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Elaine Grings, Beef Report Editor

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SOUTH DAKOTA STATE UNIVERSITY BEEF EXTENSION TEAM

2012 South Dakota Beef Report

BEEF 2012-01

Comparison of three CIDR based fixed-time AI protocols for beef heifers[1](#page-9-0)

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SUMMARY

Several effective fixed-time AI (FTAI) protocols have been developed to facilitate AI while eliminating the need for estrus detection. Among these are the 5-d CO-Synch+CIDR (5d), PG 6-d CIDR (PG-CIDR), and 14 d CIDR-PG (CIDR-PG) protocols. While each of these protocols varies in duration and approach to synchronizing estrus and ovulation, each has been reported as an effective method to facilitate FTAI in beef heifers. Therefore, the objective of this study was to compare FTAI pregnancy rates in beef heifers synchronized with these three CIDR based protocols. Virgin beef heifers (n = 801) at four locations were synchronized with one of three protocols: 1) (**5-day CO-Synch + CIDR**) an injection of GnRH (100 μg; i.m.) and insertion of a CIDR on d -5, PG (25 mg; i.m.) and CIDR removal on d 0 with a second injection of PG (>4 h after CIDR removal) on d 0 and FTAI at 72 h after CIDR removal, 2) (**PG 6-day CIDR**) PG (25 mg; i.m.) on d -9, GnRH (100 μg; i.m.) and insertion of a CIDR on d -6, PG and CIDR removal on d 0, and FTAI at 66 h after CIDR removal, or 3) (**14-day CIDR-PG**) a 14-day CIDR insert from d -30 to -16, PG (25 mg; i.m.) on d 0, and FTAI at 66 h after PG. All heifers received an injection of GnRH (100 μg; i.m.) concurrent with FTAI. Timing of treatment initiation was offset to allow all heifers to receive FTAI concomitantly and at random. Pregnancy success was determined between 35 and 40 d after FTAI by transrectal ultrasonography. Blood samples were collected approximately 12 d before the beginning of each protocol and at the initiation of each protocol to determine estrous cycling status (77%). Data were analyzed using the GLIMMIX procedures of SAS. Fixed-time AI pregnancy success did not differ between treatments (*P* = 0.13; 62.5%, 56.9%, and 53.3%, for 5-day CO-Synch + CIDR, PG 6-day CIDR, and 14-day CIDR-PG; respectively) or location (*P* = 0.16; 51.5%, 62.7%, 56.1%, and 58.6% for location 1, 2, 3, and 4; respectively). However, heifers that had reached puberty by initiation of synchronization had greater (*P* < 0.01) pregnancy success compared to heifers that were prepubertal (60.7% and 47.3%; respectively). In summary, all three protocols had similar FTAI pregnancy success, and puberty status had the greatest impact on pregnancy success.

INTRODUCTION

Numerous estrous synchronization protocols are available for facilitating artificial insemination (AI) in cattle. While some of these protocols rely on estrous detection, recently several protocols have been developed to facilitate the mass breeding of all females at a predetermined time. These "fixed-time AI" (FTAI) programs forgo estrous detection but rather synchronize ovulation. Such an approach eliminates the time and labor involved in estrous detection and allows females to be worked as a herd rather than individually. Recently, several FTAI approaches have been developed for beef heifers. Among these

 1 This project was funded by Select Sires and Pfizer Animal Health

include the 5 day CO-Synch + CIDR, PG 6-day CIDR, and the 14-day CIDR-PG protocols. While each of these protocols vary in their duration and approach to synchronizing estrus and ovulation, each have been proven effective methods to facilitate FTAI in beef heifers. Therefore, the objective of this experiment was to compare FTAI pregnancy rates between the 5 day CO-Synch + CIDR, PG 6-day CIDR, and the 14 day CIDR-PG protocols.

METHODS AND MATERIALS

Eight hundred and one virgin beef heifers (approximately 15 months of age) located at 4 locations (WY n= 116, SD n= 157, MN n=233, and UT n=295) were randomly allotted to 1 of 3 FTAI protocols (Figure 1): 1) (**5-day CO-Synch + CIDR**) an injection of GnRH (100 µg; i.m.) and insertion of a CIDR on d -5, PG (25 mg; i.m.) and CIDR removal on d 0 with a second injection of PG > 4 h after the 1st on d 0, and FTAI at 72 h after CIDR removal concurrent with GnRH administration, or 2) (**PG 6-day CIDR**) PG (25 mg; i.m.) on d - 9, GnRH (100 µg; i.m.) and insertion of a CIDR on d -6, PG (25 mg; i.m.) and CIDR removal on d 0, and FTAI at 66 h after CIDR removal concurrent with GnRH administration; 3) (**14-day CIDR-PG**) a 14-day CIDR insert from d -30 to -16, PG on d 0, and FTAI at 66 h after PG concurrent with GnRH administration. Treatment initiation was offset to allow all heifers to be FTAI at the same time and at random across treatments.

Figure 1. Heifers were allotted to 1 of 3 protocols: 1) (**5-day CO-Synch + CIDR**) an injection of GnRH (100 µg; i.m.) and insertion of a CIDR on d -5, PG (25 mg; i.m.) and CIDR removal on d 0 with a second injection of PG >4 h after the $1st$ on d 0, and FTAI at 72 h after CIDR removal concurrent with GnRH administration, or 2) (**PG 6-day CIDR**) PG (25 mg; i.m.) on d -9, GnRH (100 µg; i.m.) and insertion of a CIDR on d -6, PG (25 mg; i.m.) and CIDR removal on d 0, and FTAI at 66 h after CIDR removal concurrent

with GnRH administration; 3) (**14-day CIDR-PG**) a 14-day CIDR insert from d -30 to -16, PG on d 0, and FTAI at 66 h after PG concurrent with GnRH administration.

At the final PG administration all heifers at locations 2 and 3 (SD and MN) were marked with tail paint. At FTAI, tail paint scores were assessed $(1 = tail$ paint completely gone; $2 = tail$ paint partially gone, obvious signs of mounting; 3 = no signs of mounting, tail paint undisturbed). Pregnancy success was determined in all heifers 35 to 40 d after insemination using transrectal ultrasonography. Blood samples were collected 10 to 12 d before the beginning of each synchronization protocol and on the day the synchronization protocol was initiated to determine cycling status. Heifers with > 1.0 ng/mL of progesterone in at least one of the two blood samples were considered pubertal. Heifers that failed to have progesterone concentrations > 1.0 ng/mL in either blood sample were considered prepubertal at protocol initiation.

Sire and technician were unique to each herd. Therefore, the effect of sire and technician within each herd was analyzed separately using the GLIMMIX procedure of SAS. There was no treatment x sire or treatment x technician interactions so data was combined. Fixed-time AI pregnancy rates were analyzed using the GLIMMIX procedure of SAS with herd included as a random effect. None of the main effect interactions were significant; therefore, all interactions were removed from the model. Main effects were removed in a stepwise reducing method, and the final model included treatment and puberty status. In herds 2 and 3 where tail paint scores were used, impact of tail paint score on fixedtime AI pregnancy success was determined using the GLIMMIX procedure of SAS.

RESULTS AND DISCUSSION

All three of the implemented protocols have been previously determined to be effective and deliver satisfactory FTAI pregnancy rates (42 to 64%) when conducted in independent research trials (Mallory et al., 2010; Perry et al., 2011). However, these protocols have never been tested against each other in a controlled research trial. In the present study, FTAI pregnancy success did not differ between treatments ($P = 0.13$; Table 1) and ranged from 53% to 63%.

pregnancy success.			
Treatment	5-day CO-Synch +	PG 6-day	
	CIDR	CIDR	14-day CIDR-PG
	62.5%	56.9%	53.3%
Pubertal status	Pubertal	Prepubertal	
	$60.7%$ ^a	$47.3%^{b}$	
Tail paint score ^c		2	3
	64% ^a	58% ^a	43% ^b

Table 1. Impact of treatment, puberty status, and estrus activity on fixed-time AI

 a^{ab} Means within a row having different superscripts are different (P < 0.05) ϵ Tail paint scores 1 = tail paint completely gone; 2 = tail paint partially gone, obvious signs of mounting; 3 = no signs of mounting, tail paint undisturbed. Previous studies have reported differences in response to synchronization between pubertal and prepubertal heifers (Wood-Follis et al. 2004, Leitman et al. 2008). In the present study there was no difference in the percent of heifers that had reached puberty prior to the initiation of the synchronization protocol (73.3%, 79.2%, and 77.1% for the 14-day CIDR-PG, 5-day CO-Synch + CIDR, and the PG 6-day CIDR, respectively). However, there was an effect of puberty status on fixed-time AI pregnancy success ($P = 0.004$). Heifers that had reached puberty prior to the start of synchronization had greater fixed-time AI pregnancies compared to heifers that were prepubertal at protocol initiation (60.7% vs. 47.3%, respectively). However, there was no treatment by puberty status interaction (P = 0.87). Among all three treatments fixed-time AI pregnancy rates were greater among heifers that had reached puberty prior to the start of the synchronization protocol compared to heifers that were prepubertal at the start of the synchronization protocol (Figure 2).

Figure 2. Effect of pubertal status within each treatment on FTAI pregnancy success.

At locations 2 and 3 where estrus activity at fixed-time AI was determined by tail paint scores there was an effect of tail paint score on fixed-time AI pregnancy success (P < 0.01). Heifers that had a tail paint score of 1 (all tail paint removed; 64%) or 2 (tail paint partially removed; 58%) had greater fixed-time AI pregnancy rates compare to heifers that had a tail paint score of 3 (no tail paint removed; 43%). In addition, there tended to be a tail paint score by puberty status interaction ($P = 0.057$). Among heifers that had reached puberty prior to the initiation of the synchronization protocol there was no effect of tail paint score on fixed-time AI pregnancy success (64%, 58%, and 51% for score 1, 2, and 3, respectively), but among heifers that were prepubertal, heifers with a tail paint score of 1 or 2 had greater (P < 0.05) pregnancy success compared to heifers with a tail paint score of 3 (63%, 57%, and 26% for score 1, 2, and 3, respectively).

CONCLUSION

All three protocols delivered acceptable (> 50%) fixed-time AI pregnancy rates in beef heifers, thus allowing beef producers the option of using a long or short protocol when breeding heifers by fixed-time AI. Pubertal status had the greatest impact on fixed-time AI pregnancy success, with heifers that had reached puberty prior to synchronization having greater fixed-time AI pregnancy rates compared to heifers that were prepubertal. In addition estrus activity as determined by tail paint score had a significant impact on fixed-time AI pregnancy success. In summary, this research demonstrates that beef producers have options when synchronizing estrus in beef heifers and it is critical that heifer

development strategies are in place to ensure that heifers are pubertal at the initiation of synchronization to maximize pregnancy success with FTAI.

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BEEF 2012-02

Influence of propionate salt levels on young cow reproductive performance[1](#page-14-0)

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SUMMARY

A supplementation study was conducted to evaluate level of propionate salt (Ca-propionate) on young cow performance over two years. One hundred-twenty cows were allocated to one of three treatments at calving. Propionate salt was incorporated in a protein supplement at a rate of 0, 80 or 160 g/d. Cows were individually supplemented twice weekly at 2 lbs/d. In year 1, cows had access to pasture and hay. In year 2, cows had access to a native range pasture. Blood was collected weekly and analyzed for progesterone (P₄) to determine postpartum interval (≥ 1 ng P₄/ml). Weights and body condition scores (BCS) were assigned at calving, end of supplementation, start of breeding season, and weaning. Cow weight and BCS changed over time through the study ($P < 0.01$ but was not affected ($P > 0.10$) by treatment. Calf weight was not different (P > 0.10) between treatments. Calf weight increased through the study (P < 0.01). Pregnancy rates did not differ between treatments (P > 0.10 but were affected by cow age (P<0.01, 77% and 100% for 2- and 3-year-olds, respectively). In year 1, the percentage of cows initiating estrous cycles before the breeding season was greater (P<0.05) for cows receiving 160 g (47.6%) compared to 0 g (15.6%) of propionate salt and tended to be greater than cows receiving 80 g (P<0.10, 20.0%). Based on ultrasonography, 3-year-old cows conceived earlier (P<0.01, 184 d) than 2 year cows (207 d). In year 2, postpartum anestrous interval ($P = 0.70$), percentage of cows initiating estrous cycles before the breeding season (P = 0.54), conception rate to AI (P = 0.68), and season-long pregnancy rates (P = 0.87) were not different among treatments. In summary, propionate salt can influence reproductive performance, however, response is not consistent.

INTRODUCTION

Producers often indicate that young cows are difficult to get rebred without additional harvested feeds. Lalman et al. (1997) indicated that positive energy balance postpartum is essential for prompt rebreeding of heifers calving in thin condition. Funston (2008) stated the inadequate protein can suppress estrus and ovulation in beef cattle. The first limiting nutrient in cows grazing dormant winter range is protein (Wallace, 1987; Lardy et al., 1998; Hollingsworth-Jenkins, 1996) and protein supplementation has improved cow performance (Miner et al. 1991). Endecott et al. (2007) supplemented 2- to 4-year olds cows with glucogenic precursors postpartum and reported that addition of glucogenic precursor to a protein supplement decreased days to first estrus in postpartum 2-year-old range cows. Additionally, Waterman, et al. (2006) reported increasing metabolizable protein from ruminally undegradable protein (RUP) with or without propionate in a supplement fed to 2 year-old cows increased decreased postpartum interval, however, pregnancy rates were not different. Knowing that the inclusion of propionate salt in a protein supplement improved reproductive performance in young cows grazing New Mexico rangeland, we evaluated the influence of propionate salt on young beef cow reproductive performance within the South Dakota rangeland environment.

 $¹$ This project was funded by the South Dakota State University Agricultural Experiment Station.</sup>

MATERIALS AND METHODS

Experimental Design

Studies were conducted over 2 consecutive years at SDSU research stations. The year 1 experiment was conducted at the Cottonwood Range Livestock Research Station near Philip, SD and year 2 experiment was conducted at the Antelope Range and Livestock Research Station near Buffalo, SD. Both stations are representative of Northern Great Plains mixed-grass prairie. Sixty 2- and 3- yr old cows (920 lbs; BCS = 4.46) were used in year 1, and 60 2-yr old cows (835 lbs; BCS = 3.94) were used in year 2. Although at different locations between years, cows were from the same cow herd and had similar genetics.

Prior to calving, cows were managed as one herd. Cows were managed in a small paddock close to the headquarters prior to calving, so assistance could be provided if required. After calving (≤ 3 d) cows were allocated to one of three supplementation treatments and moved to the experimental pasture. Cows had access to native pasture and hay in year 1. Hay (6.9% CP, 59.7% TDN) was provided due to limited available standing forage. At the Antelope Station in year 2, cows had access to a native range pasture (300 acres) where primary grasses were western wheatgrass, needle and thread, green needlegrass and blue grama.

In both years, cows were blocked by expected calving date and randomly assigned to one of the three propionate salt (Ca-Propionate; NutroCal, Kemin Industries) treatments. The three supplements were formulated on an as-fed basis to be isocaloric and isonitrogenous (Table 1). Propionate salt was incorporated into the protein supplement that was fed twice weekly at 2 lbs/d for 45 or 54 days in year 1 and 2, respectively.

Table 1. Ingredient and chemical composition of protein supplement (% of DM).

^a NutroCal, Kemin Industries, Inc., Des Moines, IA; source of Ca-propionate

Measurements

Measurements were obtained similarly in both years. Cows were weighed and assigned a BCS at calving, end of supplementation (Suppl), start of breeding season (Breeding) and weaning. Calves were weighed at birth, branding and weaning.

Blood samples were collected weekly starting at 10 days postpartum by venipuncture using a 10 mL vacutainer tubes (Fisher Scientific, Pittsburgh, PA). Blood was kept on ice until centrifuging at 3000xg for 20 minutes to harvest plasma. Plasma was stored at -20**°** C until assayed for progesterone (Engel et al., 2008). Return to estrous was determined when progesterone was greater than 1 ng/mL.

In year 1, bulls were placed with cows for a 60-d breeding season. Cows were synchronized with the Select Synch + Controlled Internal Drug Releasing device (CIDR) protocol. Cows were artificially inseminated (AI) at 72 h after CIDR removal and, in year 2, bulls were placed with cows 10 days following AI for a 60-d breeding season. Pregnancy rates were determined by transrectal ultrasonography at weaning in both years and, in year 2, AI pregnancy rates were determined 35 d following AI.

Statistical Analysis

Each year was analyzed as a separate experiment because of changes in location and age groups between years. Cow BW, BCS, ADG, BCS change, calf BW, calf ADG, postpartum interval, and days to conception response to level of supplemental propionate salt were analyzed in a randomized complete block design using the MIXED procedure of SAS (PROC MIXED, SAS Institute, Cary, NC). Cows were stratified into blocks by calving date and randomly assigned within each block to propionate salt treatments. Block was considered a random effect. Fixed effects were treatment, period, treatment x period, and cow age (year 1 only). Calf sex was treated as a fixed effect and calf birthdate as a covariate. Least squares means were calculated and linear and quadratic polynomial contrasts were constructed to evaluate the influence of increasing levels of propionate salt. Period (calving, Suppl, Breeding and weaning) was considered a repeated measure. Reproductive responses were analyzed using the GLIMMIX procedure of SAS. In year 1, level of supplementation, cow and the cow age by level of supplement interaction were analyzed. In year 2, only level of supplementations was analyzed.

