TENDERNESS OF *BOS INDICUS* INFLUENCED CATTLE AS IMPACTED BY ANABOLIC IMPLANTS AND GENDER

A Thesis

by

JARRETT FRANKLIN HUDEK

Submitted to the Office of Graduate Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

May 2009

Major Subject: Animal Science

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Approved by:

Chair of Committee,	Jeff W. Savell
Committee Members,	Dan S. Hale
	Jason Sawyer
	Joe Townsend
Head of Department,	Gary Acuff

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ABSTRACT

Tenderness of *Bos indicus* Influenced Cattle as Impacted by Anabolic Implants and Gender. (May 2009)

Jarrett Franklin Hudek, B.S., Texas A&M University

Chair of Advisory Committee: Dr. Jeff W. Savell

Steers (n = 77) and heifers (n = 68) were assigned randomly to one of three treatment groups. Treatment groups were defined as: no implant, implanted twice with trenbolone acetate (Revalor S or H), or implanted twice with estrodial benzoate (Synovex S or H). Animals were fed to an estimated 10 mm backfat thickness and based on visual appraisal, were assigned a harvest date. Carcass characteristics, color space values, sarcomere length, fat and moisture determination, Warner-Bratzler shear force, and protein degradation were all measured. Implanted animals, as a whole, exhibited heavier hot carcass weights and larger ribeye areas than non-implanted animals. Animals implanted with Revalor displayed significantly lower marbling scores and lower yield grades than those from control or Synovex groups. The distribution of quality grades within treatment groups shifted, with implant groups displaying higher percentages of Select carcasses. Gender impacted percentage of extractable fat and marbling scores, with heifers displaying higher values than steers for both measurements. Both implant groups displayed higher ($P < 0.05$) Warner-Bratzler shear values following a 0- and 14-d aging periods. However, following the 21-d aging

period, differences in tenderness were no longer present between non-implanted and implanted animals. Synovex treated animals displayed longer ($P < 0.01$) sarcomere lengths than control or Revalor. Differences ($P < 0.001$) in protein degradation were found between treatment groups. Across gender groups, the non-implanted cattle displayed the greatest amount of degradation (62%), followed by Synovex (48%,) and lastly Revalor (33%), all of which were different ($P < .05$) from each other.

These results indicate that use of anabolic implants positively impacted lean muscle growth, yet was a detriment to quality. Also, tenderness was negatively impacted by the use of these compounds. However, this study found by aging product for at least 21 days, tenderness differences between implanted and non-implanted animals were significantly mitigated.

DEDICATION

I would like to dedicate this to my parents Steve and Judy. Mom, for teaching me that intelligence is a gift to be used, and dad for teaching me that it cannot be used effectively without a strong work ethic.

ACKNOWLEDGEMENTS

First and foremost I would like to extend my utmost appreciation to my committee members. Dr. Savell, thank you for being the chair of my graduate committee, as well as all of the support you have provided throughout my journey. Thank you to Dr. Hale, who always had a positive message and letting me know that he had faith in me. Thanks to Dr. Sawyer, who always made time to discuss the logistics and statistics of the project. Finally, thanks to Dr. Townsend who provided encouragement and support throughout my undergraduate and graduate degree.

The proteolysis data would not have been possible without the support of Tommy Wheeler, Peg Ekren, and Andy King at the U.S. Meat Animal Research Center in Clay Center, Nebraska. Thanks to you and your staff for the guidance and opportunity to learn new techniques.

Great appreciation goes to Willy Horne, who provided invaluable help to make this study possible. Appreciation is also expressed to my fellow graduate students, Eric Metteauer, James Dillon, Sarah West, Tiffany Muras, and Will Wiederhold who provided invaluable help and support throughout my time in graduate school. Extreme appreciation is expressed to Ashley Haneklaus who has been there to provide guidance, support, and encouragement throughout these last two years. My utmost appreciation also is expressed to Ray Riley and the workers at the Rosenthal Meat Science and Technology Center for all of your support throughout this project.

Also, I would like to give thanks to my parents, Steve and Judy, who have supported me throughout my life. Without them I would have not been able to accomplish everything that I have. Finally, I would like to express my eternal gratitude to my fiancé Meagan. Your unending love, support, and encouragement has been the greatest gift I have ever received.

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CHAPTER I

INTRODUCTION AND REVIEW OF LITERATURE

The use of growth promoting implants is widespread in the beef industry. Two commonly used anabolic compounds are trenbolone acetate (TBA) and estrodial benzoate (E_2), found in the form of Revalor and Synovex, respectively. These compounds have been shown to increase feedlot performance, average daily gain, feed efficiency, and daily dry matter intake, and profitability. In a study conducted by Johnson et al. (1996), steers implanted with a combined TBA + E_2 exhibited a 21 percent increase in average daily gain 115 days after implantation. Also, these animals exhibited a 13 percent decrease in required dry matter intake per kg gain for implanted steers versus non-implanted steers during the first 40 days after implantation.

Furthermore, these animals tend to exhibit increased hot carcass weights and ribeye area as well as decreased subcutaneous fat. Almost all studies reviewed showed that implanted steers exhibited increased hot carcass weights when compared to their non-implanted counterparts. Bruns et al. (2005) concluded that steers receiving early implants exhibited increased hot carcass weights and ribeye area. Roeber et al. (2000) noted similar results in regards to hot carcass weight and ribeye area while also noting significantly higher kidney, pelvic, and heart fat percentage among control steers versus steers implanted with Synovex Plus. Another study found that implanted animals slaughtered on day 115 in the feedyard exhibited significantly higher ribeye areas;

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however, steers slaughtered on day 143 in the feedyard did not (Johnson et al., 1996). This seems to suggest that over time the differences in carcass characteristics disappear. This is in agreement with Samber et al. (1996), who studied implant effects on calves fed to 212 days. This study found that significant differences did not exist in ribeye area, hot carcass weight, fat thickness, or KPH between the control group and animals administered an implant strategy of Synovex/Revalor and Revalor/Revalor. Although many of these reports suggested implanted animals tend to have larger ribeye areas and lower subcutaneous fat, these studies found that no differences occurred in calculated yield grades. This seems to be because increased hot carcass weights are offsetting larger ribeye areas (Roeber et al., 2000).

