of aging 0.12 -0.17 0.10 -0.08 0.14 -0.14 -0.39° -0.46°* 0.22 0.57** 0.62** 0.65*** Overall tenderness score 14. At 3 d of aging 0.16 0.16 0.09 -0.03 0.08 0.43** 0.12 0.50** -0.16 -0.47°* -0.33° -0.37° -0.29° 15. At 7 d of aging 0.04 0.35° 0.02 0.23 -0.36° 0.18 0.17 0.27° -0.29° -0.52** -0.57** 0.62** 0.65*** * P < 0.001. ** P < 0.001. ** P < 0.001.																
3. Subcuraneous fat thickness 0.66 ⁺⁺⁺⁺ 0.48 ⁺⁺⁺ 0.66 ⁺⁺⁺⁺⁺ 0.28 ⁶ 0.62 ⁺⁺⁺⁺ 0.61 ^{++++++++++++++ 0.04 for a constraint of the standard straint}																
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*** $P < 0.010.$ **** $P < 0.001.$ ***** $P < 0.0001.$ * $P < 0.0001.$ a Only variables with loadings of an absolute value > 0.4 in PC1 or PC2 from the PCA were included in this analysis.	16. At 14 d of aging	0.04	0.17	-0.16	0.06	-0.19	0.42**	0.16	0.44**	-0.15	-0.43**	- 0.56***	-0.48**	-0.51***	0.25	0.60
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^c pH@3 h, Longissimus thoracis muscle pH at 3 h postmortem.																
^d Temp@3 h, <i>Longissimus thoracis</i> muscle temperature at 3 h postmortem.	" Temp@3 h, Longissimus thora	<i>cis</i> muscle te	emperature a	t 3 n postmo	ortem.											

scores, associated with higher Temp@3 h, and longer sarcomeres than cull cows.

Rates of temperature decline have been associated to carcass weight (Thompson et al., 2006). In addition, despite the lack of association with beef tenderness, 7.6 mm of subcutaneous fat thickness would be required to reduce muscle shortening (Tatum et al., 1982). In the present study, heavy heifers and cull cows had similar carcass weight and subcutaneous fat thickness, which was close to the proposed threshold, but different LM area, kidney fat and total LM fat content. The plot of loading vectors from the two principal components (Fig. 1) suggests that, in addition to a strong positive association with overall tenderness and sarcomere length, Temp@3 h has a strong positive association with LM area and a moderate positive association with carcass kidney fat content. These two variables explained 40% of total Temp@3 h variation (30% and 10% respectively). On the other hand, PCA plot for the two principal components shows that leaner animals, L-HEIFER and STEER, had the highest pH@3 h values, which makes a moderate contribution to CP1. Lower rates of pH decline have been associated to lower muscular glycogen content at slaughter, to red-oxidative muscular fibers and faster temperature decline (Gardner et al., 2005). As L-HEIFER and STEER had the same Temp@3 h as H-HEIFER, the higher pH@3h in leaner categories could not be attributed to a faster temperature decline. As suggested by Wulf et al. (2002), L-HEIFER's and STEER's greater ultimate pH suggests that muscular glycogen content at slaughter could have limited the rate of pH decline.

Differences in LM total fat content did not affect shear force values, but could have affected sensory analysis of tenderness as suggested by Pflanzer and Felício (2009). Wheeler et al. (1994) and Wulf et al. (1997) observed small $(\leq 5\%)$ but positive associations between *Longissimus* thoracis muscle intramuscular fat/marbling and tenderness. Savell and Cross (1988) suggested that, in order to obtain beef with acceptable palatability, a 3% intramuscular fat content (fresh-tissue basis) is required; lower contents would result in tougher, drier, and less flavorful meat. In our study, only H-HEIFER was above the 3% threshold, and sensory analysis gave some evidence that the lower LM total fat content in L-HEIFER and STEER could have compromised, in part, meat tenderness. However, the multivariate analysis suggests that, if present, only a weak association exists between LM total fat content and shear force or tenderness.

The lack of slaughter group by aging period interaction effect on shear force is in line with the lack of cattle group effect on intact Tn-T content. This indicates that initial differences in tenderness could not be overcome by extending the aging period, and emphasizes the importance of controlling the relative rates of postmortem pH and temperature decline or using different hanging methods to reduce shortening, as it was suggested by Thompson et al. (2006), in order to maximize tenderness.

Muscle effects on shear force, not observed in our study, have been reported by others. Stolowski et al. (2006) observed that *gluteus medius* had lower shear force than *Longissimus thoracis* muscle with minimum aging (2 d), but that the latter had a greater aging response.

Gruber et al. (2006) observed lower shear force values in the gluteus medius than in the longgisimus of USDA Select carcasses, but not in fatter carcasses graded as upper twothird USDA Choice. A greater shear force decrease with aging in the Longissimus thoracis muscle than in the gluteus medius was also reported by Pringle et al. (1999) and Gruber et al. (2006). Gruber et al. (2006) concluded that muscles with greater initial shear force values have greater aging responses. As in our study, Stelzleni et al. (2007) did not find differences in WBSF between the Longissimus thoracis and gluteus medius when aged for 14 d. Although no muscle effects were observed in the present study, a similar trend for a muscle by aging period interaction was observed. Despite having higher initial (3 d) shear force values, shear force decrease by aging was greater in Longissimus thoracis muscle than gluteus medius.

5. Conclusions

Based on the shear force and sensory panel results obtained in the present study it can be concluded that, regardless of the aging period and muscle considered, increasing heifers' slaughter weight/age on grazing systems would not have a negative impact on beef shear force/tenderness as compared to slaughtering lighter heifers or steers of similar weight, whereas it would result in less shear force and more tenderness level than those obtained when cull cows are slaughtered. Shear force and tenderness differences appear to be generated by differences in chilling rate and sarcomere length and would not be overcome by extending the aging period.

Conflict of interest

None of the authors have any actual or potential conflict of interest including any financial, personal or other relationships with other people or organizations within three years of beginning the submitted work that could inappropriately influence, or be perceived to influence, their work.

Acknowledgments

The authors are grateful for the contribution made by Dr. Susan Duckett in reviewing the manuscript, and by Lic. Gabriela Cendoya and Mrs. Adriana Cano in performing the multivariate analysis. This study was funded by the Instituto Nacional de Tecnología Agropecuaria (INTA) PNCAR-012211, the Agencia Nacional de Promoción Científica y Tecnológica (ANPCyT) PICT2007-02240, the Instituto de Promoción de la Carne Vacuna Argentina (IPCVA), and the Universidad Nacional de Mar del Plata. None of the sponsors had any role in the study design; in the collection, analysis, and interpretation of data; in the writing of the report; and in the decision to submit the paper for publication.

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