RESULTS AND DISCUSSION

No differences in cow weight (P ≥ 0.11; Table 2) and BCS (P ≥ 0.17; Table 2) were found between treatments in either year. Cow weight changed through the study (P < 0.01; Table 3). Waterman et al. (2006) also reported that cow BW was not different between protein supplements with or without propionate salt treatments. In year 1, cow ADG had a treatment by period interaction (P < 0.05): ADG displayed a quadratic response (P < 0.05) to levels of propionate salt during the supplementation period with 80 g displaying the highest ADG, but no response during the Suppl to Breeding or Breeding to weaning periods. In year 2, cow weight change differed by period ($P < 0.01$); cows lost weight during the supplementation period (-0.70 lbs/d), and gained from Suppl to Breeding (11.2 lbs/d) and Breeding to weaning (0.84 lbs/d). Cow BCS changed through the study (P<0.01; Table 3). In year 1, cows BCS increased during supplementation (P<0.01, 0.42) and decreased from Breeding to weaning (-0.17).

Table 3. Influence of Period on Spring Calving Beef Cow Performance and Their Calf Performance

	Year 1^a			Year 2^a						
	Initial	Suppl	Brand	Breed	Wean	Initial	Suppl	Brand	Breed	Wean
Cow BW, lbs ^c	920	975		1030	1044	835	794		873	976
Cow BCSbc	4.5	4.9		4.9	4.7	3.9	4.1		4.1	4.7
Calf BW, lbs ^c	79		223		433	78		202		435

 a Initial = within three days of calving, Suppl = last day of supplementation period, Brand = day calves were branded, Breed = first day of breeding season, Wean = day calves were weaned. Supplementation period was 45 and 54 days in year 1 and 2, respectively.

 b Body Condition Score on a 9-point scale (1 = emaciated to 9 = extremely obese).

 c P-value < 0.01

Calf weight was not different (P \geq 0.38) among treatments. Calves gained an average of 350 lbs/d through the study.

Pregnancy rates did not differ between treatments ($P \ge 0.90$; Table 2) in either year. In year 1, there was no effect of cow age (P = 0.97), but based on ultrasonography, 3-year-old cows conceived earlier (P < 0.01, 183.7 d) than 2-year cows (207.0 d). Furthermore, the percentage of cows initiating estrous cycles before the breeding season was greater (P = 0.04) for cows receiving 160 g (47.6%) compared to 0 g (15.6%) propionate salt and tended to be greater than cows receiving 80 g (P = 0.07, 20.0%). In year 2, only 2-year old cows were used, and the percentage of cows initiating estrous cycles before the breeding season was not different among treatments (P = 0.28). Endecott et al. (2007) and Waterman et al. (2006) reported decreased days to first estrus with propionate salt supplements, which we observed in year 1 but not year 2.

Propionate salt did not influence cow BW, BCS or calf BW. Propionate salt supplementation influenced reproductive performance in year 1, not in year 2. Lack of significant differences in animal performance between propionate salt treatments could be due to 1) year to year variation in for forage quality and quantity and/or 2) age of animals. Year 1 had both 2 and 3 year old cows, but year 2 only had 2 year old cows on the project. In New Mexico, where propionate salt has been beneficial, cows have been grazing dormant rangeland during the breeding season (Waterman, 2006; Endecott, 2007) and do not reach

their lightest body weight until after the beginning of the breeding season. Based on the results of this project propionate salt has limited value for improving young cow's reproductive performance when grazing Northern Great Plains rangelands and breeding during the forage growing season.

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Effects of weaning age and winter development environment on heifer performance[1](#page-19-0)

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SUMMARY

Our objective was to determine if early weaning (about 125 d) vs. normal weaning (about 250 d) and wintering replacement heifers in drylot vs. rangeland affected heifer growth and reproductive performance. Heifer calves from the 2009 and 2010 calf crops (n = 104 and 73, respectively) were allocated to the 2 weaning treatments and then stratified by age into the 2 winter development treatments forming a 2 by 2 factorial arrangement of treatments. Heifers wintered in drylot received mixed grass and alfalfa hay (yr 1: 11.6% CP, 52.5% TDN; yr2: 12.3% CP, 53.4% TDN) plus 1.8 kg of a dried distiller's grain (DDGS)-based supplement/hd/d (yr1: 22.7% CP, 75.8% TDN; yr 2: 25.4% CP, 76.7% TDN). Heifers wintered on rangeland also received 1.8 kg/hd/d of the same supplement. Over the winter, each treatment was allocated to a separate pen or pasture. After estrus synchronization and timed AI, all heifers were placed on rangeland to graze through the summer. During the summer of yr 1, heifers were allocated by winter treatment to 2 pastures, and in yr 2 all 4 treatment combinations were allocated to separate pastures. Responses measured were BW, ADG, pubertal status at initiation of estrus synchronization, and pregnancy status after breeding. Pubertal status was indicated by serum progesterone > 1 ng/ml. A winter by weaning treatment interaction affected (*P*<0.001) BW and ADG both years. During the winter months, range heifers were lighter and grew slower than drylot heifers, but BW did not differ due to winter treatments at the end of the summer. However, early-weaned heifers remained lighter than normal weaned heifers at the end of the summer. Weaning treatment affected (P=0.03) fall pregnancy rate (93.2%±4.0 and 74.7%±7.98 for early- and normal-weaning, respectively) in yr 2. In yr 1, there was a difference ($P=0.006$) between drylot and range heifers (92.7%±3.52 and 72.8%±6.47, respectively) in the proportion that obtained puberty before estrus synchronization. In conclusion, producers should consider important interactions between weaning and winter management practices when establishing a replacement heifer development program that best fits the goals of their operation.

INTRODUCTION

There have been multiple research projects on different heifer development programs to evaluate effectiveness of alternative options (Olson et al., 1992; Arthington and Kalmbacher, 2003; Salverson et al., 2005). Past research has suggested that rangeland may be an effective resource to develop heifers that are destined to become range cows (Olson et al., 1992; Salverson et al., 2005). The objective of this study was to evaluate how age at weaning, 125-d-of-age (early) and 250-d-of-age (normal), and two winter development environments, rangeland and drylot, affected heifer growth and development. We hypothesized that heifers wintered on rangeland with supplementation would have lower ADG and would be lighter at initation of breeding compared to the heifers wintered in drylot, but that they would

 1 This research supported by USDA NRI competitive grant (2007-55618-18160)

still be adequately developed to have similar reproductive performance to drylot-raised heifers. We also hypothesized that range heifers that grazed alongside their mothers longer would have improved ADG after weaning and therefore normal-weaned heifers would have better reproductive performance than early-weaned heifers. We further hypothesized that wintering heifers in drylot would produce the same results for both early-weaned and normal-weaned heifers.

MATERIALS AND METHODS

All animal procedures were approved by the South Dakota State University Institutional Animal Care and Use Committee.

Design and Treatments

Heifer calves from the 2009 and 2010 calf crop (n = 104 and 73, respectively) were split into two groups to either be early weaned (**EW**, about 125 d of age) or normal weaned (**NW**, about 250 d of age). These groups were based on dam assignments to weaning treatments for another study that was ongoing. In that study, dams had been stratified into two groups and then each group was randomly assigned to either be early- or normal-weaned when they entered the study. Within the two weaning-age groups, heifers were stratified by age into two winter development treatments. These groups were either wintered in drylot (**D**) or wintered on rangeland (**R**) from weaning to breeding. This created the following four treatment combinations in a 2×2 factorial treatment structure: 1) early weaned and developed from weaning to breeding in drylot (**ED**); 2) early weaned and developed from weaning to breeding on rangeland (**ER**); 3) normal weaned and developed from weaning to breeding in drylot (**ND**); 4) normal weaned and developed from weaning to breeding on rangeland (**NR**). Heifers wintered in drylot received mixed grass and alfalfa hay (Table 1) ad libitum plus 4 lb. of a dried distiller's grain (**DDGS**)-based supplement/hd/d (Table 1). Heifers wintered on rangeland also received 4 lb/hd/d of the same supplement. During the winter when snow cover precluded grazing, Rheifers received the same hay as the Dheifers. Heifers in the ER treatment combination consumed 497 lb/hd of hay in year 1 and 671 lb/hd of hay in year 2. Heifers in the NR treatment combination consumed 482 lb/hd of hay in year 1 and 647 lbs/hd of hay in year 2. Over the winter, each treatment combination was allocated to a separate pen or pasture. After estrus synchronization and timed AI, all heifers were placed on rangeland to graze through the summer. During the summer of year 1, heifers were allocated by winter treatment to 2 pastures, and all 4 treatment combinations were allocated to separate pastures in the summer of year 2.

Collections

Heifer BW were recorded at EW (August 18, 2010; August 17, 2011), NW (November 3, 2010; November 2, 2011 [NW heifers only]), middle of the winter treatment period (March 9, 2010; February 4, 2011), first blood sampling (May 14, 2010; May 18, 2011), breeding (June 19, 2010; June 9, 2011), July pregnancy detection (July 29, 2010; July 26, 2011), end of summer grazing period (September 1, 2010; August 24, 2011), and fall pregnancy detection (November 3, 2010; October 20, 2011).

Puberty

Pubertal status of the heifers at the beginning of the breeding season was determined by analysis of serum progesterone. Blood samples were collected via jugular or coccygeal venipuncture into a 10-ml Vacutainer tube at d -10 (May 14, 2010; May 18, 2011), d 0 (May 25, 2010; May 30, 2011), and d 15

(June 9, 2010 [yr 1 only]), relative to the initial start of estrus synchronization. Blood sat at room temperature for 1 hr to clot and was then centrifuged for 20 minutes. Serum was harvested and frozen at -20º C until analysis. Serum progesterone concentrations were analyzed by a previously validated radioimmunossay (Engel et al., 2008). Heifers were defined as having reached pubertal status if serum progesterone was ≥ 1 ng/ml in either serum sample.

Table 1. Nutrient analyses of grass/alfalfa hay and $DDGS¹$.

 1 DDGS = dried distiller's grains with soluble- based cube

Breeding

On d -7 (May 25, 2010 and May 30, 2011) heifers received an estrus synchronization protocol and were bred by timed AI (June 19, 2010 and June 9, 2011). The synchronization protocol included: 100 µg GnRH (Cystorelin, Merial Marysville, Kansas) and Controlled Internal Drug Releasing device (**CIDR**) insertion on d -7; 25 mg PG (Lutalyse, Pfizer Kalamazoo, Michigan) and CIDR removal on d 0; and timed AI with 100 µg GnRH at 72-hr after CIDR removal. An error was made in yr 1 and the CIDRs were reinserted on d 8 to d 15 and heifers were bred on June 19, 2010. Timed AI was followed by a 45-d exposure to natural service to complete the breeding season. Semen-tested bulls were used with a bull:heifer exposure ratio not exceeding 1:28 both years.

Conception to AI was determined by trans-rectal ultrasonography on d 40 after AI in yr 1 and d 47 after AI in yr 2 (July 29, 2010 and July 26, 2011). Overall pregnancy rate was determined by rectal palpation in yr 1 and trans-rectal ultrasonography in yr 2 on d 90 (November 3, 2010 and October 20, 2011) after the breeding season.

Statistics

Heifer BW and ADG were analyzed using the MIXED procedure of SAS. The model included weaning treatment, winter treatment, and their interaction as independent variables. Time of weighing (or the intervals between weighing for ADG) and its 2- and 3-way interactions with weaning and winter treatments were included as repeated measures. Animal was included as a random effect and was considered the experimental unit.

Pregnancy rates and puberty status were analyzed using the GENMOD procedure of SAS with the use of the logit structure for binomial data. The model included independent variables of weaning treatment and wintering treatment as well as their interaction.

RESULTS AND DISCUSSION

Weaning treatment, winter treatment, and weigh period interacted for BW and ADG during both years (*P* < 0.001) (Tables 2, 3, 4, and 5). In both years, R heifers were lighter and grew slower than D heifers during the winter months. Within each winter treatment the EW heifers were also lighter than the NW heifers. Once spring green-up occurred, R heifers had an increase in ADG and continued to gain more than the D heifers throughout the summer. At the end of the study in year 1, there was no difference in BW between the two NW groups, and they were both significantly heavier than the EW groups. At the end of year 2, there was no difference between wintering treatments, however EW heifers were still lighter than NW heifers. This agrees with other studies by Lusby et al. (1981); Olson et al. (1992); and Arthington and Kalmbacher (2003).

Table 2. Effect of weaning at an average of 125- or 250-d of age and development from weaning to breeding in a drylot or on range on BW in 2010 heifers

 a,b,c Means within a row with different superscripts differ ($P < 0.05$).

 1 August 18, 2009 for early-weaned and November 3, 2009 for normal-weaned.

a,b,c Means within a row with different superscripts differ (*P* < 0.05).

	Early Weaned			Normal Weaned		
Date	Drylot	Range	Drylot	Range		
Birth, lb	84.5 ± 2.6	84.5 ± 2.4	82.3 ± 3.0	83.2 ± 2.9		
8/17/2010, lb	387.0 ± 15.4	386.1 ± 14.7	370.9 ± 17.8	377.3 ± 17.0		
Weaning ¹ , lb	387.0 ± 11.4^a	386.1 ± 10.9^a	542.7 ± 13.2^b	538.8 ± 12.8 ^b		
2/4/2011, lb	608.5 ± 10.8^b	$519.4 \pm 10.3^{\circ}$	$667.3 \pm 12.5^{\circ}$	578.2 ± 12.1^b		
5/18/2011, lb	729.7 ± 11.5^b	649.9 ± 11.0^a	716.5 ± 13.3^b	720.5 ± 12.8^b		
Breeding (6/9/11), lb	698.9 ± 10.7°	$670.6 \pm 10.2^{\circ}$	746.0 ± 12.3^b	752.0 ± 11.9^b		
Pregnancy Check	$724.7 \pm 10.5^{\circ}$	735.7 ± 10.1^{ab}	759.7 ± 12.1^b	812.9 ± 11.7^c		
$(7/26/11)$, lb						
$8/24/11$, lb	820.4 ± 12.3 ^a	840.2 ± 11.8 ^a	886.8 ± 14.1^b	923.1 ± 13.7^b		

Table 4. Effect of weaning at an average of 125- or 250-d of age and development from weaning to breeding in a drylot or on range on BW in 2011 heifers

 a,b,c Means within a row with different superscripts differ ($P < 0.05$).

¹ August 17, 2010 for early-weaning and November 2, 2010 for normal-weaned.

Table 5. Effect of weaning at an average of 125- or 250-d of age and development from weaning to breeding in a drylot or on range on ADG in 2011 heifers

			Normal Weaned		
Drylot	Range	Drylot	Range		
1.30 ± 0.035 ^c	0.77 ± 0.033 ^a		0.53 ± 0.018^b		
			$1.85 \pm 0.40^{\circ}$ $1.03 \pm 0.049^{\circ}$		
		Early Weaned $1.30 \pm 0.042^{\circ}$ 0.77 $\pm 0.040^{\circ}$ 0.73 ± 0.051^a 1.21 $\pm 0.048^b$ $1.58 \pm 0.097^{\circ}$ $2.22 \pm 0.095^{\circ}$	$1.74 \pm 0.042^{\circ}$ $1.32 \pm 0.048^{\circ}$ 0.19 \pm 0.022 ^a $0.64 \pm 0.057^{\circ}$ $0.63 \pm 0.025^{\circ}$		

a,b,c Means within a row with different superscripts differ (*P* < 0.05).

In y 1, more D heifers obtained puberty before the breeding season than R heifers (*P* = 0.006; 92.7 ± 3.5% vs. 72.8 ± 6.5%, respectively). However, after heifers were initially exposed to progestin (immediately before the CIDR were re-inserted), there was no difference in pubertal status (*P* > 0.05; 96.8 ± 1.8%). In y 2 there was no difference (*P* > 0.05) in the percentage of heifers that obtained puberty between treatments (99 ± 2.8%). Other studies have also shown that as long as heifers obtain an appropriate percentage of mature BW by initiation of breeding, winter gain should not affect puberty at breeding (Lemenager et al., 1980; Clanton et al., 1983; Lynch et al., 1997).

AI conception rate did not differ among treatments in either y 1 or 2 ($P > 0.05$; 53.7 \pm 7.1% and 48.2 \pm 8.5%, respectively), consistent with no differences in percentage of heifers that were pubertal at initiation of breeding. In yr 1, there was also no difference in overall pregnancy rate between treatments (*P* > 0.05; 86.7 ± 5.0%). This supports previous finding by Lynch et al. (1997), Martin et al. (2008), and Funston and Larson (2011). However, in y2 more (*P* = 0.03) EW heifers were pregnant at fall pregnancy diagnosis than NW heifers (93.2% \pm 0.040 and 74.7% \pm 0.080, respectively).

Wintering heifers on rangeland or early weaning could be a beneficial option for certain heifer development programs. A producer needs to look at important interactions between weaning and winter treatment when selecting a development program that best fits the goals of their operation.