Many studies have shown steers treated with growth promoting implants, when compared to control animals, displayed decreased intramuscular fat. Bruns et al. (2005) studied the effect of stage of growth and implant on carcass composition. They found that steers implanted early (d 0 on feed) with a combined TBA + E_2 exhibited lower marbling scores and decreased percentage of carcasses grading Premium Choice and an increased percentage of Select compared to animals receiving no implant. However, they found the delayed implant (d 57 on feed) group did not display significantly lower marbling scores from the control group. A separate study utilizing 10 different implant combinations found similar results. Platter et al. (2003) reported that the control group displayed significantly higher marbling scores than all 10 implant groups. It is interesting to note, however, that while the control group exhibited the highest marbling score, the percentage of Prime and Choice did not significantly differ between the treatment groups and the non-implanted cattle. Rather, they found that the distribution shifted, with the

percentage of carcasses with marbling scores of Modest^{00} and higher being significantly reduced in certain implant strategies (Platter et al., 2003). Research conducted by Johnson et al. (1996), however, found that marbling scores were not significantly different between implanted and non-implanted steers on either the day 115 or 143 slaughter groups. While the majority of the research conducted utilized steers, two separate studies conducted by Kniffen et al. (1999) and Schneider et al. (2007) researched the effects of growth promotants on heifers. Schneider et al. (2007) found that heifers implanted once with TBA showed no difference in marbling scores compared to that of the control group. A separate study conducted by Kniffen et al. (1999) studied the effects of estrodial-releasing implant on carcass characteristics in beef heifers. No differences were found in quality grades between the treatment groups in this study.

Even though animals implanted with growth promotants have displayed increased feedlot performance and lean tissue accretion, some research has suggested that animals implanted with growth promotants tend to be tougher than their non-implanted counterparts. While most of the research is in agreement on effects of carcass characteristics due to implants, the effect on tenderness is inconsistent. Many of the studies suggest that implanted animals tend to have higher Warner-Bratzler shear (WBS) values. Platter et al. (2003) concluded that samples from all ten implant groups exhibited higher ($P < 0.05$) WBS values than the control group. Similarly, two different studies concluded that the implants containing trenbolone acetate + estrodial (Revalor) produced steaks with significantly higher WBS values than control steaks. Samber et al. (1996) noted that the control group had significantly lower WBS values than all treatments in which the first implant administered was Revalor. In this study, however,

WBS values were not significantly different between the control group and steers in which the first implant administered was not Revalor. Roeber et al. (2000) reported different results in which steers implanted with Revalor S twice or any groups administered Synovex did not produced significantly higher WBS values than control animals. However, animals in this study that were administered only one implant of Revalor S displayed higher ($P < 0.05$) shear values than control animals. This is also contrary to the findings of Schneider et al. (2007) who did not report any differences between no implant and one implant for any aging period tested. Barham et al. (2003), utilizing a population of *Bos indicus* influenced steers, found that significant differences existed in shear force values between non-implanted and implanted animals following 0-, 7-, and 14-day aging periods. However, the same study found that those differences disappeared following the 21-day aging period. While most studies measured tenderness by evaluating shear force values, there are a few that used consumer panels. Two separate projects involving growth promoting implants and tenderness utilized consumer panels to evaluate the ability of consumers to identify the differences between steaks from treatment and control steers. Of these two projects, the research conducted by Barham et al. (2003) analyzed the effects of aging on consumer perception, whereas the study performed by Platter et al. (2003) did not. Conclusions from both studies showed lower overall likeness scores for implant-treated steaks. However, Barham et al. (2003) noted that consumers could no longer differentiate treatment and control steaks following the 14-day aging period. Furthermore, Roeber et al. (2000) reported that consumer tenderness scores were less desirable for steaks from animals administered two doses of Revalor S than from the control group.

Bos indicus breeds in beef production systems have been used traditionally to increase hybrid vigor, especially in semitropical and tropical climates where these breeds provide additional advantages for heat, disease, and insect resistance (Cole et al., 1963; Crockett et al., 1975). In these regions, *Bos indicus* influenced animals also tend to have increased average daily gain and higher slaughter weights. Huffman et al. (1990) utilized a population of steers of various percentages of Brahman influence and measured feedlot performance in sub-tropical Florida. Steers of ¾ Brahman, ¼ Angus breeding had higher ($P < 0.05$) averaged daily gains than their Angus counterparts. Furthermore, steers possessing increased percentages of Brahman influence displayed increased daily dry matter intake. Cattle displaying *Bos indicus* traits, however, have been discriminated at the packer level (Johnson et al., 1990) due to perceived inconsistencies in tenderness. Several studies have reported that meat from *Bos indicus* influenced cattle displayed increased shear force values (Crouse et al., 1989; Koch et al., 1976). Furthermore, Huffman et al. (1990) found that marbling score decreased in calves exhibiting at least 50% Brahman influence.

The effects of growth promotants on carcass characteristics and tenderness are well documented in steers. However, there has been very little research done exploring the impact of gender. Moreover, there is limited research on tenderness as a result of the interaction between growth promoting implants and gender among cattle with *Bos indicus* breeding. Such data would be beneficial to the beef industry allowing anabolic implants to be more efficiently utilized in improving yields and/or final product while minimizing tenderness and other quality problems. Possibly by incorporating growth promoting implants, days on feed, and aging into a production protocol, differences

between control and implanted animals could be significantly mitigated. Therefore, the object of this study was to study the effects of a control, intermediate (Synovex), and aggressive (Revalor) implant strategy on carcass characteristics, tenderness, and postmortem proteolysis on *Bos indicus* influenced cattle.

CHAPTER II

MATERIALS AND METHODS

Animals

Bos indicus type (5/8 Angus X 3/8 Brahman or 5/8 Angus X 3/8 Nellore) steers (n = 77) and heifers (n = 68) of known parentage were utilized from the Texas AgriLife Research Station in McGregor, Texas for this study. Calves then were separated by gender and assigned randomly to one of three treatment groups. Implant regime and treatment breakdown is displayed in Table 1. These implant treatments were chosen to compare a control with an intermediate and an aggressive implant strategies which is often used in the cattle feeding industry. Treatments groups were as follows: 1 (**CON**) = no implant / no implant; 2 intermediate (**SYN**) = Synovex / Synovex; 3 aggressive (**REV**) = Revalor / Revalor. Synovex-S (200 mg progesterone, 20 mg estradiol benzoate) and Revalor-S (120 mg trenbolone acetate, 24 mg estradiol) were used for the steers whearas Synovex-H (200 mg testosterone propionate, 20 mg estradiol benzoate) and Revalor-H (140 mg trenbolone acetate, 14 mg estradiol) were used for the heifers. Implants were administered at 0 day on feed (if applicable) followed by another implant at 84 days on feed. To ensure consistency in handling, cattle from the control group were run through cattle chutes and handled in the same manner as those animals receiving implants.

Table 1. Implant regime and treatment breakdown for animals utilized in study

		Day implant was administered (d)	
	n	0	84
Steer			
Control	24	no implant	no implant
Synovex	25	200 mg progesterone, 20 mg estradiol benzoate	200 mg progesterone, 20 mg estradiol benzoate
Revalor	28	120 mg trenbolone acetate, 24 mg estradiol	120 mg trenbolone acetate, 24 mg estradiol
Heifer			
Control	22	no implant	no implant
Synovex	21	200 mg testosterone propionate, 20 mg estradiol benzoate	200 mg testosterone propionate, 20 mg estradiol benzoate
Revalor	25	140 mg trenbolone acetate, 14 mg estradiol	140 mg trenbolone acetate, 14 mg estradiol

Carcass Evaluation

Animals were fed to an estimated backfat thickness of 10 mm through visual appraisal and were harvested in two groups. Representatives of all three treatments were present in both harvest groups. Group 1 was harvested at day 115 and group 2 at day 150. Animal identification was maintained throughout the harvest process and final hot carcass weight was collected. Carcasses underwent a 48 hour chill period and then were evaluated for USDA quality grade and yield grade data (USDA, 1996). Trained personnel from Texas A&M University collected data for adjusted fat thickness, ribeye area, kidney, pelvic, and heart fat, overall maturity, marbling, and dark cutter score. Color data were obtained using a Hunter Mini-scan colorimeter (Hunter Labs, Inc., Reston, VA; Illuminant A, 10° observed); two readings were obtained per animal. Following grade data collection, approx. 16 cm inch samples of the strip loin then were collected from the right side of the carcass and shipped back to the Rosenthal Meat Science and Technology Center in College Station, TX. Strip loins were fabricated as follows: one slice off the anterior end for fat and moisture determination, three 2.54 cm steaks for Warner-Bratzler shear force determination, and one 1.27 cm steak for sarcomere length determination.

Warner-Bratzler Shear Force

Upon strip loin fabrication, the three steaks utilized for shear force testing were assigned to three aging periods, 0-d, 14-d, and 21-d, vacuum packaged and frozen upon reaching the designated aging treatment. Aging period is based on days post packaging at the commercial packing plant facility, which was approximately 48 hours after harvest. Steaks were thawed at 4°C for 24 hours before cooking. Steaks were cooked to an

internal temperature of 70°C on an electric grill (Hamilton Beach Indoor/Outdoor Grill, Hamilton Beach/Proctor Silex, Inc., Southern Pines, NC) and were turned once at 35°C. Temperature was monitored using type K thermocouples attached to a Thermocouple Input Benchtop Meter (model BS 6001A, Omega Engineering, Inc., Stamford, CT). Six 1.27 cm diameter cores were removed parallel to muscle fiber direction. Cores then were sheared using a Universal Testing Machine with a Warner-Bratzler shear attachment (model SSTM-500, United Calibration Corporation, Huntington Beach, CA). Cores and any remaining sample were frozen for proteolysis determination.

Chemical Fat and Moisture Analysis

The slice designated for fat and moisture determination was frozen in liquid nitrogen and pulverized using a Waring Blender. Approximately 3 g of sample was weighed and stuffed in a Fisher brand filter paper thimble. Two samples from each animal were obtained and allowed to dry in an oven at 100°C for 12 hours. Samples then were cooled for approx 30 min and were re-weighed. Moisture was expressed as the difference between pre- and post-drying weights. Chemical fat analysis was determined using a modified version of the ether extraction method (AOAC, 2000). Samples were placed in a soxhlet, and 2000 mL flasks underneath were filled with boiling chips and 1000 mL of ether. Extraction was carried out over an 18-hour period. Samples then were placed underneath a chemical hood to allow the ether to evaporate. Thimbles were oven dried again for 12 hours at 100°C and then reweighed to determine extractable fat percentage.

Sarcomere Length

Sarcomere length was determined on two samples representing the lateral and medial sections of each steak. Samples (4 g) were placed in a Vitris flask containing 20 mL of cold buffer solution (.25 M sucrose, .002 M KCl, pH 7.0) and homogenized at low speed for 15 sec. A drop of homogenate then was placed on a glass microscope slide and covered with a cover slip. Sarcomere length was measured using a Spectra-Physics model 155SL helium-neon laser (0.95 mW). The slide was placed under the laser and moved from one edge to another until a diffraction pattern was observed. Ten independent measurements were taken from each sample and calculated using the formula outlined by Cross et al. (1981).

Proteolysis

Protein degradation was determined using the procedure outlined by Wheeler and Koohmaraie (1999). Cooked samples from shear force determination were powdered, then shipped to the U.S. Meat Animal Research Center for western blot procedure.

Powdered samples were homogenized with Tris-EDTA extraction buffer for 20 seconds. A .5 ml aliquot was taken and 2X Treatment Buffer added, then heated in a water bath, mixed repeatedly then reheated for an additional 5 minutes. Samples were then centrifuged (Eppendorf 5414 C, Eppendorf AG, Hamburg, Germany) for 20 minutes. Protein concentrations were determined using the micro-BCA protein assay (Pierce, Rockford, IL) using a 1:5 dilution of supernatant and 1X treatment buffer. BCA reagent was added to each sample, allowed to incubate for 30 minutes, and read at 562 nm (SPECTRAmax Plus 384, Molecular Devices Corp., Sunnyvale, CA). Samples were

then diluted to 3 mg/ml using protein denaturing buffer containing 2X treatment buffer, 10% MCE, and .8% bromophenol blue.

Electrophoresis was performed using 10% gels (1.5 M tris, pH 8.8; 30% acrylamide, 10% SDS, 10% ammonium persulfate (APS), and TEMED) with a 4% stacker (.5 M tris, pH 6.8). Zero hour standards pooled from the longissimus muscle of multiple animals were loaded onto the gel at 18 micrograms in triplicate. Diluted samples were loaded at 15 micrograms, 9 samples per gel. Gels then were ran at 200 volts for 45 min. using a Bio-Rad Power supply (Bio-Rad, Hercules, CA). Subsequently, gels then were transferred onto a PVDF (polyvinylidene difluoride) membrane at 200 mA for one hour using towbin transfer buffer plus 10% methanol. Membranes were blocked using StartingBlock T20 (TBS) blocking buffer (Thermo Scientific, Rockford, IL) for 30 minutes. Purified D3 (batch UNMC1) primary antibody was added and allowed to incubate for 1 hour. Membranes were washed with TTBS (20 mM Tris, 137 mM NaCl, 5 mM KCl, .05% Tween 20) once at 15 min. and twice at 5 min. to remove excess antibody. Pierce anti-mouse IgG (Thermo Scientific, Rockford, IL) was used as the secondary antibody at a dilution of 1:10,000 and allowed to incubate for 1 hour. Membranes then were washed with TTBS once for 15 min., then 4 more times for 5 min. Antibody detection was assessed by exposing the membrane to SuperSignal West Dura Extended Duration Substrate (Pierce, Rockford, IL). Imaging and analysis was completed using the ChemiImager 4000 digital imaging system (Alpha Innotech, San Leandro, CA). Membranes were exposed for five min. and then quantified by measuring band density (IDV). The three 0 hour standards were averaged and used as a reference. Desmin degradation was expressed as a percentage of the reference for each blot, or:

% Degradation = (1.0- (IDV of Sample/ μ IDV of 0 hr Standard)) x 100.