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Effects of weaning age and winter development environment on heifer grazing distribution[1](#page-25-0)

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SUMMARY

The objective of this experiment was to determine if early weaning (approximately 125 d) vs. normal weaning (approximately 250 d) and wintering replacement heifers in drylot versus rangeland affected heifer grazing distribution during the subsequent summer. Heifer calves from the 2009 and 2010 calf crop (n = 104 and 73, respectively) were allocated to the 2 weaning treatments and then stratified by age into the 2 winter development treatments. During the summer of yr 1 heifers were allocated to 2 pastures by winter treatment, and in yr 2, all 4 treatment combinations were allocated to separate pastures. A subset of heifers from each group was selected to wear global positioning system (GPS) collars (n=2 and 5 in yr 1 and 2, respectively). Readings were taken from the GPS every 15 min in yr 1 and every 65 s in yr 2. The GPS coordinates were then analyzed relative to ecological sites, water locations, fence locations, and temperature using Arc GIS (ESRI, Redlands, CA). Winter treatment affected (*P*<0.05) preference index (PI) for claypan and loamy sites in 2010, and distance from water in 2011. Day of sampling affected (*P*<0.05) claypan and loamy site PI in 2010 and thin claypan site PI in 2011. Day of sampling interacted with winter treatment (*P*<0.05) for distance from water in 2010, sand and thin claypan site PI in 2010 and thin claypan site PI in 2011, while day of sampling interacted with weaning treatment for distance from water in 2011. A winter by weaning treatment interaction affected (*P*<0.05) thin claypan site PI in 2011. Temperature had an effect on distance to fencelines in 2010 (*P*<0.001). There was a temperature interaction with wintered treatment effect on distance to water in 2011 (*P*<0.001). There was a three-way interaction (*P*<0.05) between weaning treatment, winter treatment and ambient temperate on the distance from water and between weaning treatment, winter treatment and day of sampling on claypan and sand site PI in 2011. In conclusion, winter development influenced patterns of range utilization. Day-of-sampling interactions indicated that range heifers did not adjust preferences and thus were already adapted to the range environment, whereas drylot heifers adjusted preferences over time suggesting they re-learned how to utilize the range environment.

INTRODUCTION

Past research has suggested that rangeland may be an effective resource to develop heifers that are destined to become range cows (Olson et al., 1992; Salverson et al., 2005). Learning and retention of grazing skills may play an important role in the development of productive range cows that are capable of harvesting adequate grazed nutrients to meet their requirements, therefore requiring minimal outside feed resources. Factors that affect grazing behavior include, but are not exclusive to, early life experiences, presence of peers, physiological or nutritional state of the animal, inherited senses, and spatial memory (Launchbaugh and Howery, 2005). Some specific factors that have been found to

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influence cattle grazing habits are vegetation biomass available in the area and water location (Gillen et al., 1984; Owens et al., 1991; Pinchak et al., 1991; Cibils et al., 2008).

The objective of this study was to determine how weaning treatment and winter development environment of beef heifers affected grazing distribution and pasture utilization throughout the following summer months. We hypothesized that range-raised heifers would have a more broad pasture distribution and better utilization. This was based on the idea that range heifers grazing throughout winter would retain grazing skills and cues learned from dams while suckling and that drylot heifers would have a re-learning period before they would be able to fully utilize the pasture. We also hypothesized that normal-weaned heifers would have better grazing habits than early-weaned heifers because they spent more time grazing with their dams learning and retaining grazing skills.

MATERIALS AND METHODS

All animal procedures were approved by the South Dakota State University Institutional Animal Care and Use Committee.

Design and Treatments

Heifer calves from the 2009 and 2010 calf crop (n = 104 and 73, respectively) were split into two different groups to either be early weaned (**EW**- about 125 d) or normal weaned (**NW**- about 250 d). These groups were based on cow assignments to weaning treatments for another study that was currently ongoing. In that study, cows were stratified into two groups and then each group was randomly assigned to either be early- or normal-weaned when they entered the study. Within the two weaning-date groups, heifers were stratified by age into two groups and each group was randomly assigned to a post-weaning development treatment. These groups were either developed through the winter from weaning to breeding in drylot (**D**) or developed from weaning to breeding on rangeland (**R**). This created the following four treatments: 1) early weaned and developed in drylot (**ED**); 2) early weaned and developed on rangeland (**ER**); 3) normal weaned and developed in drylot (**ND**); or 4) normal weaned and developed on rangeland (**NR**). Heifers developed in drylot received mixed grass and alfalfa hay (yr 1: 87.1% DM, 11.6% CP, 52.5% TDN; yr 2: 87.3% DM, 12.3% CP, 53.4% TDN) ad libitum plus 4 lb of a dried distiller's grain (**DDGS**)-based supplement/hd/d (yr 1: 93.4% DM, 22.7% CP, 75.8% TDN; yr 2: 91.8% DM, 25.4% CP, 76.7% TDN). Heifers developed on rangeland also received 4 lb/hd/d of the same supplement. During the winter when the ground was covered in snow to a depth that precluded grazing, range heifers received the same hay as the drylot heifers. Heifers in the ER treatment consumed 498 lb/hd of hay in yr 1 and 672 lb/hd of hay in yr 2. Heifers in the NR treatment consumed 483 lb/hd of hay in yr 1 and 648 lb/hd of hay in yr 2. More hay was fed to both treatment combinations in year 2 because of a greater number of days with heavy snow cover. Over the winter, each treatment combination was allocated to a separate pen or pasture. After estrus synchronization and timed AI (June 19, 2010 and June 9, 2011), all heifers were placed on rangeland to graze through the summer. During the summer of yr 1, heifers were allocated by winter treatment to 2 pastures, and all 4 treatments were allocated to separate pastures during the summer of yr 2.

Year 1 GPS Locations

Two heifers from each treatment combination were selected based BW close to the mean treatment group 365-d BW and average phenotype to represent their group's grazing habits. These heifers wore a global positioning system (GPS) – very high frequency (VHF) collar (GPS-Log-V2, Kedziora Innovation

Group Mannsville, NY) that recorded location at 15-minute intervals throughout the summer grazing season. Data were collected over a 74-d period from June 19 to August 31, 2010.

Year 2 GPS Locations

Five heifers were randomly selected from each group to wear a GPS collar using the same selection criteria as year 1. These collars contained a Garmin GPS unit (eTrex Legend H, Garmin, Olathe, Kansas) that recorded location every 65 seconds. Data were collected over a 76-d period from June 9 to August 24, 2011.

GPS Analysis

Location data points for each heifer were mapped onto a layer in ArcMap (Rock Wars, Golden CO) and intersected with a layer for 4 ecological sites (claypan, loam, sand, and thin claypan) that were common to all pastures. The number of GPS points on each ecological site per day was divided by the total GPS points that given day, resulting in the percentage of time spent each day on a given ecological site. That number was then multiplied by the percentage of the total area occupied by that ecological site to calculate the preference index (**PI**).

Fencelines and water sources were mapped using a GeoExplorer[®] GeoXH (Trimble[®] GPS Navigation; Sunnyvale, CA). With the use of Arc Map, the distance between every GPS data point for each heifer and the nearest water source and fenceline was calculated. Mean distances from the fencelines and water sources for each heifer were found for each hour of each day over the entire summer grazing period. Hourly ambient temperature readings were collected from an onsite weather station.

Statistical analysis

Preference indexes for ecological sites were analyzed by repeated measures using the MIXED procedure of SAS. The model included the independent variables of weaning treatment, wintering treatment, and day of the grazing season, along with their 2- and 3-way interactions. Day of the grazing season was considered a continuous variable and therefore treated as a covariate. Fence and water distances were also analyzed by repeated measures using the MIXED procedure of SAS using the same model.

The temperature effect on distance from water and fenceline was also analyzed as described above, with the model consisting of the independent variables of weaning treatment, wintering treatment, temperature and their 2- and 3-way interactions.

RESULTS AND DISCUSSION

Ecological site PI

In yr 1 R heifers preferred (*P* = 0.01) claypan ecological sites more than D heifers (PI of 6.46 ± 0.661 and 1.14 \pm 0.410, respectively). There was a tendency ($P = 0.10$) for R heifers to increase their PI by 0.07 \pm 0.028 each day of the grazing season. However, over the summer all groups had an increase of 0.04 \pm 0.018 units of PI for claypan sites (*P* = 0.02). In yr 2, there was a three-way interaction between day, weaning treatment, and winter treatment on claypan PI. The following regression equations were generated:

ED claypan PI = $33.0 + (-0.500 \times day)$ ER claypan PI = $-0.04 + (0.19 \times day)$ ND claypan PI = $0.09 + (0.01 \times day)$ NR claypan PI = $2.5 + (-0.03 \times day)$

ER and ND did not have a preference for claypan sites and NR had a slight preference. None of these preferences changed over the summer. In contrast, ED heifers had an extremely high initial PI for claypan sites, which then decreased through the grazing season (*P* < 0.001)

Drylot heifers had a greater initial preference (*P* < 0.001) for loam ecological sites than R heifers (1.76 ± 0.54 and 0.31 \pm 0.080, respectively) and all groups had an increase ($P < 0.001$) of 0.01 \pm 0.002 preference units each day in yr 1. There were no differences in PI for loam sites in yr 2.

In yr 1, R heifers had a greater initial preference (*P* = 0.02) for sand sites than D heifers and increased preference throughout the summer *(P* = 0.001), whereas D heifers had no change in preference as the summer progressed. The following regression equations were generated:

> D sand PI = $0.57 + (0.004 \times day)$ R sand PI = $2.40 + (0.03 \times day)$

In yr 2, there was an interaction among period, weaning treatment and winter treatment (*P* = 0.007) for sand site PI. The following regression equations were generated:

> ED sand PI = $0.3 + (0.03 \times day)$ ER sand PI = $0.59 + (-0.004 \times day)$ ND sand PI = $1.43 + (-0.01 \times day)$ NR sand PI = $1.96 + (0.05 \times day)$

Normal-weaned R heifers initially had a higher PI than other treatments that increased (P < 0.001) each day. All other groups had a smaller initial preference that did not change during the summer.

Thin clay pan site (TCP) preference in yr 1 was initially higher in D heifers than R heifers. The following regression equations were generated:

> D TCP PI = $1.04 + (-0.006 \times day)$ R TCP PI = $0.84 + (0.001 \times day)$

Over the summer D heifers decreased (*P* < 0.001) preference and R heifers had no change. In yr 2, there was a weaning treatment by winter treatment interaction on thin clay pan site preference. Normal R heifers had a PI of 0.553 ± 0.087, which was similar to ED with a PI of 0.73 ± 0.082 and different from ER (0.95 ± 0.081) and ND (1.06 ± 0.087) . ED was also similar to ER, but different from ND. All groups had a 0.005 ± 0.002 increase in PI over the summer (*P* < 0.001).

Distance from Fences

In yr 1, the R heifers' average distance from the fenceline was greater ($P = 0.03$) than the D heifers (459.3 ± 16.5 ft and 316.3 ± 13.4 ft, respectively). However, in yr 2 there was a tendency (*P* = 0.09) for a weaning by winter treatment interaction that indicated ED heifers mean distance was further from the fence than the NR heifers (449.5 \pm 41.7 ft and 262.5 \pm 49.5 ft, respectively). Heifers mean distance from the fence increased as temperature increased at 5.3 ± 1.3 ft (P < 0.001) per degree C in yr 1. However, in yr 2 there was only a tendency ($P = 0.09$) for a 4.1 \pm 2.4 ft increase in distance from the fenceline per degree C as temperature increased.

Distance from Water

Past research has found that the location of water influences pasture distribution (Gillen et al., 1984; Owens et al., 1991; Pinchak et al., 1991; Cibils et al., 2008) and our finding support this. In yr 1, there was an interaction between day and winter treatment ($P = 0.02$). The R heifers started the grazing season at a closer mean distance to water than the D heifers (387.2 \pm 123.0 ft and 1650.3 \pm 112.2 ft, respectively), and the average distance for R heifers did not change (*P* > 0.05) over the summer but the D heifers moved closer to water at a rate of 9.74 ± 3.12 ft/day (*P* = 0.002). This suggests that once the D heifers became more familiar with where the water source was in relation to other resources, they did not venture as far away. In yr 2, there was no change over time in relation to the winter treatment, but the average distance to water was greater ($P < 0.001$) for the R than the D heifers (1446.9 \pm 60.0 ft and 564.3 ± 57.7 ft, respectively). There was also a day by weaning treatment interaction (*P* = 0.002). The following regression equations were generated:

EW distance to water = 1036 ft + $(2.46$ ft \times day)

NW distance to water = 1377 ft - $(8.46 \text{ ft} \times \text{day})$

The EW heifers started the grazing season closer to water than NW, and the EW mean distance to water did not change (*P* > 0.05) over the season while the NW heifers reduced distance over time (*P* < 0.001). Early-weaned D heifers had a tendency ($P = 0.07$) to be closer to water than ER heifers in yr 2 (324.2 \pm 69.0 ft and 1775.0 \pm 88.3 ft, respectively). There was also a tendency ($P = 0.09$) in yr 2 for the mean distance from water to decrease by 3.0 ± 1.8 ft/day. This suggested that the ER heifers had improved grazing distribution relative to the other treatments combinations because they had spent the greatest amount of post-weaning time in the grazing setting on rangeland.

There was an interaction between temperature and winter treatment for influence of temperature on distance from water in yr 1 (*P* < 0.001). The following regression equations were generated:

D distance to water = 1352 ft + $(-35.5 \text{ ft} \times \text{°C})$

R distance to water = 202 ft + $(-4.9 \text{ ft} \times \text{°C})$

Drylot-developed heifers traveled further from water than range-developed heifers at cooler temperatures, but ventured closer to water as temperature increased. In yr 2 there was a three-way interaction between temperature, weaning, and winter treatments $(P = 0.007)$. The following regression equations were generated:

> ED distance to water = 127 ft + $(-1.8 \text{ ft} \times \text{°C})$ ER distance to water = 412 ft + (7.2 ft \times °C) ND distance to water = 270 ft + (-2.82 ft \times °C) NR distance to water = 497 ft + (-7.56 ft \times °C)

Both ED and ND heifers started closer to the water than ER and NR heifers and did not change distance with temperature. However, ER heifers increased distance and NR heifers reduced distance as temperature increased. This suggested that other factors besides water location affected grazing distribution.

Heifer grazing distribution is based on learned experiences, social cues and pasture environment. In this study, heifers that were wintered on rangeland initially utilized the pasture more evenly because the drylot heifers apparently needed to have a learning period. Past research has shown that water and vegetation location influence cattle pasture distribution and our results support that. By wintering heifers on rangeland, a producer will have heifers that do not require a learning period during the summer and will be able to better utilize their pastures.

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Comparing Pfizer GeneSTAR and Igenity PROFILE DNA tests in crossbred cattle¹

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SUMMARY

Our objective was to compare the performance of Pfizer's GeneSTAR and Igenity's PROFILE DNA tests in crossbred beef cattle. Hair follicles were collected from 394 crossbred steers that were part of the South Dakota State University Calf Value Discovery project (n = 145) and calves that were fed at the SDSU Southeast Research Farm (n = 249) in 2011. Hair follicles were sent to Pfizer Animal Genetics and Igenity for testing with their GeneSTAR and PROFILE DNA tests, respectively. Marbling score (100-999 scale), ribeye area, fat thickness, carcass weight, yield grade, quality grade, and % kidney, pelvic, and heart fat was collected post-harvest on all steers. Using this dataset, we first asked whether each DNA test was associated with marbling scores. If the DNA tests were not associated with marbling, then the tests may not be useful for predicting genetic merit for marbling in crossbred cattle. The GeneSTAR and PROFILE DNA tests were associated with marbling scores, but this association was not high. Second, we asked whether GeneSTAR and PROFILE DNA test results were associated with each other. If test results were not correlated, then using both DNA test results together may improve genetic predictions. GeneSTAR and PROFILE tests for tenderness were highly correlated, suggesting that DNA markers for tenderness are similar for both tests. GeneSTAR and PROFILE DNA tests for marbling and feed efficiency were not highly correlated, raising the possibility that both tests could be used jointly to improve genetic predictions for these traits. However, using both GeneSTAR and PROFILE DNA tests jointly did not appreciably improve predictions for marbling score in our dataset. We conclude that, although both GeneSTAR and PROFILE DNA tests were associated with marbling score, the correlation between these tests and marbling was low in our sample of crossbred cattle. Further, using GeneSTAR and PROFILE DNA tests for marbling and feed efficiency may improve genetic predictions for these traits, but we did not find evidence that using both tests jointly improves predictions for marbling.

INTRODUCTION

Genomic tests have the potential to allow producers to more accurately predict genetic merit for their cattle. More accurate genetic predictions will improve response to selection and allow producers to manage their cattle more effectively. Several DNA tests are available to beef producers, but most of these tests are specific to only a single breed (e.g., Angus). However, two commercially available DNA tests are marketed to producers raising cattle of any breed type and composition.

GeneSTAR is one of these DNA tests. GeneSTAR is marketed by Pfizer Animal Genetics and predicts genetic merit for three traits: marbling, feed efficiency, and tenderness. The DNA test results are reported to producers as "Most Valuable Predictions" (MVP). Lower MVP for feed efficiency and tenderness and higher MVP for marbling are more desirable. Along with the MVP, the % ranking of the individual relative to all other animals in the Pfizer GeneSTAR database is reported.