Statistics

Mixed-model procedures of SAS (v. 9.1.3; SAS Institute, Cary, NC) were used for analyses. Analysis of variance was used to separate responses. Treatment, gender and aging time were included in the model as fixed effects; harvest group was included as a random effect. When main effects were significant ($P < 0.05$), least squares means were reported and separated using the p diff procedure of SAS. Pearson correlations were calculated using the PROC CORR function of SAS.

CHAPTER III

RESULTS AND DISCUSSION

Carcass Characteristics

Table 2 represents least squares means and standard errors for hot carcass weights (HCW) and longissimus muscle area (LMA) stratified by gender x treatment group. A gender, treatment effects and treatment by gender interaction was noted ($P < 0.05$) for hot carcass weight. Steers exhibited heavier ($P < 0.0001$) hot carcass weights than heifers and the control group exhibited lighter ($P < 0.05$) weights than both Synovex (SYN) and Revalor (REV). Animals implanted with Synovex and Revalor displayed a HCW increase of 6 and 8 percent, respectively. However, there was no difference between the two implant groups. This is in agreement with the results found by Bruns et al. (2005) and Roeber et al. (2000) who found that while implant groups demonstrated heavier HCW, few differences resulted between implant groups. Within the heifers, no significant difference was observed between the control group and the implant groups for HCW. This differs from the results found by Schneider et al. (2007), who reported that heifers from the implant groups exhibited heavier hot carcass weights than those from the control group. Least square means for other USDA yield grade related carcass traits stratified by gender and treatment groups are reported in Table 3. No differences were observed for KPH percentage. This is contrary to findings by Johnson et al. (1996), who found that implanted animals slaughtered on day 143 produced lower percentages of KPH than non-implanted animals from the same slaughter group. A small, yet significant

($P < 0.05$) difference was found in adjusted fat thickness for gender. Steers exhibited an adjusted fat thickness of .92 cm, whereas heifers presented an adjusted fat thickness of 1.04 cm. There were not differences noted between treatments. This was expected as animals were fed to a constant fat thickness of 10 mm through visual appraisal. Gender and treatment significantly impacted LMA, however, no interaction was observed. Steers exhibited larger longissimus muscle areas compared to heifers. Furthermore, animals from both implant groups exhibited larger longissimus muscle areas than the control. Many studies have reported similar findings, with implant groups displaying larger LMA than non-implanted animals (Platter et al., 2003; Roeber et al., 2000).

A treatment effect for yield grade was observed ($P < 0.05$); Revalor-implanted animals had lower yield grades than Synovex-implanted and control animals. No differences were observed between the Synovex-implanted and control groups (Table 3). This differs from the findings of Roeber et al. (2000), who reported no differences in yield grades between implanted and non-implanted animals.

Table 2. Least squares means ± standard error (SEM) for hot carcass weight and longissimus muscle area for treatment x gender group

	n	HCW, kg	SEM	LMA, cm^2	SEM
Steer					
Control	24	294.86[c]	8.6	77.42[cd]	2.07
Synovex	25	333.51[a]	8.6	81.45[b]	2.06
Revalor	28	320.59[b]	8.4	82.60[ab]	2.01
Heifer					
Control	22	273.25[d]	8.8	74.42[d]	2.10
Synovex	21	283.48[cd]	8.9	81.44[ab]	2.13
Revalor	25	287.13[cd]	8.5	80.29[abc]	2.05
			P value		
Gender effect		< 0.0001		0.014	
Treatment effect		< 0.0001		< 0.0001	
Gender x Treatment effect		0.0405		0.971	

[a-d]Means within a column lacking a common superscript letter differ ($P < 0.05$)

Table 3. Least squares means ± standard error (SEM) for hot carcass weight (kg), adjusted fat thickness (AFT) cm, longissimus muscle area (LMA) cm^2, kidney, pelvic, and heart fat percentage (KPH), and USDA yield grade (YG)

	n	AFT, cm	LMA, cm^2	KPH, %	YG
Gender					
Steer	77	.924 ± .04[b]	80.35 ± 1.80[a]	1.54 ± .09	2.52 ± .067
Heifer	68	1.04 ± .04[a]	77.46 ± 1.78[b]	1.64 ± .09	2.51 ± .072
Treatment					
Control	46	1.03 ± .05	75.71 ± 1.87[b]	1.69 ± .10[a]	2.62 ± .087[a]
Synovex	46	1.01 ± .05	79.98 ± 1.88[ab]	1.58 ± .10[ab]	2.58 ± .088[ab]
Revalor	53	.91 ± .05	81.44 ± 1.84[a]	1.49 ± .10[b]	2.33 ± .081[b]
			P value		
Gender		0.043	0.014	0.166	0.913
Treatment		0.175	< 0.0001	0.076	0.031
Gender x Treatment		0.173	0.971	0.163	0.107

[ab]Means within a column and main effect lacking a common superscript letter differ ($P < 0.05$)

Least squares means and standard error for marbling scores are presented in Table 4. Gender ($P < 0.05$) and treatment ($P < 0.005$) impacted marbling scores, however, no gender x treatment interaction was observed. Heifers displayed a higher marbling score than steers. Also, non-implanted animals displayed higher marbling scores than those from the REV group. Those from the Synovex group were numerically intermediary to the CON and REV groups and did not differ from either group. Many studies (Bruns et al., 2005; Platter et al., 2003) have documented similar results, where Revalor implanted animals displayed lower marbling scores than their non-implanted counterparts. Percentages for USDA quality grades between gender and implant group are displayed in Figures 1 and 2, respectively. All groups exhibited lean and skeletal maturity scores within the "A" score. Treatment group appeared to shift the distribution of quality grades between steers and heifers. Within the steers, a noticeable shift in quality grade was observed. The control group presented 44 percent of all carcasses grading low Choice or higher, whereas the REV group had a low Choice or higher percentage of 35. Within the heifers, a noticeable increase in the percentage of carcasses grading Select occurred, with the SYN group displaying an increase of 10%, and REV 20% as compared to the control group. This corroborates with the results of Schneider et al. (2007), who found that the percentage of re-implanted carcasses grading Choice or higher significantly decreased ($P < 0.05$). Furthermore, Samber et al. (1996) showed a significant decrease in Choice grading carcasses among those administered three implants of Revalor.

Table 4. Least squares means ± standard error of means (SEM) for Marbling Score

	Marbling Score[2]
Gender	
Steer	351.80 ± 8.42[b]
Heifer	381.98 ± 8.97[a]
Treatment	
Control	383.± 10.88[a]
Synovex	373.24 ± 10.91[ab]
Revalor	344.29 ± 10.15[b]
	P-value
Gender	0.0154
Treatment	0.0259
Gender x Treatment	0.9476

[1]Marbling Score – 100-199 – devoid, 200-299 – traces, 300-399 – slight, 400-499 – small, 500-599 – modest

[ab]Means within a column and main effect lacking a common superscript letter differ ($P < 0.05$)

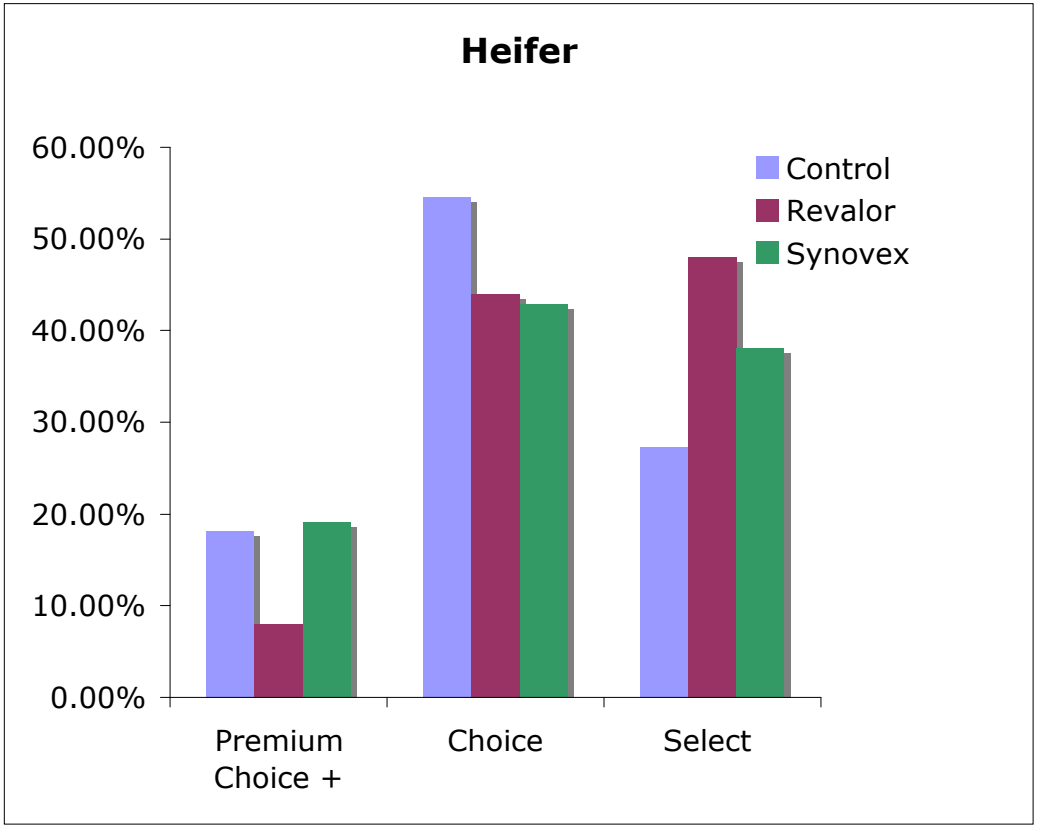


Figure 1. Percentages of USDA quality grades for heifers by treatment

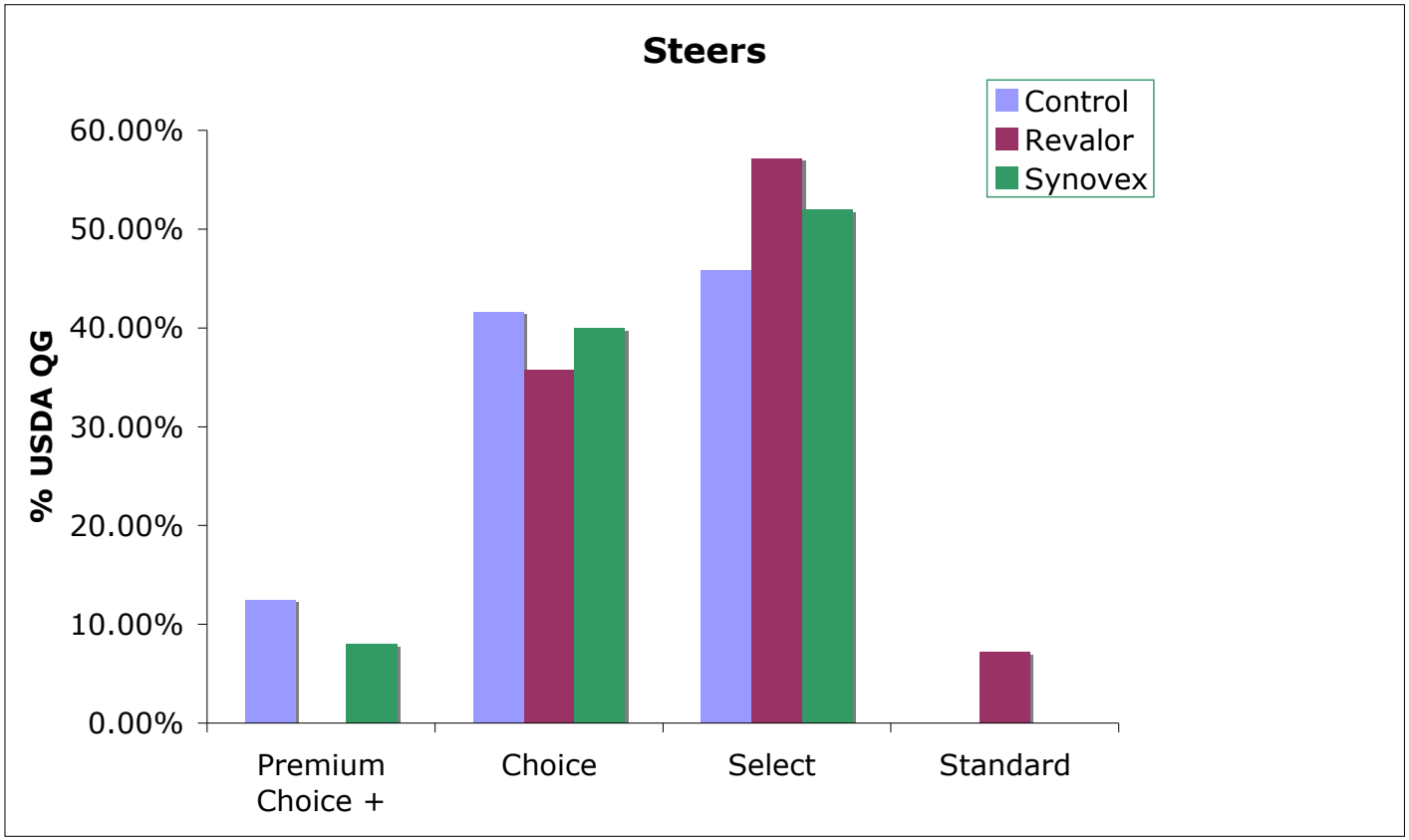


Figure 2. Percentages of USDA quality grades for steers by treatment

Color scores are reported in Table 5. No differences between gender and treatment were observed for lightness (L*). However, there was a gender effect for redness (a*) values with steers exhibiting higher (more red) values than heifers. Also, there was a treatment interaction for yellowness (b*) values with the REV group displaying significantly lower (less yellow) values than either the CON or SYN group.

Table 5. Least squares means ± SEM for color scores

	L*[1]	a*[2]	b*[3]
Gender			
Steer	40.37 ± .34	41.23 ± 1.07[a]	28.85 ± 2.57
Heifer	41.31 ± .36	40.17 ± 1.08[b]	28.64 ± 2.57
Treatment			
Control	41.18 ± .43	40.94 ± 1.10	29.29 ± 2.58[a]
Synovex	40.85 ± .44	40.82 ± 1.10	29.11 ± 2.59[a]
Revalor	40.49 ± .40	40.33 ± 1.09	27.82 ± 2.58[b]
		P value	
Gender	0.0566	0.0224	0.6718
Treatment	0.5156	0.5019	0.0240
Gender x Treatment	0.3555	0.7220	0.9004

1- L*-lightness 0-100,
2- a*- positive – red, negative – green
3- b*- positive – yellow, negative – blue
[ab]Means lacking a common superscript letter differ ($P < 0.05$)

Warner-Bratzler Shear Force