The second commercially available DNA test for cattle of all breed types is Igenity's PROFILE test. Igenity's PROFILE predicts genetic merit for more traits than GeneSTAR: residual feed intake, ADG, marbling, % choice, yield grade, fat thickness, heifer pregnancy rate, stayability, maternal calving ease, and docility. Unlike GeneSTAR, PROFILE reports DNA test results to producers as a categorical 1-10 score. A score of 1 is most favorable for residual feed intake and yield grade, while a score of 10 is most favorable for all other traits.

Although both tests have been evaluated independently, GeneSTAR and PROFILE test results have not been compared to each other in the same sample of animals. Presently, producers have little to no information on whether GeneSTAR or PROFILE should be used in their herds. Further, genetic predictions may be improved by using both DNA tests jointly if correlations between GeneSTAR and PROFILE tests for the same trait are low. Our objectives were as follows:

1) Test whether Pfizer's GeneSTAR and Igenity's PROFILE DNA tests were associated with marbling score in a sample of crossbred beef cattle.

2) Estimate the correlation between the GeneSTAR and PROFILE DNA test results. If the correlation between GeneSTAR and PROFILE Marbling tests is low, test whether using both tests jointly improves genetic predictions for marbling.

MATERIALS AND METHODS

Animal and Trait Data

Crossbred steers were sampled from the 2011 South Dakota State University (SDSU) Calf Value Discovery (CVD) project (n = 145) and the SDSU Southeast Research Farm (n = 249). For the 2011 CVD project, 9 producers enrolled steer calves. Calves were finished at a commercial feedlot (Vander Wal Yards, Bruce, SD) for 211 or 231 days prior to slaughter. Carcass data from these steers was collected in June 2011. Carcass data collected includes ribeye area, marbling (100-999 scale), fat thickness, HCW, dressing %, USDA yield and quality grade, and % kidney, pelvic, and heart fat (% KPH).

Crossbred steers sampled from the SDSU Southeast Research Farm were either purchased at an auction barn (n = 178) or raised at the SDSU Cottonwood Research Station (n = 71). At the SDSU Southeast Research Farm, carcass data was collected in June and August 2011. Carcass data collected includes HCW, USDA yield and quality grade, ribeye area, marbling (100-999 scale), fat thickness, and % KPH.

DNA Sample Collection

Hair follicles were removed from the tail switch of steers by hand and placed into hair follicle sample collectors following the instructions of the two DNA testing companies. Hair follicle samples from each steer were sent to Pfizer and Igenity for DNA testing. Pfizer and Igenity extracted the DNA from hair follicles and tested DNA with their GeneSTAR (Pfizer) and PROFILE (Igenity) marker panels. The DNA test results were subsequently returned to the investigators.

Data Analysis: Effect of DNA tests on marbling scores

The effects of each DNA test on marbling score were estimated with a linear model. Marbling score was the dependent variable and the independent variables were marbling DNA test score (MVP for

GeneSTAR and 1-10 categorical scores for PROFILE), herd (CVD or Southeast Research Farm), and HCW. All independent variables were fixed effects except for HCW, which was modeled as a covariate. Steers at the Southeast Research Farm were concurrently part of an externally funded nutrition study. The nutrition study did not affect carcass performance and thus nutritional treatment was not included in our model.

The effect of the PROFILE DNA test scores for ribeye area, fat thickness, and USDA yield and quality grade were also estimated on their respective traits with the same linear model as described above. The GeneSTAR DNA test does not estimate genetic merit for these traits, so only the PROFILE DNA test was evaluated. All statistical analyses were completed in JMP v 8.0 software (SAS, Cary, NC). A *P* value less than 0.05 was considered to be statistically significant.

Data Analysis: Correlation between GeneSTAR and PROFILE DNA tests

For comparing GeneSTAR and PROFILE DNA test results, Pearson correlation coefficients were estimated between all pairs of DNA test scores. Because the correlation coefficient between marbling GeneSTAR and PROFILE DNA test scores was low, we included both marbling GeneSTAR and PROFILE test scores in our linear model described above for the statistical analysis for objective 1. As for objective 1, correlation coefficients were estimated with JMP v 8.0 software.

RESULTS AND DISCUSSION

Descriptive Statistics

Table 1. Description of carcass traits in our study population.							
Carcass trait	Sample mean	SD	Minimum	Maximum			
HCW , lbs. (n = 391)	756.8	71.44	424.9	954.6			
Dressing % ($n = 145$)	62.9	2.42	45.0	67.0			
Ribeye area, in ² (n = 390)	11.86	1.047	9.57	15.01			
Backfat, in $(n = 390)$	0.51	0.150	0.10	1.00			
% KPH 1 (n = 390)	2.06	0.207	1.52	2.65			
Yield grade, $1-5$ (n = 391)	2.88	0.697	1.00	4.00			
Marbling, 100-999 (n = 390)	483	90.4	250	755			

Table 1. Description of carcass traits in our study population.

 1 % Kidney, pelvic, and heart fat

¹ Pfizer Animal Genetics, Kalamazoo, MI

PROFILE score	Sample mean	SD	Minimum	Maximum
Residual feed intake ($n = 388$)	6.41	1.17	3	9
ADG ($n = 389$)	5.85	1.04		9
Tenderness ($n = 377$)	6.29	1.95		10
Marbling ($n = 389$)	6.65	1.11	4	10
% Choice (n = 389)	6.65	1.11		10
Yield grade ($n = 389$)	6.26	1.05	3	9
Backfat (n = 389)	5.56	0.99	3	8
Ribeye area ($n = 388$)	4.74	0.96		8
Heifer pregnancy rate ($n = 388$)	5.50	1.13		8
Stayability ($n = 388$)	6.55	1.11	3	10
Maternal calving ease ($n = 389$)	5.03	1.03		8
Docility ($n = 388$)	6.08	0.99		9

Table 3. Description of Igenity PROFILE test results.

 $¹$ Neogen Corporation, Lansing, MI</sup>

Objective 1 Results

The Pfizer GeneSTAR marbling MVP and Igenity marbling PROFILE were significantly associated with marbling score ($P < 0.05$). A 100-unit change in marbling score resulted in a 1.7-unit change in GeneSTAR MVPs and a 5.56-unit change in PROFILE score. The amount of variation in marbling score explained by the GeneSTAR marbling and PROFILE tests was 0.01 and 0.049 respectively. The PROFILE test scores for yield grade and fat thickness were significantly associated with their respective phenotypes (*P* < 0.05). The PROFILE score for ribeye area, however, was not significantly associated with actual ribeye areas (*P* $= 0.38$).

Objective 2 Results

Pearson correlation coefficients were estimated for all pairs of DNA test results (Table 4). Most of the correlation coefficients were low (r < 0.20). The highest correlation coefficients were usually between DNA tests for carcass traits. The correlation between the tenderness GeneSTAR and PROFILE DNA panels was -0.62. This coefficient is negative because of the definitions of the DNA test scores: larger values are more favorable for the tenderness PROFILE and smaller values are more favorable for the tenderness GeneSTAR MVP. Correlation coefficients between feed efficiency and marbling GeneSTAR MVP and PROFILE scores were lower. The correlation coefficient between feed efficiency MVP and PROFILE scores was 0.14 and the correlation coefficient between marbling MVP and PROFILE scores was 0.13.

Because correlation estimates were low between DNA test results for marbling, we tested whether using marbling DNA test scores jointly could improve predictions for marbling score. Including both DNA tests in our model for marbling score did not appreciably improve marbling score predictions. Both marbling DNA tests accounted for only 5.4% of the variation in marbling score. We could not repeat the same analysis for feed efficiency because individual feed efficiency data was not available.

Within DNA panels, the highest correlation was between marbling and % choice PROFILE scores. The correlation coefficient was 1.0 between these DNA panels. Thus, both panels include the same DNA markers, which makes sense because % choice is only determined by marbling and maturity. Maturity is not a genetically determined trait. The correlation between fat thickness and yield grade PROFILE scores was also high (r = 0.66), suggesting both panels harbor similar DNA markers.

Interpretation of Results

In our sample, both DNA panels were statistically associated with marbling score; however, the effect sizes were small. These results are similar to what is reported by the companies offering these genetic tests. Why were the effect sizes so small? Many non-genetic factors affect marbling and other carcass traits in cattle. For example, the CVD project animals were taken from 9 cow-calf producers which each raised and fed their cattle differently during the pre-weaning phase. These environmental differences between calves before entering the feedlot would have affected marbling and other carcass traits. Additionally, each of these genetic tests only include a small number of genes that affect marbling score. Many genes that affect marbling have not yet been identified and thus could not be included in the DNA tests.

The GeneSTAR and PROFILE tenderness tests were highly correlated, suggesting that each company is using similar genes to estimate genetic merit for tenderness. This result is not surprising because several genes with large effects on tenderness have been discovered (e.g., calpastatin and u-calpain). Genetic testing has the potential to increase the accuracy of our genetic predictions for a large number of traits. However, genetic selection is only one of several tools available to beef producers for improving carcass characteristics. Environmental effects (e.g., nutrition, management) also affect carcass traits. Additionally, genetic tests available to crossbred commercial beef cattle only include a fraction of the total number of genes that affect economically important traits. As the technology improves, these tests should become better predictors of genetic merit for commercial producers.

Table 4. Pearson correlation coefficients^{1,2} between DNA score panels³

¹ Off-diagonals represent the correlation coefficient between two different DNA panels. For example, r = 0.218 is the correlation between the GeneSTAR marbling Most Valuable Prediction (MVP) and GeneSTAR feed efficiency MVP.

 2 95% confidence intervals for correlation coefficients ranged from 0.11 to 0.20.

 3 FE = Feed efficiency; Marb = Marbling; Tend = Tenderness; RFI = Residual feed intake; % Ch = % Choice; YG = Yield grade; Fat = Fat thickness; REA = Ribeye area; HP = Heifer pregnancy rate; Stay = Stayability; MCE = Maternal calving ease; Doc = Docility; MVP = Most valuable prediction

BEEF 2012-06

Relationship between fat content and NE values for some ethanol byproducts[1,](#page-37-0)[2,](#page-37-1)3

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SUMMARY

A finishing phase study was conducted to measure the impact of the fat content of ethanol byproducts on the relative Net Energy values for these feedstuffs.

The three feedstuffs with varying crude fat content used included a commercially available corn gluten feed corn distillers grains blend (CGD); a corn wet distillers grains without solubles (WDG); and corn wet distillers grains plus solubles (WDGS). Byproducts were incorporated as 40% of the finishing diets (DM basis) replacing corn and SBM components of the control (CO) diet. There were 6 pens of 7 or 8 yearling steers on each treatment during the 130 d experiment.

The assayed average dietary fat content for the CO, CGD, WDG, and WDGS diets were 2.91%, 4.95%, 5.34% and 6.58%, respectively. Increasing fat content of diets (and byproducts) was associated with increased dietary NE content. Diet NE_G values derived from steer performance were 60.6, 59.7, 62.0 and 64.6 Mcal/cwt for CO, CGD, WDG and WDGS, respectively. Assuming that the substituted grain had a NE_G value of 68 Mcal/cwt, the derived NE_G values for the three byproducts were 65.8, 71.5 and 78.1 Mcal/cwt for the CGD, WDG and WDGS diets, respectively. Regression of diet fat content against NE_G content indicated that fat represented 2.02 times the NE content of non-fat diet components (P < .02; r^2 $= 0.26$).

INTRODUCTION

It is a logical priority in the advancement of renewable biofuels technology to seek to capture as much of the energy potential as possible from the basal feedstuff. The energy rich oil component of corn becomes a key focus of this strategy. The impact of this is an increase in production of lower fat content distillers grains. We now see a range of 4 to 15% fat in the distillers grains that are incorporated into feedlot diets.

Many finishing diets are based upon a high inclusion level of biofuels byproducts. An important question is how the changing fat content of the byproducts will impact dietary NE levels and cattle performance.

 ¹ IACUC approval 11-034E.

² Project funded by the Beef Nutrition Program and the Ag Exp Station, SDSU.

³ The authors wish to express appreciation to R. Knock, DakotaLand Feeds for assistance in coordinating availability of specialty feeds.

MATERIALS AND METHODS

This experiment was conducted at the Ruminant Nutrition Center during June through October 2010. Yearling steers were on test for 130 d. The finishing diets were formulated to contain 8.5% roughage and a 50:50 ratio of Dry Rolled Corn:High Moisture Corn grain base. Byproducts were included at 40% of the diet replacing, corn and a protein supplement. Three byproducts were used to provide varying dietary fat content. Those included a commercially available corn gluten-feed distiller's grains blend (CGD), wet distiller's grains without solubles (WDG) and wet distillers grains with solubles (WDGS). These byproducts were received as needed in 20T loads and bagged to preserve quality while feeding.

Step-up diets of 50, 30 and 16% roughage were used to adapt steers to the final diets. Byproduct inclusion was at 30% in the first two step-up diets. Final test diets were first offered at d 19. Each feed ingredient was sampled weekly. Actual diet formulations and compositions were back calculated weekly from feed batching records and the weekly ingredient assay values. Diets shown in Table 1 reflect the actual formulation and composition values for the study for days 21 to 130. Feed was mixed and delivered twice daily.

Steers (n = 190) were from a single source and purchased through a South Dakota sale barn. Standard Ruminant Nutrition Center receiving protocols were followed. The BW recorded during processing was used for allotment purposes. The allotment involved stratifying the allotment BW similarly across four treatments and then across 6 replicate pens within treatment. At the onset of the study, there were 8 steers per pen in replicates 1 to 5 and 7 steers per pen in replicate 6.

Individual BW was acquired at the onset of the experiment and again at 28, 56, 84, 111 and 130d. Revalor S™ implants (Intervet) were administered concurrent to weighing steers on d 28. Weighing was done prior to making morning feed deliveries. Feed records were summarized at intervals corresponding to weigh dates. Shrink was not applied to interim performance data. At termination steers were comingled and slaughtered as a single lot. Individual identity was maintained during slaughter and matched to camera grading data acquired from the packing plant.

Cumulative live performance was calculated by applying a 3% shrink to the BW measured at d 130. Carcass adjusted performance was calculated using a final BW derived from HCW (HCW/0.625) to exclude potential diet effects on fill.

Data were analyzed as appropriate for a completely random design experiment with pen representing the experimental unit. Tests for linear or quadratic responses to diets were based upon equally spaced polynomials and were not weighted for actual dietary fat content. The dietary NE_M and NE_G values were calculated for each individual pen based upon BW, DMI and live weight gain. The NE_G value for each byproduct was estimated based on actual substitution level in the diet and an assumed NE $₆$ of 68</sub> Mcal/cwt for the displaced corn-supplement mixture. Regression analysis was used to predict the effect of dietary fat (actual) on the diet NE_G values derived from steer performance.

RESULTS AND DISCUSSION

Dietary targets were met (Table 1). Byproduct inclusion averaged 40.2% and did not differ (P > .20) among diets CGD, WDG and WDGS. Hay content (8.2%) did not differ among diets. As anticipated, crude protein was higher ($P < .05$) in diets containing byproducts. The rationale was to meet the CP

requirements in the CO diet and assume no additional CP response in the higher CP content byproduct diets. The dry supplement used in the control diet contained 75% CP. The formulation was 91.2% SBM, 8.8% urea. All diets differed (P < .001) in EE content. The magnitude of differences was not as large or as uniform as anticipated because of higher than anticipated EE content of the CGD feed (Table 2).

Adaptation to diets was faster when byproducts were fed. This was a reflection of differences in dietary starch content influences on acid challenges to the rumen during adaptation and feeding aggression by the steers. Even at the lower DMI, the Control step-up diets resulted in twice as much daily corn intake compared to the corresponding steps of diets containing byproducts. Intakes by the steers fed the CO diet lagged behind the others through 56 d on feed (Table 3). It appears that the adaption to the more readily fermentable CO diet resolved during the period of 57 to 84 d on feed. During this period intakes were similar and CO steers exhibited higher ADG and lower F/G. The influence of WDGS to cause higher DMI became more pronounced as the days on feed progressed.

Cumulative performance was very good. Rankings appear similar whether assessing performance on a live BW or carcass adjusted BW basis. Increasing dietary fat content caused linear improvements (P < .01) in ADG, DMI and F/G. Most of this response was due to the WDGS treatment.

Generally carcass differences were small and consistent with increased DMI and growth rate. Dressing percentage and HCW increased linearly (P < .01) with increasing dietary fat (Table 5). There was also a trend for ($P = 0.11$) for Yield Grade to increase across treatments. The WDG treatment tended (P = 0.11) to cause lower marbling scores and that was reflected in a 20% point decline in premium grade carcasses. In several previous studies we observed reduced Quality Grade relative to total carcass fatness associated with feeding ethanol byproducts. The response has been inconsistent and we have not been able to identify a component within these feeds that leads to the effect.

There were 6 pen replicate estimates of dietary NE for each treatment. The NE values increased linearly across diets (Table 6). The CGD diet contained 2% more fat than the CO diet and only 0.4% less fat than the WDG diet, but had a NE_G value that ranked numerically lower than the CO diet. In contrast, the NE_G of the WDG diet was intermediate to the values for the CO and WDGS diets. This indicates that the NE of the CGD diet was driven by feed components other than fat. The estimate of byproduct NE_G content was calculated merely by substitution. Since actual byproduct inclusion levels were similar the statistical inferences are identical for the byproduct and complete diet NE_G estimates.