Least squares means and standard errors for Warner-Bratzler shear force by treatment group (Table 6) and gender (Table 7) were reported. Treatment and Day significantly impacted Warner-Bratzler shear force values yet no treatment x day interaction was observed. The CON group had the lowest shear values on day 0, with no significant difference noted between REV and SYN. A significant drop in WBS value was observed for all treatments on day 14, with the CON group exhibiting a 28 % decrease followed by SYN with a 24% reduction, and finally REV at 22% between day 0 and 14. Therefore, because the rate of reduction in WBS value was similar for all treatments, meat from implanted animals benefited from postmortem enzymatic tenderization just as greatly as non-implanted animals. CON steaks displayed lower ($P < 0.001$) shear values than either of the implant groups following the 14-day aging period. Many studies (Schneider et al., 2007; Samber et al., 1996) have produced similar results in which implanted animals displayed higher shear force values after a 14-day aging period. Furthermore, standard errors were reduced following 14-day aging period, suggesting less variation between animals. However, following the 21-day aging period, all differences in WBS values had disappeared between the implant and control groups. This differs from the findings of Schneider et al. (2007), who found that differences still existed among treatment groups for steaks aged 21 days.

Table 6. Least squares means ± SEM for Warner-Bratzler shear (WBS) force (kg) for implant group by aging period (d)

	Aging period		
Treatment	0 d	14 d	21 d
Control	3.00 ± .187[b]	2.40 ± .129[b]	2.43 ± .121[a]
Synovex	3.54 ± .188[a]	2.83 ± .130[a]	2.78 ± .121[a]
Revalor	3.72 ± .176[a]	3.02 ± .122[a]	2.73 ± .112[a]
		P value	
Treatment	0.011	0.0006	0.092
Day	< 0.0001	-	-
Treatment x Day	.7477	-	-
Gender x Treatment	0.681	0.5936	0.942

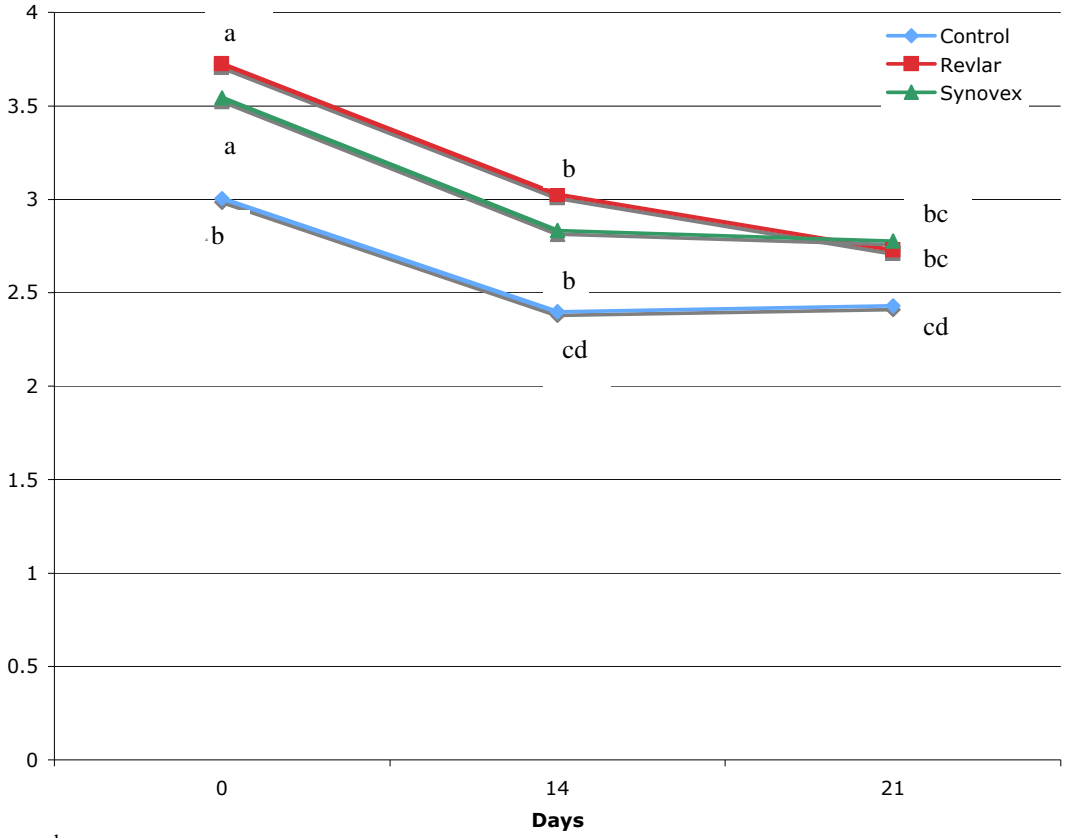
[ab]Means lacking a common superscript letter differ ($P < 0.05$)