Obviously, there are several nutrition issues in play as we substitute ethanol byproducts for corn in finishing diets. Even so, these diets do allow a cursory approach to estimating how byproduct fat content may impact NE_G . When diet NE_G were regressed against dietary mean fat content the ensuing equation was: NE_G = 57.32 + 1.02(%EE) (P = 0.02; r^2 = 0.26). This suggests that NE_G changes 1 Mcal for every 1% point change in dietary fat. The r^2 is not compelling, but 1 Mcal NE_G/1% point fat is biologically reasonable. It infers that the fat has 2-fold the NE_G content of the fuel (carbohydrate) that it replaced.

We were fortunate to receive excellent cooperation from an ethanol producer that allowed us to have WDG and WDGS produced in the same production runs. That allows us to make a more direct comparison of these two products. Returning the solubles increases the fat content of the byproduct and dilutes the fiber component (Table 2). When the difference in NE_G between only the WDG and WDGS is divided by the change in fat content the coefficient becomes 2.02 Mcal $NE₆/1%$ point fat. The coefficient is probably inflated by the NE_G value for the non-fat fuel contained in solubles being higher than the NE_G content of the non-fat fuel (fiber) of the distillers' grains.

These results support the argument that removing fat from biofuel byproducts will lower the NE value for the feeds. The rate of change is approximately 1.02 Mcal $NE₆/cut$ for each 1% point change in fat content. The WDG-WDGS comparison emphasizes the importance of components other than fat in affecting NE content of these feedstuffs. Knowledge of manufacturing processes that cause these compositional differences would be helpful when pricing and doing formulations. More extensive feed characterization is recommended to make effective predictions of relative energy values of these evolving feedstuffs.

Table 1. Actual formulation and composition for control, corn gluten feed-distiller's grains blend (CGD), wet distiller's grains (WDG) and wet distiller's grains plus solubles (WDGS) diets.^{1, 2}

 1 DM basis.

 2 Based on weekly ingredient analyses; n = 17; mean (std dev.).

³Supplement contained 640 g /T monensin, 156g /T tylosin and provided minerals and vitamins to meet or exceed NRC requirements.

 4 Based upon tabular values for ingredients; assuming co-product NE at par with corn (100/68).

Table 2. Composition of corn gluten feed-distiller's grains blend (CGD), wet distiller's grains (WDG) and wet distiller's grains plus solubles (WDGS) byproducts $fed¹$

 1 All values (except DM) are DM basis.

 2 Number of lots used and analyzed in this study.

	Control	CGD	WDG	WDGS	SEM	$p = \frac{2}{3}$
1 to 28 d						
Initial BW, lb	851	851	852	854	2.0	
d28 BW, lb	980	992	995	995	5.0	0.050
ADG, lb	4.57	4.99	5.12	5.12	0.178	0.050
DMI, lb	20.65	22.19	21.84	22.43	0.122	0.001
F/G	4.55	4.46	4.30	4.43	0.148	0.467
29-56 d						
d56 BW	1072	1089	1096	1105	5.4	L < 0.001
ADG	3.28	3.47	3.60	3.91	0.138	L < 0.01
DMI	21.45	23.14	22.26	22.58	0.286	$L = 0.08; Q = 0.03$
F/G	6.61	6.71	6.20	5.80	0.246	$L = 0.02$
57 to 84 d						
d84 BW	1204	1204	1218	1228	1.6	L < 0.02
ADG	4.71	4.10	4.38	4.42	0.265	Q < 0.10
DMI	24.08	24.14	23.39	24.09	0.317	NS ³
F/G	5.19	5.94	5.44	5.51	0.299	$Q = 0.15$
85 to 111 d						
d111 BW	1312	1310	1325	1351	7.8	L 0.002; Q 0.10
ADG	3.99	3.92	3.96	4.58	0.125	L 0.005; Q 0.017
DMI	25.72	25.91	25.35	26.66	0.309	L 0.12; Q 0.09
F/G	6.52	6.66	6.46	5.88	0.182	L 0.02; Q 0.07
112 to 130 d						
d130 BW	1380	1387	1388	1431	9.0	$L < 0.002$; Q 0.06
ADG	3.60	4.04	3.37	4.22	0.205	NS
DMI	25.65	25.90	25.13	27.21	0.374	$L < 0.05$; Q < 0.05
F/G	7.19	6.44	7.64	6.51	0.365	NS

Table 3. Interim periods cattle performance summary when control, corn gluten feed-distiller's grains blend (CGD), wet distiller's grains (WDG) and wet distiller's grains plus solubles (WDGS) diets were fed.¹

 1 non shrunk BW basis.

² Probability of linear (L) and quadratic (Q) contrasts of relative diet fat content.

 3 NS $P > 0.20$.

	Control	CDG	WDG	WDGS	SEM	$P=1$
Initial BW, lb	851	851	852	854	2.0	
Live Basis						
Final BW, $lb2$	1339	1345	1346	1388	8.7	L 0.002; Q 0.06
ADG, lb	3.76	3.80	3.81	4.11	0.074	L 0.005; Q 0.102
DMI, lb	23.36	24.14	23.49	24.38	0.162	L0.005
$F/G,$ lb	6.23	6.36	6.17	5.93	0.106	L 0.039; Q 0.106
Carcass Adjusted ³						
Final BW, lb	1333	1343	1347	1394	9.4	L 0.001; Q 0.072
ADG, lb	3.71	3.79	3.82	4.16	0.080	L 0.002; Q 0.120
$F/G,$ lb	6.30	6.39	6.16	5.87	0.129	L 0.019; Q 0.170

Table 4. Cumulative performance summary on live and carcass weight adjusted basis when control, corn gluten feed-distiller's grains blend (CGD), wet distiller's grains (WDG) and wet distiller's grains plus solubles (WDGS) diets were fed.

 1 Probability of linear (L) and quadratic (Q) contrasts of diet fat content.

 2 d130 BW with 3% shrink.

 3 HCW/0.625.

Table 5. Carcass traits, Quality Grade, and Yield Grade distributions when control, corn gluten feeddistiller's grains blend (CGD), wet distiller's grains (WDG) and wet distiller's grains plus solubles (WDGS) diets were fed.

	Control	CDG	WDG	WDGS	SEM	P ¹
Dress, % $2^{\overline{2}}$	62.25	62.42	62.56	62.75	0.214	L 0.010; Q NS^4
HCW, lb	833	839	843	871	5.7	L 0.001; Q 0.064
Ribfat, in.	0.50	0.52	0.54	0.55	0.021	L 0.125; Q NS
REA, in^2	12.74	12.48	12.47	12.90	0.246	L NS; Q 0.162
Marbling 3	569	578	533^{\ddagger}	572	12.1	NS
YG	3.21	3.39	3.46	3.49	0.098	L 0.1101; Q NS
Y1 & 2, % Y 3, % $Y3.5 - 4.0, %$ Y4, %	23.9 47.8 21.7 6.5	17.0 38.3 36.2 8.5	18.2 38.6 29.6 13.6	17.4 30.4 39.1 13.0		Chi square NS
Prime & Prem. Choice, % Low choice, % Select, % No Roll, %	32.6 43.5 21.7 2.2	36.1 44.7 19.2 $\pmb{0}$	11.4 59.1 29.6 0	34.8 43.5 21.7 0		Chi square NS

Table 6. Performance based calculations of diet and by-product NE values for corn gluten feed-distiller's grains blend (CGD), wet distiller's grains (WDG) and wet distiller's grains plus solubles (WDGS).

 $¹$ Derived using actual performance data in NE calculations published by Galyean (2005).</sup>

² Assumed corn = 68 Mcal/cwt. Eq. [(Test NE_G - Control NE_G) / % co-product] + 68.

BEEF 2012-07

Cost analysis of cattle feedlot designs[1](#page-44-0) , [2](#page-44-1)

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SUMMARY

A cost analysis of fixed and non-fixed costs of gain was conducted on 3 cattle feedlot designs. The three facility designs compared were conventional open pens (OPN), open pens with shelter over the feeding area (OS), and a monoslope confinement barn (MON). The OPN design was the least expensive facility to build and operate. However, because of poorer cattle performance (*P* < 0.05), it was not the most cost effective. The MON design had significantly higher operating costs when compared to the OPN or OS designs, especially for the tractor/spreader (*P* < 0.05), skid loader (*P* < 0.10), labor (*P* < 0.05), and straw (*P* < 0.05). The operating and fixed costs, combined, made the MON design the most expensive cost of gain design (*P* < 0.05). The OS design was the most expensive facility to build but with current feed prices and the cattle performance, it was the most cost effective cattle feeding design.

INTRODUCTION

Various cattle feeding facility designs exist in the northern plains states with little information available to aid managers in making facility decisions. Cattle performance (ADG, F/G, and health), fixed, and nonfixed costs must all be considered to determine the most cost effective feedlot design. Fixed costs will include land and construction costs. Non-fixed, or operating costs include machinery hours, labor, and bedding. The amount and composition of manure generated by cattle housed in each design could also impact economic outcomes. This study was conducted to determine which cattle feedlot design is the most cost effective.

MATERIALS AND METHODS

The data were collected at the South Dakota State University Foundation Opportunities Farm near Lennox, South Dakota. The Opportunities Farm is home to a 960-head cattle feedlot consisting of three contrasting designs, confinement (MON), open (OPN), and partially covered (OS). Each design consists of four pens with a capacity of 80-head per pen. The confinement design is a 360 ft x 40 ft building with each pen being 90 ft wide and 40 ft deep (45 ft² per head). The open design is an earthen-mound pen design that is 80 ft wide and 275 ft deep (275 ft² per head). The partially covered design combines a building and earthen-mound pens. The building is 320 ft x 35 ft and covers the feed ally, bunk, water fountain, and front 20 ft of the pen. The earthen lots are 215 ft deep, combined with the building, to allow 235 ft² per head of pen space. All pens included 80 ft of fenceline feedbunk.

 1 Authors extend appreciation to E. Loe, S. Holt, and M. Loewe for their contributions to compilation of this data.

 2 This study was funded by the Beef Nutrition Program.

Land area required per design was calculated by using the stocking density for each respected design. This value was used to calculate the acres required per design and was multiplied by the current land value of \$4,680/ac. The land expense was annualized over 20 years. The actual 2004 construction costs were corrected for inflation to estimate 2010 construction costs of OPN \$521.23, OS \$741.73, and MON \$682.24. Each design was depreciated over 20 years. To establish a daily fixed cost per head, construction and land costs were summed, assuming a 90% occupancy rate.

Cattle feeding activities contributing to the non-fixed costs were recorded daily for a 3-yr period. These activities included machinery hours, labor, and straw and were totaled by week and divided by actual head days. This established an amount of each input required per head per day for each design. Values were assigned for each input (Table 1) and the cost per 1,000 head per day was calculated as the nonfixed cost.

Table 1. Unit of Measurement for Inputs

 1 South Dakota Agricultural Land Market Trends 1991-2010, Janssen, et al

² Farm Business Management, University of IL at Urbana-Champaign April 2010

³ Ag Decision Maker, IA State University March 2010

⁴ USDA-SD Ag Market News, Weekly East River SD Hay Market 6 August 2010

The historical database of cattle performance differences across pens by design at the Opportunities Farm was used to calculate costs of gain. There were 27 contemporary group comparisons in that database, spanning 7-yr. In the database, each group of cattle arriving at the Opportunities Farm was allocated across the three feedlot housing designs. This balanced data set allowed for a comparison, by system, of estimated ADG and F/G.

The fixed and non-fixed costs were summed to determine a total cost per 1,000 head per day. This number was divided by the respective ADG for each design to establish the non-feed cost of gain. Differences were calculated between the OPN vs. MON and OPN vs. OS. With these two comparisons, the feed to gain of each respective design was multiplied by the cost of feed to establish where feed price, feed costs and non-feed cost of gain reach equilibrium.

The costs generated during this study are not reflective of full yardage costs. Items assumed equal across designs were not accounted for. Therefore results represent only the relative differences in cost of gain. Items assumed equal across designs included:

- Sick / death loss
- Time required to pull a sick animal
- Weather
- Feed preparation and delivery
- Cattle gender
- Manure value
- Carcass merit, other than weight (Wulf, 2007)

The amount of each input and non-feed cost of gain was tested by constructing an ANOVA table using the Proc GLM (generalized linear model procedure) in SAS (SAS Institute, Cary, NC). It was a randomized block design, where year was included as the block and design means tested with error=year(system). Quarterly least square means were tested using the Fisher t-test. The yearly least square means were used to compare relative costs between systems.

RESULTS AND DISCUSSION

Loe (2007) reported that cattle performance differed between designs (*P* < 0.01). Cattle fed in the MON and OS covered designs achieved faster gains and were more efficient (Table 2).

Table 2. Cattle performance differences for open (OPN), open with shelter (OS), and monoslope confinement (MON) housing systems

	MON	OPN	OS	SEM	P Value
ADG	3.57^{a}	3.51 ^b	3.62 ^a	0.075	0.008
F/G	ა.75 $^{\mathrm{b}}$	7.00 ^a	6.77°	0.149	0.002

¹Loe (2007 internal data summary)

a,b Means within main effect without common superscripts differ

Several of the operating inputs differed between designs. The confinement design required more (*P* < 0.05) tractor/spreader and skid loader hours, labor hours, and bedding (Table 3). The open and partially covered designs both required more loader tractor hours (P < 0.10).

Table 3. Amount of variable inputs for open (OPN), open with shelter (OS), and monoslope confinement (MON) housing systems¹

¹Inputs are per day per 1,000 head

 a, b Means within row without common superscripts differ ($P < 0.05$)

 d, e Means within row without common superscripts differ ($P < 0.10$)

The MON design was the most expensive to operate (non-fixed costs), with no difference between the OPN or OS covered design.

Figure 1. Costs/100lb of gain for open (OPN), open with shelter (OS), and monoslope confinement (MON) housing systems. FNFCG : Fixed Non-Feed Cost of Gain; VNFCG : Non-Fixed Non-Feed Cost of Gain; NFCG : Total Non-Feed Cost of Gain.

a,b,c Means within main effect without common superscripts differ (*P*<0.05)

A unique and powerful aspect of this data set is the 7-yr of commercial pen scale cattle performance available. Performance comparisons were replicated 27 times. These three feedlot designs are at the same location, and have common nutrition and management. This allows for the key comparison of costs per unit of production. Differences in operating costs per 100 lb of live weight gain (LWG) are shown in Figure 1. The fixed non-feed cost of gain (FNFCG) reflects depreciation cost for the land and facilities assuming a 90% occupancy rate. Costs ranged from \$2.36 for the OPN to \$3.19/cwt for the OS design. Statistical analysis was not applied to these costs because we have only a single construction cost observation for each design. The differences in FNFCG (\$0.83) are relatively small when total cost of gain is exceeding \$80/cwt.

The variable cost/LWG (VNFCG) were higher (P<.05) in MON; and similar between the OPN and OS designs. When fixed and variable costs were pooled the NFCG differed (P< .05) for each design. The NFCG difference between the MON and OPN designs was attributable to the additional \$1.97 /CWT cost of gain for tractor/manure spreader, \$1.03 /CWT cost of gain for skid loader, and \$2.58 /CWT cost of gain for bedding. The difference in NFCG between the OPN and OS covered facilities was primarily due to the difference in fixed costs (land and construction).

The added expense associated with providing shelter in feedlot pens has to be offset by improved cattle performance, especially improved feed efficiency. In this study the MON and OS systems cost more to build and to operate but did result in improved feed efficiency over feeding in OPN pens. The economic value of the improved feed efficiency is dependent on the cost of feed. Cost equilibrium would be where saving in feed cost/LWG brought about by improved efficiency equals the added NFCG expense. Comparing the confinement and open designs, feed must reach roughly \$489/T before the superior feed

efficiency of cattle fed in the confinement system would offset the \$6.11 /cwt higher NFCG. Comparing the open and open with shelter designs, feed must cost more than \$122/T for the added feed efficiency of cattle fed in the partially covered facility to offset the added cost to build the facility.

In the current feed cost environment open with shelter design is the most cost effective facility design for feeding cattle in this environment.

BEEF REPORT 2012-08

SDSU Calf Value Discovery 2011 Summary Report[1](#page-49-0)

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INTRODUCTION

The Calf Value Discovery Program (CVD) allows cow-calf producers to gain knowledge of the finishing segment of the beef cattle industry and the marketing of fed cattle. Specifically, CVD provides an opportunity for cow-calf producers to learn how their calves perform in a feedlot and their carcass value when sold in a value-based marketing system. Each producer taking part in the program could consign a minimum of 5 steers weighing between 500 and 800 pounds to the CVD program. Animals were finished in a calf-fed program using typical diets and management protocols at VanderWal Yards (Bruce, SD). Carcass and feedlot performance information from calves were returned to producers for use in making future management decisions to improve profitability.