Table 7. Least squares means ± SEM for WBS for gender by aging period (d)

	Aging Period		
Gender	0 d	14 d	21 d
Steer	3.37 ± .149[a]	2.75 ± .106[b]	2.62 ± .094[b]
Heifer	3.48 ± .156[a]	2.76 ± .111[b]	2.67 ± .099[b]
		P value	
Gender	0.603	0.922	0.683

[ab]Means lacking a common superscript letter differ ($P < 0.05$)

Figure 3 depicts mean shear force values for all three treatment groups throughout the aging periods. The greatest drop in shear force values for all three groups came at 14 days, at which point the control group displayed the lowest shear values. However, no differences were observed between the two implant groups. It is apparent that the differences in shear force are the result of the higher initial shear force exhibited by the implant groups. This is supported by the findings of Schneider et al. (2007), who found that heifers receiving two implants displayed higher day 0 shear force values than those receiving either one or no implant. Furthermore, Roeber et al. (2000) reported higher initial shear force values for groups implanted with Revalor S. However, many studies have reported no differences existed in shear force between implant and non-implanted animals. Most notably is the Barham et al. (2003) study which utilized *Bos indicus* influenced animals. Results from this study indicated steaks from implanted animals received lower tenderness scores by sensory panels after a 14-day aging period. However, steaks from the same aging period revealed no differences in shear force. Furthermore, differences in tenderness detected by sensory panels diminished following the 21-day aging period.



[a-d]Values lacking a common superscript letter differ ($P < 0.05$)

Figure 3. Least squares means for WBS force value (kg)

Chemical Fat and Moisture Analysis

In regards to extractable fat analysis (Table 8), a gender effect ($P < 0.05$) was noted. Heifers displayed higher extractable fat percentage than steers, and Revalor displayed lower extractable fat percentage than either control or Synovex. The differences between genders may be attributed to the higher marbling scores exhibited by heifers. The sample for fat determination was taken off of the anterior end of the loin sample, opposite the 12th rib interface where marbling scores were measured. Although no treatment effects were observed, REV animals displayed a lower percentage of extractable fat, again corresponding with the marbling scores noted earlier. This is supported by the findings of Savell et al. (1986), who reported differences in ether extractable fat between marbling scores although the percentages reported here are lower than those presented in the study.

Sarcomere Length

Least squares means for sarcomere length are displayed in Table 9. A treatment effect was observed, however, there were no differences noted between genders. Sarcomere lengths from SYN group displayed the longest sarcomere lengths and differed significantly from REV group, but not from the control group. It is unclear the mechanism behind these differences.

Table 8. Least squares means and SEM for extractable fat and moisture percentage

	% Fat		% Moisture	
	Mean	SEM	Mean	SEM
Gender				
Steer	2.66[a]	.1516	69.84	2.53
Heifer	3.15[b]	.1614	69.63	2.55
Treatment				
Control	3.15[a]	.1959	68.74	2.60
Synovex	2.99[ab]	.1965	69.58	2.60
Revalor	2.57[b]	.1826	70.88	2.58
		P value		
Gender	0.0277		0.8427	
Treatment	0.0887		0.2526	
Gender x Treatment	0.9576		0.8420	

[ab]Means within a main effect lacking a common superscript letter differ ($P < 0.05$)

Table 9. Least squares means and SEM for sarcomere length (μm)

	Sarcomere Length, μm	
	Mean	SEM
Gender		
Steer	2.10	.220
Heifer	2.06	.220
Treatment		
Control	2.08[ab]	.221
Synovex	2.14[a]	.221
Revalor	2.02[b]	.221
	P value	
Gender	0.2696	
Treatment	0.0231	
Gender x Treatment	0.4261	

[ab]Means lacking a common superscript letter differ ($P < 0.05$)

Proteolysis

Desmin degradation was determined using the cooked cores from the Warner-Bratzler shear 14 d aged sample. Treatment group impacted desmin degradation percentage; however, gender did not. Least squares means and standard error are presented in Table 10. The CON group displayed the greatest amount of desmin degradation with 62%, followed by SYN displaying 55%, and REV exhibited the least amount of degradation with 38%. Heifers implanted with Revalor presented the least amount of degradation at 29%, significantly lower than all other groups except their steer counterparts. Control steers displayed the greatest amount of degradation at 64% which was different ($P < .05$) from all other implant gender combinations.

Koohmaraie (1994) reported that degradation of desmin results in the fragmentation of myofibrils most likely through the disruption of the transverse cross-linking between myofibrils thereby positively impacting tenderness. Therefore, increased degradation results in lower shear force values. Taylor et al. (1995) studied the relationship between myofibrillar degradation and tenderness in cattle. Results from this study concluded that the degradation of the Z-disk might not be as influential in tenderness as previously thought. Rather, the ability of calpains to more easily break down intermediate filaments such as desmin may play a bigger role in postmortem tenderization.

The primary advantage to using the cooked cores of the WBS samples to determine desmin degradation is to more directly study the relationship between degradation and shear force. Wheeler et al. (2002) first reported such correlations (r = -

.69) between 14-day shear force value and percent desmin degradation. They also reported a high correlation (r = .80) for desmin degradation and sensory panel tenderness scores. In this study, a significant correlation was noted between percent degradation and WBS values for the treatment groups. SYN displayed a moderate correlation (r = -.56) as reported in Table 11, suggesting that lower protein degradation for SYN may be a driving factor for the increased shear force values displayed by Synovex implanted animals. Revalor displayed a low, yet still significant ($P = 0.05$) correlation (r = -.27). The control group showed the lowest correlation between percentage degradation and WBS value. This may be attributed to the lower initial shear force values exhibited by these animals. Figure 4 represents one of the western blots prepared to determine proteolysis. Each band present represents one animal, from a gender x treatment group. It is evident in this figure that the CON group exhibited the greatest amount of degradation with the first CON sample displaying 64% degradation. Furthermore, the REV samples showed the least amount of degradation, the lowest reading among the REV samples exhibited 0% degradation. Higher shear force values displayed by the Revalor-implanted animals may then be attributed to something other than the unaltered state of desmin even after 14 days postmortem.