MATERIALS AND METHODS

Eleven cow-calf operations in South Dakota and Minnesota consigned calves to the 2011-2012 CVD program. The number of animals consigned by producers ranged from 5 to 72. Calves were received on November $8th$ and $9th$, 2011. Upon receipt, calves were vaccinated against viral and bacterial respiratory (Bovi-Shield Gold BVD, Inforce 3, One-Shot, Pfizer Animal Health, Kalamazoo, MI) and clostridial pathogens (Ultrabac-7, Pfizer), dewormed (Dectomax Pour-On, Pfizer), individually identified, and weighed. Calves were implanted on d 35 (Synovex S, Pfizer) and on d 105 (Revalor-S, Merck Animal Health, Summit, NJ). Cattle were fed a finishing diet based on high moisture ground ear corn, modified wet distillers grains, and corn silage as a group in a single pen. Cattle were visually evaluated for degree of finish and sold in semi-load lots when deemed to have approximately 0.4 inches of backfat. Slaughter dates were May 11, June 1 and 15, 2012 (184, 205, and 219 days on feed, respectively). Animals were sold on a quality/yield grid at Tyson Fresh Meats, (Dakota City, NE).

For each animal, individual BW was recorded at arrival at the feedyard, on d 35, d 105, and one day before shipment to slaughter. A four percent shrink was used for all weights. Since cattle were fed in a single pen, individual feed intake was calculated based on animal performance and diet energy content using NRC (2000) equations. At slaughter, individual identification was tracked through the plant and individual carcass data, including HCW, 12^{th} -rib fat thickness, ribeye area, percent kidney, pelvic, and heart fat, marbling score, and USDA Quality and Yield Grades, were reported by the plant.

Actual expenses included feed, based on calculated individual intake as a fraction of actual feed delivery, yardage (\$0.35 per animal daily), and veterinary expenses. Feeding expenses associated with calves that died during the program was distributed equally across all other

 ¹ Salaries and research support provided by state and federal funds appropriated to South Dakota State University.

animals in the program. Actual grid prices were used to calculate carcass value and feeding period profitability. To estimate feeder calf price, and overall profitability, average feeder steer prices from South Dakota Auction Market Summary report (USDA Agricultural Market Service report SF_LS795) for the weeks ending October 31, and November 7 and 14, 2012 were regressed on selling weights. The resulting equation was used to calculate feeder calf price for each calf: Price (\$/100 lb) = 277.65 - (BW \times 0.1147) (r^2 = 0.81). The Nebraska weighted average carcass price for negotiated direct sales was used (USDA Agricultural Market Service report LM CT158) for the weeks animals were sold was used to estimate carcass value and profitability had animals been sold on a dressed basis.

To estimate what factors were associated with feeding performance or profit and quality grade for calves that finished the 2011-2012 CVD program, calves were divided into thirds based on profit. A generalized linear model was used to separate means between groups (PROC GLM, SAS Institute, Cary, NC). The association of USDA Quality Grade among profit groups was determined by χ^2 (Proc Freq, SAS). Means were considered different when $P \le 0.05$.

RESULTS AND DISCUSSION

Four calves (1.63%) died during the program, and the remaining 240 animals finished the program and were included in the analysis. Overall cattle performance data is included in Table 1. Calves were placed with an average weight of 569 ± 87 lb, but the range in weights was 459 lb. Some calves were accepted into the program whose initial weights were outside the recommended range of 500 to 800 pounds. Average final BW for steers (average DOF = 201) was 1,245 ± 110.1 lb, and ADG was 3.37 lb/d. Dry matter intake of was 20.2 lb/d, which was 2.2% of average BW, and F:G ratio was 6.01 on average. The 2011-2012 winter was extremely mild in eastern South Dakota, with little precipitation. This likely contributed to the good performance. On average, steers were slaughtered at target fat thickness, but considerable variability existed among steers. Steers graded well, with 67.1% Grading US Choice or Prime, with only 9 Yield Grade 4 carcasses and 2 Yield Grade 5 carcasses. In a pen-based scenario, approximately 10% of carcasses should have Yield Grades of 4 and 5 for maximal profit (Walter and Hale, 2011). In this case, had some animals been fed longer, overall quality grade might have been improved.

On average, feeding costs were \$565.31 per animal. This equates to a total cost of gain of \$83.62/100 lb. When carcasses were sold on a grid marketing basis, price ranged from \$1,173.37 to 1,954.15, but had carcasses been sold on a dressed basis this range would have been narrower (\$1,184.49 to \$1,863.22). When including the value of the feeder calf, there was a \$633.86 dollar per animal range in return from a loss of \$220.95 to a profit of \$412.91. However, on average total profit was \$48.20 per animal. Since 1973, feeding cattle has been approximately a breakeven business (Walter and Hale, 2011). For the whole group, carcasses sold for \$11.42 per carcass more on a grid basis, than on a dressed basis. This resulted in approximately the same profit advantage to selling on a grid for the whole group. However, on an individual basis, there was less potential profit for the highest profit animals when they were sold on a dressed basis. Generally, animals that had the best potential to match the grid should have been sold on a grid, but there would have been less price risk for animals that did not conform to the grid by selling on a dressed basis.

When sorted according to profit groups, the High profit steers had heavier placement and final weights, and had a greater ADG compared to the Low and Middle profit groups. Typically, higher profit in cattle feedlots is associated with superior feed efficiency, but because DMI was

calculated from NRC (2000) models, DMI and subsequently F:G may be overestimated for some high gaining animals. Because of this higher DMI, High profit steers had greater feed costs than Low, with Middle intermediate. However, superior performance and carcass value overcame this expense. In addition, while High profit steers were placed on feed weighing 154 pounds more than the Low profit steers, they were on feed 9 fewer days, which helped reduce yardage and feed costs. On average, costs associated with treating illness were significantly greater for Low profit groups than Middle and High.

Steers in the High profit group had greater HCW, dressing percentage, and ribeye area than Low profit steers, with Middle profit steers intermediate. Similarly, High Profit steers had greater marbling scores, and lower kidney pelvic and heart fat, and tended to have lower yield grades than Low profit steers, with Middle profit steers intermediate. Twelfth-rib fat thickness, however, was not different among profit groups, indicating that the previously mentioned differences in carcass characteristics were not influenced by degree of finish. Superior HCW and quality grade resulted in carcass values of \$1,714.08 for High profit steers compared to \$1,551.27 and \$1,376.34 for Middle and Low, respectively. However, because of the higher feed costs for High and Middle compared to Low the difference in feeding period profit was not as great as the difference in carcass value (High minus Low profit = \$300.10). In addition, High and Middle steers had greater placement weights, and thus greater feeder calf values compared to Low. Therefore, the difference in total profits between High and Low was only \$149.18 per animal. Had steers been sold on a dressed basis, the carcass value and profit to Middle steers would have been similar to when animals were sold on a grid. For Low profit steers, selling on a dressed basis would have an advantage of \$8.36 in carcass value compared to selling on a grid, but High profit steers were valued \$40.05 more per carcass on the grid than on a dressed basis.

For individual producers, the Calf Value Discover program provides feedback on feeding performance and carcass characteristics of calves. In addition, viewing animals from an individual ranch in the context of the entire group can provide a benchmark for comparison with cattle from other operations. In general, cattle with greater potential to perform and producer heavier carcasses were more profitable than those with lower weight gains. Higher profit steers also had superior USDA Quality Grades and tended to have better Yield Grades. Ultimately, market conditions and input prices can greatly impact feeding profitability from year to year, but these data provide useful guidelines for making selection and marketing decisions in the future.

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Item	Mean	Standard Deviation	Minimum	Maximum
Days on Feed	201	14.2	184	219
BW, lb				
$D0$	569	87.0	340	799
D 105	934	107.5	641	1,273
Final	1,245	110.1	979	1,549
ADG, lb/d	3.37	0.412	2.17	4.97
DMI, lb/d	20.2	1.84	15.9	26.8
F:G	6.01	0.40	5.01	8.17
HCW, lb	795	75.2	615	966
Dress., %	63.8	1.76	58.1	68.1
12-th rib fat thickness, in.	0.46	0.14	0.11	1.30
Rib eye area, in. ²	13.0	1.7	9.8	18.9
KPH, %	1.91	0.19	1.41	2.52
Marbling Score ^a	421.6	77.6	285.0	770.0
Yield Grade	2.50	0.75	1.0	5.0
Quality Grade Distribution	N	Percent		
Prime, %	$\overline{2}$	0.8		
Choice, %	159	66.3		
Select, %	76	31.7		
No roll, %	3	1.3		

Table 1. Overall performance and carcass characteristics of cattle enrolled in the 2011-2012 South Dakota Calf-Value Discovery Program.

 a^a Marbling score: 300-399 = Slight, 400 – 499 = Small, 500 -599 = Modest.

Item	Mean	Standard Deviation	Minimum	Maximum
Feeder calf cost, \$/steer	914.91	84.92	641.18	188.67
Feeding costs, \$/steer				
Feed cost	444.36	36.78	331.79	543.60
Treatment costs	7.08	17.12	0.00	101.03
Total Feeding cost	565.31	42.51	438.20	702.43
Grid marketing profit				
Carcass value, \$/carcass	1,547.23	156.97	1,173.37	1,954.15
Carcass price, \$/100 lb	192.23	5.98	173.14	223.13
Live price, \$/100 lb	122.57	5.54	102.58	148.73
Feeding profit, \$/steer	963.11	135.80	636.13	1,349.09
Total profit, \$/steer	48.20	86.37	-220.95	412.91
Dressed marketing profit				
Carcass value, \$/carcass	1,535.81	144.31	1,184.49	1,863.22
Feeding profit ^a , \$/steer	951.70	122.61	647.76	1,265.19
Total profit, \$/steer	36.79	68.12	-219.20	211.38

Table 2. Feeding expenses and carcass values of cattle enrolled in the 2011-2012 South Dakota Calf-Value Discovery Program.

^aFeeding profit is carcass value minus feeding cost.

		Profit Group			
Item	Low	Middle	High	SEM	P-Value
n	80	80	80		$\overline{}$
Days on Feed	206 ^a	201 ^b	197 ^b		0.003
BW, lb					
D ₀	487 ^a	578^b	641°	6.69	< 0.001
D 105	834 ^a	941^{b}	$1,028^c$	7.94	< 0.001
Final	1,136	$1,255^b$	$1,343^c$	7.87	< 0.001
ADG, lb/d	3.17 ^a	3.38^{b}	3.57 ^c	0.042	< 0.001
DMI, lb/dd	18.6 ^a	20.3^{b}	21.6 ^c	0.154	< 0.001
F:G	5.94 ^a	6.01 ^{ab}	6.09 ^b	0.044	0.054
HCW, lb	716 ^a	801 ^b	868^c	4.76	< 0.001
Dress., %	63.0^{a}	63.8^{b}	64.6 ^c	0.183	< 0.001
12-th rib fat thickness, in.	0.45	0.48	0.47	0.016	0.41
Rib eye area, in. ²	12.0 ^a	13.0 ^b	14.1 ^c	0.162	< 0.001
KPH, %	2.01 ^a	1.89^{b}	1.82^c	0.020	< 0.001
Marbling Score ^e	402 ^a	414 ^a	449 ^b	8.42	< 0.001
Yield Grade	2.60	2.56	2.35	0.083	0.07

Table 3. Feedlot performance and carcass characteristics of steers enrolled in the 2011-2012 South Dakota Calf-Value Discovery Program according to profit group.

a,b,c Means differ; P-values noted in table.

dCalculated from BW and ADG

 e^{e} Marbling score: 300-399 = Slight, 400 – 499 = Small, 500 -599 = Modest.

				Profit Group			
		Low		Middle	High		
Quality Grade	n	Percent	n	Percent	n	Percent	P-Value
Prime	0	0		0.1		0.1	0.02
Choice	44	55.0	48	60.0	67	83.8	
Select	24	30.0	30	38.5	12	1.5	
No Roll	2	0.3		0.1	0	0	
Yield Grade							
	5	6.3	9	11.3	8	10.0	0.24
	29	36.3	23	28.8	36	45.0	
	40	50.0	42	52.5	36	45.0	
	5	6.3	4	5.0	Ω	$\mathbf{0}$	
		1.3		1.3	0	0	

Table 4. Quality and Yield Grade Distributions of steers enrolled in the 2011-2012 South Dakota Calf-Value Discovery Program according to profit group.

		Profit Group			
Item	Low	Middle	High	SEM	P-Value
Feeder calf cost, \$/steer	831.98 ^a	929.83^{b}	982.91^{b}	6.44	< 0.001
Feeding costs, \$/steer					
Feed costs	420.16^a	445.47 ^b	467.67°	3.51	< 0.001
Treatment costs	12.44^{b}	6.27 ^a	3.55 ^a	2.25	0.01
Total costs	546.52 ^a	565.07^{b}	584.35 ^c	4.45	< 0.001
Grid marketing profit					
Carcass value, \$/carcass	$1,376.34^a$	$1,551.27^b$	$1,714.08^c$	8.36	< 0.001
Carcass price, \$/100 lb	189.68^a	191.55^{b}	195.45^c	0.614	< 0.001
Live price, \$/100 lb	119.10^a	122.24^{b}	126.36 ^c	0.524	< 0.001
Feeding profit ^d , \$/steer	810.89 ^a	967.45^{b}	$1,110.99^c$	6.51	< 0.001
Total profit, \$/steer	-21.09 ^a	37.62^{b}	128.09 ^c	6.82	< 0.001
Dressed marketing profit					
Carcass value, \$/carcass	$1,384.70^a$	$1,548.71^{b}$	$1,674.03^c$	9.21	< 0.001
Feeding profit ^ª , \$/steer	819.25^a	964.90 ^b	$1,070.94^c$	7.40	< 0.001
Total profit, \$/steer	-12.73 ^a	35.07 ^b	88.03 ^c	6.09	< 0.001

Table 5. Feedlot performance and carcass characteristics of steers enrolled in the 2011-2012 South Dakota Calf-Value Discovery Program according to profit group.

a,b,c Means differ; P-Values noted in table.

^d Feeding profit is carcass value minus feeding cost.

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Investigation of protease activity in early postmortem muscle subjected to alternative chilling conditions[1](#page-57-0)

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SUMMARY

The objective of this study was two-fold: 1) determine the conditions (temperature, pH) that exist in early postmortem muscle of normally-chilled and delay-chilled beef carcasses to provide a model for *in vitro* work, and 2) determine the mechanism by which specific early postmortem temperature/pH conditions found in normally-chilled and delay-chilled muscle influence the enzymes that regulate the aging process *in vitro*. For objective 1, seven market-ready calves (HCW 848 ± 17.5 lb) were harvested with the right sides subjected to normal chilling (2.3°C) and the left sides subjected to ambient temperature (delay chilling; 22.6°C) for an additional 4.75 h and then allowed to normally chill. Carcasses were monitored for *longissimus dorsi* muscle temperature, pH, postmortem proteolysis, and sarcomere length. Steaks aged for 1, 7 and 14 d were evaluated for tenderness using Warner-Bratzler shear force (WBSF) and carcasses were swabbed over the rump, flank, and brisket for total aerobic plate counts. Delay-chilled carcasses had a faster pH decline and a slower rate of carcass cooling (*P*<0.05). No differences were seen in sarcomere length, total plate counts, or in postmortem proteolysis of troponin T (TnT)(*P*>0.05). WBSF was not different at 1 and 7 d (*P*>0.05), but was less in steaks from normallychilled carcasses at 14 d (*P*=0.0144). Further data analysis indicated a strong, negative correlation between 14 d WBSF and the change in pH from 6 to 12 hr postmortem (Figure 3, r = -0.8105, *P*=0.0004). These results were utilized to design the methodology for objective 2, where isolated myofibrils were subjected to μ -calpain digestion at 4 or 22°C with either a fast or slow initial pH decline. Digests were evaluated for pH, µ-calpain activity, and intact TnT degradation. Digestions with a fast initial pH decline had lower pH values in the early time points of the incubation (*P*<0.05). No differences were detected in µcalpain activity or in the degradation of intact TnT (*P*>0.05) between the fast and slow pH decline treatments. Meanwhile, a temperature x time interaction was revealed in μ -calpain activity and in the degradation of intact TnT (*P*<0.05). Additionally, intact TnT and µ-calpain activity decreased over time (*P*<0.05) while warmer digestions resulted in a tendency for reduced µ-calpain activity (*P*=0.0854) and a significant reduction in intact TnT (*P*=0.0105).

INTRODUCTION

Postmortem proteolysis (also known as the 'aging' process) is generally accepted to be the factor resulting in the improvement in tenderness associated with meat stored at refrigerated temperatures over time. This improvement in tenderness is due to a loss of muscle cell structure caused by enzymes known as proteases, which specifically degrade structural proteins to cause an overall weakening of muscle structure. This weakened structure requires less force to shear and ultimately results in a more

 1 This research was funded in part by a grant from the South Dakota Beef Industry Council. Salaries and research support also provided by state and federal funds appropriated to South Dakota State University.

tender product for consumers. However, debate continues concerning the activity of these proteases under early postmortem conditions. Numerous studies (Goll et al., 1992; Koohmaraie, 1988; Koohmaraie et al., 1996) provide compelling evidence that postmortem tenderization of beef is primarily modulated by the calpain protease system, specifically μ -calpain. These highly abundant enzymes are calcium dependent, have access to their substrates postmortem, require no ATP for activation, and have the ability to reproduce postmortem changes *in vitro* (Huff-Lonergan et al., 1996; Koohmaraie, 1988). Despite this convincing evidence, calpains may lack the ability to function under postmortem conditions (Boehm et al., 1998). In particular, *in vitro* proteolytic assays show that the activity of μ–calpain decreases rapidly during storage.

Much of the controversy surrounding the postmortem influence of μ -calpain is related to the ability of this enzyme to function at temperature and pH conditions found in early postmortem muscle. Prior to death, living muscle pH is approximately 7.4 and ultimately declines to a pH of 5.3 to 5.6 over a 24-hour period (Aberle et al., 2001). In addition, muscle temperature declines from body temperature to approximately 4 \degree C over this same time period. Optimal pH and temperature conditions for μ -calpain are similar to those found in living muscle tissue; therefore, we evaluated delayed chilling to optimize conditions for µ-calpain activity. Delayed chilling is a nontraditional method where carcasses are held in an unchilled environment for a period of time prior to chilling (Smulders et al., 1992) increasing the potential to improve beef tenderness (Fields et al., 1976; Yu et al., 2008). Therefore, the objective of this study was twofold: 1) Determine the conditions (temperature, pH) that exist in early postmortem muscle of normally-chilled and delay-chilled beef carcasses to provide a model for *in vitro* work, and 2) Determine the mechanism by which specific early postmortem temperature/pH conditions found in normally chilled and delay chilled muscle influence the enzymes that regulate the aging process *in vitro*.

MATERIALS AND METHODS

Experiment 1 – Sample Collection

Seven market weight calves (HCW 848 ± 17.5 lb) were harvested at the South Dakota State University Meat Lab using standard harvesting procedures. After final inspection (~1.25 hr post-exsanguination), sides were divided into different treatments: the right side of each carcass was placed into a cooler (normal chilling; 2.3°C) while each left side was subjected to ambient temperature (delay chilling; 22.6°C) for an additional 4.75 hr and then allowed to normally chill. Muscle temperature was recorded every minute using a Temprecord Multitrip temperature recorder (Sensitech Inc., Beverly, MA), placed in the center of the *longissimus dorsi* at the 13th rib. From each side (starting at the 13th rib and moving posterior), a 30-g, cross-sectional sample of the *longissimus dorsi* was removed at 0.75, 1.5, 3, 6, 9, 12, 24, 48, 72, 168, and 336 hr post-exsanguination. This sample was divided for different measurements: the medial portion was used for determination of pH and the central portion was used for the evaluation of proteolysis. Samples for proteolysis analysis were vacuum packaged and immediately frozen at -20°C, whereas pH samples were immediately evaluated. Three locations (rump, flank, and brisket) were swabbed on each carcass side for analysis of microbial counts. Additionally, at 24 hr postmortem, a sample from the *longissimus dorsi* was removed and frozen at -20°C for later analysis of sarcomere length. Myofibrils from each sample were purified according to a procedure by Weaver et al. (2008) and sarcomere length was determined using a procedure by Mohrhauser et al. (2011). To compare the treatment effect on aging, three 2.54-cm steaks were removed from the $9th - 12th$ rib section for Warner-Bratzler Shear Force (**WBSF**) analysis and aged for 1, 7, and 14 d. Steaks for WBSF were vacuum packaged and stored at 4°C until completion of the aging periods, and frozen at -20°C until further analysis.

Experiment 2 – Digestion of Myofibrils with µ-calpain

Activity of µ-calpain was evaluated during the digestion of already purified myofibrils with differing rates of pH decline to mimic conditions found in muscle during the conversion of muscle to meat. Due to limitations in creating temperature declines that mimic early postmortem conditions, digestions occurred at either 4°C or 22°C with either a fast or slow pH decline during the first hour of the reaction. The desired pH at one hour for digestions subjected to a fast pH decline was 5.6-5.8 while the desired pH for the slow pH decline was 6.2-6.4. Myofibrils were digested by µ-calpain according to a modified procedure of Huff-Lonergan et al. (1996). Eight mL of glycerinated myofibrils were centrifuged at 3100 x g for 6 min at 4°C and washed with 4 mL of 5 mM Tris-HCl (pH 8.0). The samples were then centrifuged at 3100 x g for 6 min at 4°C and washed with 4 mL of 50 mM MES-Tris, (pH 6.8). Samples were again centrifuged but at 1100 x g for 6 min at 4˚C and washed with 4 mL of 50 mM MES-Tris, (pH 6.8). Finally, samples were centrifuged again at 1100 x g for 6 min at 4˚C and resuspended with 4 mL of 50 mM MES-Tris, (pH 6.8). Protein concentration was then determined using the Biuret procedure and 10 mL of myofibrils were placed in SnakeSkin dialysis tubing, (10K MWCO, Thermo Fisher Scientific, Rockford, IL) with a protein concentration of 4 mg/mL by adding 50 mM MES-Tris, (pH 6.8). One-hundred μ M CaCl₂ and 15 mM 2-mercaptoethanol were added to each reaction tube. The digestion reaction was started by adding µ-calpain to myofibrils (0.45 units/mL of myofibrils) [µ-calpain (48.2 U/mg protein) purified according to the procedure of Thompson and Goll (2000) with minor modifications as described by Maddock et al. (2005)]. Aliquots were removed at desired time points (0, 0.17, 0.33, 0.5, 1, 3, 24, and 72 hr) and placed into 0.5 volumes and 0.1 volumes of pyronin Y sample buffer [3 mM EDTA, 3% (wt/vol) SDS, 30% (vol/vol) glycerol, 0.003% pyronin Y, 30 mM Tris-HCl, pH 8.0] and 2-mercaptoethanol, respectively. Samples were denatured at 100˚C for 5 min, cooled, and stored at -20˚C. Samples were then analyzed for troponin T (TnT) degradation using the gel electrophoresis, transfer conditions, and western blotting techniques listed previously. The digestion pH was adjusted by placing the dialysis tubing containing the reaction into a 50 mM MES, 100 μ M CaCl₂ and 15 mM 2-mercaptoethanol buffer containing either lactic acid or Tris-base. Calpain activity was also evaluated in this study using a procedure from Koohmaraie (1990).

RESULTS AND DISCUSSION

Experiment 1

As expected, carcass sides subjected to delay chilling showed a slower temperature decline (Figure 1a). Delay-chilled carcasses had higher internal temperatures at 3, 6, 9, and 12 hr (*P*<0.05). At 24 hr, temperatures were did not differ between the two treatments (*P*=0.9409). As a result of the different chilling methods, differences in pH decline were also found (Figure 1b). Delay-chilled carcasses demonstrated a faster pH decline resulting in lower pH values at 6, 12, and 24 hr (*P*<0.05).

Steaks aged for 1 and 7 d showed in no differences in WBSF as a result of chilling methods (*P*>0.05; Table 1). However, when aged for 14 d, steaks from normally-chilled carcasses required less force to shear, indicating that they were more tender than steaks from delay-chilled carcasses (*P*=0.0144). Most interestingly, a strong, negative correlation between 14 d WBSF and the change in pH from 6 to 12 hr postmortem (Figure 2, r = -0.8105, *P*=0.0004) was observed. This relationship indicates that a large change in pH between 6 and 12 hr postmortem may result in a lower 14 d WBSF, or more tender meat.

Figure 1. a) Postmortem *longissimus dorsi* temperature declines from carcasses normally and delayed-chilled. Temperatures were significantly different at 3, 6, 9, and 12 hr (*P*<0.05). **b)** Postmortem pH decline of normally chilled and delay-chilled beef carcasses. Measurements of pH were significantly different at 6, 12, and 24 hr (*P*<0.05).

Variable	Normal	Delayed	SEM	P-value
1-d WBSF, kga	6.92	6.34	0.45	0.1869
7-d WBSF, kg ^a	4.36	4.76	0.50	0.2659
14-d WBSF, kga	3.79	4.56	0.31	0.0144
Sarcomere length, µm	1.77	1.82	0.04	0.3336

Table 1. Least square means of Warner-Bratzler Shear Force and sarcomere length of *longissimus dorsi* from normal- and delay-chilled carcasses.

^aWBSF = Warner-Bratzler Shear Force

Figure 2. Correlation of 14-d Warner-Bratzler Shear Force (WBSF) to the change in pH occurring from 6 to 12 hr postmortem (r = -0.8105, *P* = 0.0004).

Despite this, proteolysis of TnT was not different between treatments at all time points (Figure 3, *P*>0.05). Although the breakdown of TnT is generally regarded as a good indicator of protease activity and postmortem proteolysis, it remains uncertain whether breakdown of this regulatory protein aids in the improvement of tenderness. Therefore, other myofibrillar proteins may be broken down at different rates due to varied postmortem conditions.

Figure 3. Degradation of troponin T due to normal and delayed chilling methods. Abundance of intact proteins is expressed relative to the abundance of troponin T at 0.75 hr postmortem (*P*>0.05).

Some research has reported differences in sarcomere length due to delayed chilling; however, no differences in sarcomere length were discovered in this study (*P*=0.3336; Table 1). Additionally, no differences were reported in total aerobic plate counts (*P*>0.05; Table 2). However, it should be noted that swabs of delayed-chilled carcasses had numerically higher microbial counts at 8 d postmortem.

Variable	Normal	Delayed	SEM	P-value
1-d rump, $CFUa$	1741	1000	439	0.0890
8-d rump, CFU ^a	1109	30394	21173	0.3658
1-d flank, CFU ^a	1227	1264	489	0.9585
8-d flank, CFU ^a	3637	241140	149300	0.3036
1-d brisket, CFU ^a	2297	2486	622	0.8120
8-d brisket, CFU ^a	44536	138811	72530	0.3935

Table 2. Least square means of total aerobic microbial counts from swabs of rump, flank, and brisket locations at 1 and 8 d from normal and delay-chilled carcasses.

^aCFU = colony forming units

Experiment 2

The pH decline of the digestions performed at 4°C can be observed in Figure 4a. As designed, digestion conditions resulted in differing pH at various time points. Compared to the incubations subjected to a slow pH decline, digestion reactions subjected to a fast pH decline had significantly lower pH values at 0.17, 0.33, and 1 hr (*P*<0.05), while pH values were not different at 3, 24, and 72 hr (*P*>0.05). Similarly, the fast and slow initial pH declines of the digestions performed at 22°C were different (Figure 4b). Digestion reactions subjected to a fast pH decline had significantly lower pH values at 0.5 and 1 hr (*P*<0.05), while pH values tended to be lower at 0.17, 0.33, and 3 hr (*P*<0.10).

Figure 4. a) pH of *in vitro* digestions of purified myofibrils subjected to different pH declines at 4°C. Measurements of pH were significantly different at 0.17, 0.33, and 1 hr (*P*<0.05). **b)** pH of *in vitro* digestions of purified myofibrils subjected to different pH declines at 22°C. Measurements of pH were significantly different at 0.5 and 1 hr (*P*<0.05) and tended to be different at 0.17, 0.33, and 3 hr (*P*<0.10).

For digestions performed at 4°C and 22°C, remaining µ-calpain activity as a percentage of activity at 0 hr can be found in Figure 5. Results indicated a temperature x time interaction (*P*=0.0066). Assayed activity of µcalpain was lower in myofibril digestions performed at 22°C compared to digestions performed at 4°C at 0.17, 0.33, 1, and 3 hr (P<0.05), and tended to be lower at 0.5 hr (*P*=0.0939). However, at 72 hr, µ-calpain activity was lower in digestions performed at 4°C compared to those at 22°C (*P*=0.0306). Activity of µcalpain tended to be lower when incubated at 22°C (*P*=0.0854). Additionally, µ-calpain activity decreased over time as expected (*P*<0.0001), while the initial rate of pH decline had no effect on µ-calpain activity (*P*=0.8632). Overall, the influence of temperature on µ-calpain activity in this study was expected as room temperature (22°C) is a more optimal condition for µ-calpain activity. At this more ideal temperature, µcalpain has been shown to be more active, yet assayed results indicate lower activities in this study, probably due to the acceleration of autolysis, resulting in more self-destruction of the enzyme.

Figure 5. Percentage of initial µ-calpain activity remaining at given time points during *in vitro* digestions of purified myofibrils subjected to fast or slow initial pH declines and temperatures of 4°C or 22°C. Different rates of pH decline did not change μ calpain activity at these specific time points (*P*>0.05). A significant temperature x time interaction was indicated to influence µ-calpain activity (*P*=0.0066). Calpain activity was significantly influenced by temperature at 0.17, 0.33, 1, 3, and 72 hr (*P*<0.05), while a tendecy was seen at 0.5 hr (*P*=0.0939).

Just as in experiment 1, degradation of the myofibrillar protein TnT was evaluated in experiment 2 in order to determine if different combinations of temperature and rates of pH decline affected protein degradation. A chart illustrating the breakdown of intact TnT as a result of different pH declines at 4°C and 22°C can be seen in Figure 6. Representative western blot images for TnT disappearance due to differing pH declines at 4°C and 22°C are found in Figures 7 and 8, respectively. Similar to the µ-calpain activity results, a temperature x time interaction was revealed in the proteolysis of TnT (*P*=0.0073). Less intact TnT was found in myofibrils digested at 22°C compared to digestions at 4°C at 0.33, 0.5, 1, 3, 24, and 72 hr (*P*<0.05). As expected, intact TnT decreased over time (*P*<0.0001). Additionally, warmer temperatures resulted in a greater disappearance of intact TnT of isolated myofibrils incubated with µ-calpain (*P*=0.0105). Rate of initial pH decline had no significant impact on the degradation of intact TnT (*P*=0.4461); however, Western blots for TnT appeared to show slightly greater amounts of TnT degradation products for myofibrils subjected to a fast initial pH decline, although this was not measured (Figure 8).

Figure 6. *In vitro* µ-calpain degradation of troponin T subjected to slow pH or fast pH decline within the first hour of enzymatic digestions at 4°C or 22°C. Abundance of intact proteins is expressed relative to the abundance of troponin T at 0 hr postmortem. Different rates of pH decline did not change the amount of intact troponin T at these specific time points (*P*>0.05). A temperature x time significant interaction was indicated to influence intact troponin T (*P*=0.0073). The amount of intact troponin T was significantly influenced by temperature at 0.33, 0.5, 1, 3, 24, and 72 hr (*P*<0.05).

Figure 7. Representative western blot image for *in vitro* µ-calpain degradation of troponin T subjected to an initial fast pH or slow pH decline at 4°C.

Figure 8. Western blot image for *in vitro* µ-calpain degradation of troponin T subjected to an initial fast pH or slow pH decline at 22°C.

IMPLICATIONS

This study allowed for the evaluation of the relationship of temperature and pH with meat tenderness and provided a further understanding of the aging process of beef. Additionally, an *in vitro* procedure was developed to more closely mimic postmortem conditions in beef carcasses. This study indicates that the delayed chilling of beef carcasses did not improve meat tenderness, with no improvements in postmortem protein degradation. At the same time, it appears that carcasses subjected to delay chilling may have the potential to allow for more aerobic bacteria growth during postmortem storage, which could ultimately be detrimental to maintaining and improving food safety. Unfortunately, more questions remain concerning the relationship of early postmortem conditions and meat tenderness, especially regarding the enzyme responsible for meat aging, μ -calpain.

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The influence of caspase-3 on the calpain enzyme system during meat aging[1](#page-66-0)

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SUMMARY

Tenderness is a key component of palatability, which influences consumers' perception of meat quality. There are a variety of factors that contribute to variations in tenderness, including postmortem proteolysis. A more complete understanding of this biological mechanism regulating tenderness is needed to ensure consistently tender beef. Numerous reports indicate µ-calpain is primarily responsible for the degradation of proteins postmortem. Additionally, it has been shown that caspase-3 can cleave calpastatin, the inhibitor of µ-calpain. Therefore, the objective of this study was to determine if *in vitro* degradation of calpastatin by caspase-3 can enhance the postmortem breakdown of myofibrillar proteins by µ-calpain. Bovine *semitendinosus* muscles were excised from two carcasses 20 min postmortem. Muscle strips were dissected from the *semitendinosus*, restrained to maintain length, and placed in a neutral buffer containing protease inhibitors. Upon rigor completion, myofibrils were isolated from each strip and sarcomere length was determined. Samples with similar sarcomere lengths were selected to minimize the effect of sarcomere length on proteolysis. Myofibrils were then incubated at 22°C with µ-calpain, µ-calpain + calpastatin, caspase-3 + calpastatin, or µ-calpain + caspase-3 + calpastatin for 0.25, 1, 3, 24, 48, or 72 hr at a pH of 6.8. Proteolysis of troponin T and calpastatin was evaluated using SDS-PAGE and western blotting techniques. Analysis of western blots confirmed significant degradation of calpastatin by caspase-3. Additionally, western blots revealed intact calpastatin disappeared rapidly due to digestion by μ -calpain. While caspase-3 did not significantly degrade troponin T, all µ-calpain digestion treatments resulted in substantial troponin T breakdown. Degradation of troponin T did not differ between the μ -calpain + calpastatin and μ -calpain + caspase-3 + calpastatin digestions. Results of this study indicate caspase-3 cleavage of calpastatin does not enhance *in vitro* degradation of myofibrillar proteins by µ-calpain.