Table 10. Least squares means and SEM for percent desmin degradation

	% Degradation	SEM
Gender		
Steer	46.38	4.52
Heifer	49.37	4.46
Treatment		
Control	62.70[a]	4.79
Synovex	48.04[b]	4.80
Revalor	32.87[c]	4.69
	P-value	
Gender	0.3467	
Treatment	< 0.0001	
Gender x Treatment	0.6206	

[a-c]Means within main effects lacking a common superscript letter differ ($P < 0.05$).

Table 11. Pearson correlation for 14-d shear force and muscle traits

	Treatment group		
Trait	Control	Synovex	Revalor
Marbling Score	-.19ns	-.30**	-.23ns
% Degradation	-.25*	-.57***	-.27**
Fat	-.11ns	-.27*	-.17ns
a* value	-.07ns	-.007ns	-.25*
Sarcomere Length	-.11ns	.17ns	-.08ns

*– $P < 0.01$, ** – $P \leq 0.05$, ***– $P < 0.001$
ns – not significant

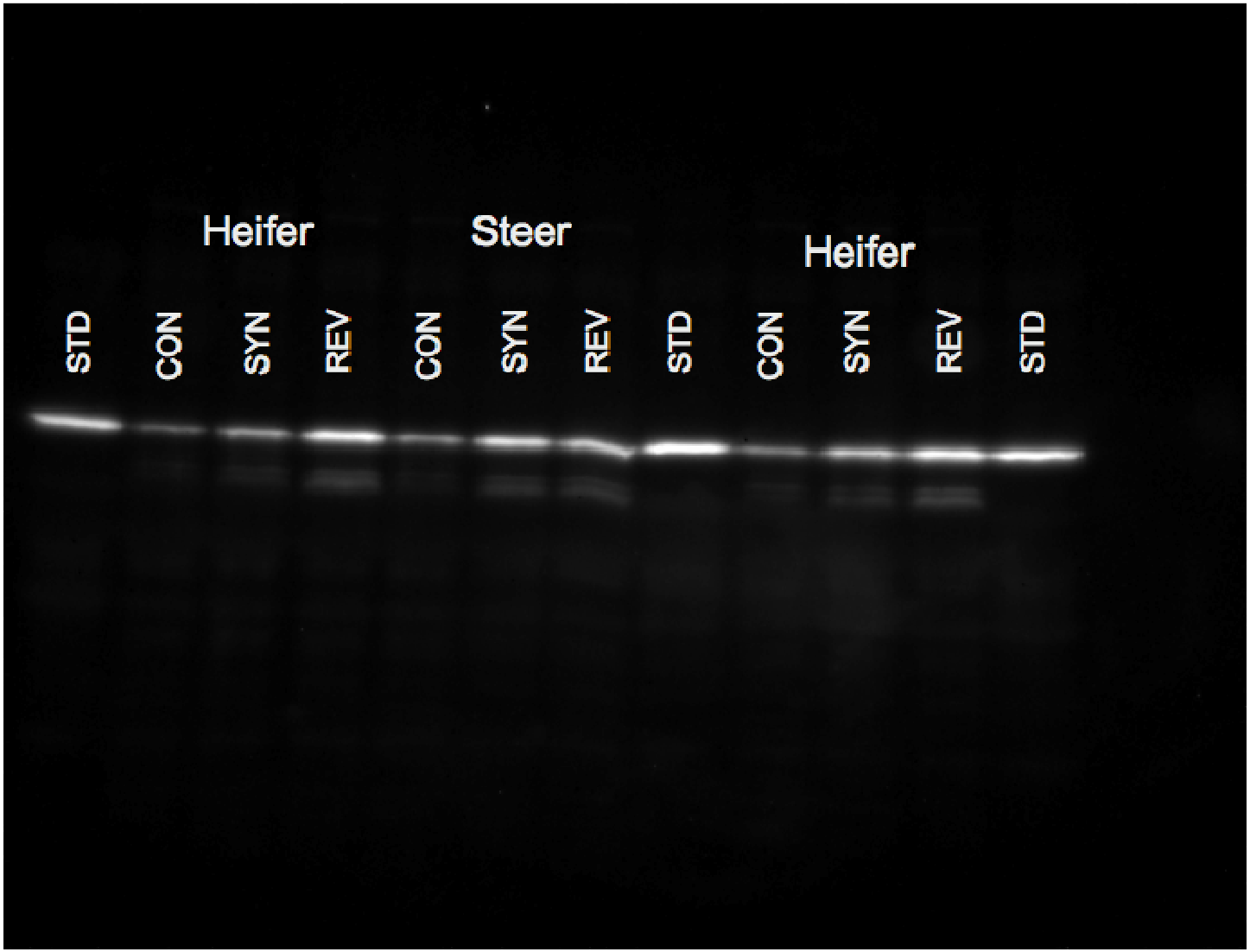


Figure 4. Example of western blot, showing desmin degradation

CHAPTER IV

SUMMARY AND CONCLUSIONS

The feedyard performance and carcass leanness benefits of anabolic growth implants have been well documented. In this study, animals from implant groups displayed heavier carcass weights and increased longissimus muscle areas as well as lower yield grades. However, these compounds had a negative impact on quality as shown by lower marbling scores and a decreased percentage of Choice grading carcasses as well as a decrease in tenderness as determined by WBS.

The 2000 National Beef Quality Audit listed reduced quality grade and tenderness due to implants as one of the top five issues facing the industry (Roeber et al., 2002). Furthermore, it has been reported that tenderness is a driving factor in the value of beef (Savell and Shackelford, 1992). This study found that animals from the implant groups displayed higher shear force values following the 0- and 14-day aging periods. However, following the 21-day aging period, all differences between the control group and implant groups had been mitigated. The increased toughness from implanted animals may result from lower percentages of desmin degradation, where all three treatment groups expressed significant differences. Furthermore, it was discovered that correlations did exist between shear force value and percent degradation for both implant groups. This implies that the lack of desmin degradation may be a driving factor in the decreased tenderness of implanted animals.

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