INTRODUCTION

Tenderness is one of the key factors affecting a consumer's perception of beef palatability. However, due to the variety of factors that contribute to the development of this attribute, consistently tender beef has remained elusive (Koohmarie, 1995; Brooks et al., 1998). After rigor mortis is complete, specific muscle proteins are degraded by endogenous enzymes. This breakdown results in the tenderization of meat during storage (Wheeler and Koohmarie, 1994). Unfortunately, the enzyme systems responsible for this degradation and subsequent improvement in tenderness continue to be under investigation. Multiple reports indicate μ -calpain is primarily responsible for the degradation of proteins postmortem (Taylor et al., 1995; Geesink et al., 2006; Mohrhauser et al., 2011). The caspase system has also been investigated due to its role in programmed cell death (Orlowski, 1999, Goll et al., 2008, Bernassola et al., 2010). However, research has provided little evidence of the involvement of the

 1 This research was funded in part by a grant from the South Dakota State University Griffith Research Award. Salaries and research support also provided by state and federal funds appropriated to South Dakota State University.

caspase enzymes in the direct breakdown of structural proteins of postmortem bovine skeletal muscle (Kemp et al., 2009; Mohrhauser et al., 2011). Still, caspase-3 may serve another role as it has been reported that caspase-3 can cleave calpastatin, the inhibitor of μ -calpain (Wang et al., 1998; Kemp et al., 2009). Thus, preceded by the hypothesis that caspase-3 breakdown of calpastatin can enhance μ calpain activity and indirectly improve tenderness, the objective of this study was to determine if *in vitro* degradation of calpastatin by caspase-3 can enhance the postmortem breakdown of myofibrillar proteins by µ-calpain.

MATERIALS AND METHODS

Two A maturity steers were slaughtered at the South Dakota State University Meat Laboratory using standard procedures. The semitendinosus from the left side of each carcass was removed approximately 20 min postmortem. A 10-g sample from each excised semitendinosus was vacuum packaged and frozen at −20°C to serve as a time 0 sample. Muscle strips (1 cm wide × 25 cm long) were then excised from the superficial portion of the muscle parallel to muscle fiber orientation and attached to wooden applicator sticks to maintain sarcomere length. The muscle samples were then placed in a neutral rigor buffer containing protease inhibitors to inhibit endogenous proteases. Samples were stored at 4°C overnight with constant stirring. Upon completion of rigor, myofibrils were isolated from semitendinosus muscle strips according to the procedure by Weaver et al. (2008) and sarcomere length determination was completed following the procedure of Mohrhauser et al. (2011).

Myofibrils from samples with similar sarcomere lengths were assigned to 1 of 4 digestion protocols: 1) digestion by μ -calpain, 2) μ -calpain + calpastatin, 3) caspase-3 + calpastatin, or 4) μ -calpain + caspase-3 + calpastatin. All treatments were subjected to enzymatic digestion at room temperature (~22°C) using a modified procedure from Weaver et al. (2009). Following digestion samples were subjected to SDS-PAGE and western blotting to visualize the degradation of calpastatin and TnT.

RESULTS AND DISCUSSION

Calpastatin is a four-domain, specific inhibitor of the calpain enzymes that can bind calpain at each domain. Taylor et al. (1995) indicated µ-calpain and calpastatin are colocalized in postmortem muscle; thus, it could be conceived that any µ-calpain activity 24 hours postmortem may be completely inhibited by calpastatin (Boehm et al., 1998). Therefore, it has been suggested that proteases other than µcalpain could play a role in meat tenderization. Previous research has shown the ability of caspase-3 to cleave calpastatin (Wang et al., 1998; Kemp et al., 2009). With this premise, it is postulated that caspase-3 could play an indirect role in the improvement of meat aging by enhancing the activity of μ calpain. Results from this study validated the breakdown of calpastatin by caspase-3 (Figure 1 and 2) as there was an obvious, significant difference between the relative abundance of intact calpastatin between 0 and 72 hr of enzymatic digestion (p < 0.0001). However, this rate of calpastatin degradation appears trivial when compared to μ -calpain's ability to degrade calpastatin (Figure 2). After only 0.25 hr, minimal amounts of intact calpastatin remained in the µ-calpain + calpastatin enzymatic digestion, while virtually all calpastatin had been degraded by 1 hr (Figure 2). These figures are negligibly different in the µ-calpain + caspase-3 + calpastatin digestion as calpastatin degradation rates were similar between the µ-calpain + calpastatin and µ-calpain + caspase-3 + calpastatin treatments (*P*>0.05).

Figure 1. Western blots, prepared from 10% polyacrylamide resolving gels transferred to a polyvinylidene difluoride membrane of isolated myofibrils from bovine *semitendinosus* digested with caspase-3 + calpastatin for 0, 0.25, 1, 3, 24, 48, and 72. Blots were labeled with immunoreactive anti-calpastatin (Domain IV, 1F7E3D10, Calbiochem, Billerica, MA). Time is indicated at the bottom of the blot. Arrows indicate molecular weight of standards. Intact = location of intact calpastatin.

Figure 2. *In vitro* degradation of calpastatin by µ-calpain+calpastatin, caspase-3+calpastatin, and µ-calpain+caspase-3+calpastatin. Abundance of intact calpastatin is expressed relative to the abundance of intact calpastatin at 0 hr.

Troponin T (TnT) has been the hallmark protein evaluated in postmortem proteolysis as its degradation appears to be an excellent indicator of the extent of protein degradation in myofibrils during aging (Koohmaraie, 1994). Although it is questionable whether TnT degradation has a direct effect on meat tenderness due to its regulatory function, the disappearance of intact TnT and appearance of 28-32 kDa degradation products are the most noticeable changes that take place during postmortem aging as measured by Western blots (Koohmaraie, 1994). Figure 3 shows representative images of Western blots labeled with a monoclonal antibody against TnT. Visual assessment of the blots reveal a qualitative decrease of intact TnT (bands 1-3) over time in myofibrils subjected to incubations with µ-calpain (Figure 3a). Meanwhile, visual evaluation of Western blots for the caspase-3 + calpastatin digestion indicate the slight appearance of TnT degradation product at 48 and 72 hr (Figure 3b, bands 4-5). However, no significant differences in intact TnT were indicated in this treatment (*P*>0.05; Figure 4). This coincides

with previous research showing little involvement by caspase-3 in the direct postmortem degradation of structural proteins (Mohrhauser et al., 2011; Kemp et al., 2009).

a) μ -Calpain + Calpastatin μ -Calpain+Caspase-3+Calpastatin

b) Caspase-3 + Calpastatin

Figure 3. Western blots, prepared from 15% polyacrylamide resolving gels transferred to a polyvinylidene difluoride membrane of isolated myofibrils from bovine *semitendinosus* digested for 0, 0.25, 1, 3, 24, 48, and 72 hr with a) µcalpain + calpastatin and μ -calpain + caspase-3 + calpastatin, and b) caspase-3 + calpastatin. Blots were labeled with immunoreactive anti-troponin T (JLT-12, Sigma-Aldrich, St. Louis, MO). Time is indicated at the bottom of the blots. Arrows indicate molecular weight of immunoreactive bands. Bands 1, 2, and 3 indicate the location of intact troponin T.

These results were verified through quantification as can be seen in Figure 4. As anticipated, μ -calpain rapidly degraded TnT over time (*P*<0.05; Figure 4). This degradation was slowed considerably when myofibrils were digested with µ-calpain + calpastatin (*P*<0.05; Figure 4). Thus, although physiological activity ratios of *μ*-calpain:calpastatin are typically 1:4 in beef, 1:2.5 in lamb, and 1:1.5 in pork (Ouali and Talmant, 1990; Koohmaraie et al., 1991), this study found that utilizing a lower amount of calpastatin *in vitro* produces desired inhibiting effects on µ-calpain. Unfortunately, the relative amount of intact TnT remaining for the μ -calpain + calpastatin digestion compared to the μ -calpain + caspase-3 + calpastatin digestion were not different throughout the course of the enzymatic incubation (*P*>0.05), although the addition of caspase-3 resulted in a lower numerical amount of intact TnT at several time points (Figure 3 and 4).

Figure 4. *In vitro* degradation of troponin T by µ-calpain, µ-calpain+calpastatin, caspase-3+calpastatin, and µcalpain+caspase-3+calpastatin. Abundance of intact troponin T is expressed relative to the abundance of intact troponin T at 0 hr.

IMPLICATIONS

In conclusion, this study confirms that although both caspase-3 and μ -calpain cleave the inhibitor of μ calpain, calpastatin, it provides support to previous research that caspase-3 is not significant in the direct breakdown of myofibrillar proteins, and that µ-calpain should be considered the primary protease responsible for the degradation of these key proteins during beef aging. Finally, this study provides evidence that caspase-3 does not enhance *in vitro* myofibril degradation by µ-calpain by significantly degrading calpastatin.

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BEEF REPORT 2012-11

SDSU Cow/Calf Teaching and Research Unit[1](#page-72-0)

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SUMMARY

The SDSU Cow/Calf Unit (CCU) is a multi-purpose facility that provides resources for Animal Science courses and research projects. Cattle produced at the facility are also utilized by Little International, Block and Bridle, and livestock judging teams. The facility is managed by Kevin Vander Wal and generally employs 4 to 5 undergraduate students.

BREEDING PROGRAM

Although the CCU has a multi-purpose mission, the breeding program is primarily centered on the production of bulls and females that can be used for teaching purposes and sold to the general public. Artificial insemination is used extensively. The program primarily utilizes proven sires; however, each year a percentage of the females are bred to younger, lower accuracy sires. The objective of the breeding program is to produce docile cattle that have excellent calving ease, growth, and carcass characteristics. Average expected progeny differences of the cows, heifers, and AI sires used in 2011 are included in Tables 1 and 2.

Table 1. Average expected progeny differences of Angus cows, heifers, and AI sires used in 2011.

 a CED = calving ease direct; BW = birth weight; WW = weaning weight; YW = yearling weight; SC = scrotal circumference; Milk = maternal milk; Marb = marbling score; REA = ribeye area; Doc = docility ^b $\mathbb{S}W$ = wean value; $\mathbb{S}B$ = beef value

Table 2. Average expected progeny differences of SimAngus cows, heifers, and AI sires used in 2011.

	Expected progeny differences ^a								Indexes'	
	СE	BW	ww	YW	MLK	Marb	REA	API		
Cows	12.6	-2.7	23		4.8	0.59	0.17	130	69	
Heifers	13.9	-3.1	23	58	5.6	0.60	0.35	141		
AI sires		-2.0	46	85		0.47	0.60	142	82	

 a CE = calving ease; BW = birth weight; WW = weaning weight; YW = yearling weight; MLK = milk; Marb = marbling score; REA = ribeye area

 P^b API = all-purpose index; TI = terminal index

 1 The authors would like to acknowledge Pfizer Animal Health for product donations (Eazi-Breed CIDRs, Lutalyse, and Factrel) toward the synchronization research projects. Salaries and research support also provided by state and federal funds appropriated to South Dakota State University.

REPRODUCTIVE PERFORMANCE

In 2011, 107 Angus and 45 SimAngus females were bred. All of the females are bred at least one time via artificial insemination (AI) and followed by clean-up bulls. The majority of both the Angus and SimAngus cows were part of a larger research project with Kansas State University and the University if Florida to compare synchronization strategies. Cows were synchronized using the 7-day CO-Synch + CIDR protocol (Figure 1) with (n = 52) or without (n = 52) an injection of prostaglandin $f_{2\alpha}$ (Lutalyse) 3 days prior to insertion of the CIDR. All heifers ($n = 45$) were also included in a research project to compare the 5-day (Figure 2) and the 6-day CO-Synch + CIDR fixed-time AI protocols (Figure 3).

Figure 1. 7-day CO-Synch + CIDR synchronization protocol used on cows during the 2011 breeding season. Cows either did (n = 52) or did not (n = 53) receive an injection of prostaglandin $f_{2\alpha}$ (Lutalyse) 3 days prior to insertion of the CIDR.

Figure 2. PG 6-day CIDR fixed-time AI synchronization protocol for beef heifers.

Figure 3. 5-day CO-Synch + CIDR fixed-time AI synchronization protocol for beef heifers.

Regardless of treatment, the cows achieved an overall 62.6% first service conception rate (63% and 60%, for with and without prostaglandin $f_{2\alpha}$, respectively) to timed AI and a 100% overall conception rate. Among the heifers pregnancy rates were greater (*P* < 0.01) for heifers receiving the PG 6-d CIDR (64%) compared to the 5-d CIDR (42%). Overall heifers achieved a 66.7% first service conception rate to timed AI and a 95.6% overall conception rate.

SALES

Each April, the SDSU Seedstock Merchandising class coordinates an annual bull sale at the CCU. The sale is designed to be a learning experience for the students and they are responsible for advertising, promotional videos, developing the sale catalog, and customer service. In 2012, the bull sale attracted

customers from SD, IA, MN, KS, NE and AL. In addition to the 17 Angus and 10 SimAngus yearling bulls in the sale, a special female offering was included that allowed the buyer to have choice of any cow/calf pair from the SDSU herd and the option to take a second pair for 75% of their initial bid. Results of the sale are presented in Table 3.

Table 3. Results from 2012 annual bull sale.

*One special lot was offered where the buyer would have choice of the pairs in the pasture and the option to take a second pair for 75% of their original bid.

In addition to the annual bull sale, a small group of bred females are offered to the general public in a phone auction each fall. In the fall of 2011, 32 bred females were sold in a fall female sale for an average of \$1770 per head (Table 4).

NEW FACILITY

The current CCU is nearly 60 years old and, while it is a functional facility for managing a cowherd, it has significant limitations as a teaching and research facility. One of the components of the Land-Grant mission is teaching. Our objective is to provide our students with the best education possible and a large component of educational process is experiential learning…learning by doing. The current facility is not well equipped to provide a positive learning experience to students. A second component of the Land-Grant mission is research. Conducting research that answers production related questions and provides new technology to producers is essential to the long-term viability of the beef industry in South Dakota. Other than a small group of replicated pastures, the current CCU is not equipped to support research. The final component of the Land-Grant mission is Extension and outreach. Effective transfer of new knowledge and technology from the university to end users allows for the most current research findings to be adopted by the industry. Our current facility is not conducive to utilization by Extension personnel for meetings or demonstrations.

As such, for several years an effort has been underway to develop a new cow-calf facility. In 2012, approval was received from the South Dakota Board of Regents and Legislature to begin the design process. A design team has been selected and the design and fundraising processes are ongoing. If you would like to receive more information on the new facility or if you are interested in contributing to the project, please contact Dr. Cody Wright (cody.wright@sdstate.edu; 605-688-5448) or Mike Barber at the SDSU foundation (mike.barber@sdsufoundation.org; 605-697-7475).

BEEF REPORT 2012-13

Cottonwood and Antelope Range and Livestock Research Stations Unit Report[1](#page-75-0)

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SUMMARY

The Cottonwood and Antelope Range and Livestock Research Stations, located in western South Dakota, are used for research projects focused on the needs of range livestock producers in that region. The stations are comprised primarily of native rangeland that is grazed by cattle at both stations, and also by sheep at Antelope. The philosophy of the research efforts has been focused on conducting applied research to solve problems and address rangeland and livestock management opportunities relevant to the livestock producers and land managers of the region.

INTRODUCTION

These Agricultural Experiment Stations conduct natural resource research, including range management and livestock research applicable to the semiarid, rangeland environment of western South Dakota and the Northern Great Plains.

The Cottonwood Station, located near Philip, comprises about 2640 acres of rangeland. It has a 12-pen feedlot in addition to facilities for grazing cattle research. It has a large hoop barn structure that is used for indoor educational activities. Cottonwood will soon host a new office complex that will have a lab, commodity storage, and shop in addition to grinding and drying rooms for sample preparation. Research at the Cottonwood Station is currently focusing on replacement heifer development and management of first calf heifers. The combination of rangeland pastures and feedlot facilities at the Cottonwood Station allows flexibility for a variety of projects to address the complex nutritional demands of maintenance, reproduction, and growth that challenge the productivity of heifers and young, growing cows.

The Antelope Station, located near Buffalo, comprises about 8165 acres of rangeland. Livestock at this station include both sheep and cattle. The sheep on this station are owned by NDSU and provide a basis for joint research opportunities. The carrying capacity at this station is for 300-plus cows. The larger number of mature cows on the ranch-scale sized area of land at the Antelope Station provides the opportunity to evaluate alternative management strategies in a total production-system setting. The sheep at the Antelope Station have been utilized in a similar philosophy to evaluate the influence of management alternatives in a system setting.

The size and location of these stations have proven valuable for a variety of wildlife and habitat research protocols.

 $¹$ Salaries and research support provided by state and federal funds appropriated to South Dakota State University.</sup>

² Director, West River Ag Center

CURRENT RESEARCH PROJECTS

Research Activities conducted during 2010/12 at each station include:

Cottonwood

- 1. Impact of maternal nutrition on expression of genes regulating offspring growth, carcass composition and meat quality. A project funded by USDA-AFRI that focuses on the effects of maternal nutrient restriction on offspring growth, performance and carcass characteristics. Investigators are A.D. Weaver (PI), K.R. Underwood, A.E. Wertz-Lutz, R.H. Pritchard, and J.M. Reecy.
- 2. Impact of maternal nutrient restriction on phenotypic expression in different leptin genotypes of offspring and meat quality. A project funded by the South Dakota Beef Industry Council that focuses on the potential interactions between leptin genotype and management and resultant fat deposition in cattle. Investigators are A.D. Weaver (PI) K.R. Underwood, A.E. Wertz-Lutz, R.H. Pritchard.
- 3. Influence of propionate salt levels on young cow reproductive performance: Funded through USDA Hatch Formula Grant. Investigators are J.A. Walker, G.A. Perry and K.C. Olson.
- 4. De-oiled distillers grains as a protein supplement for cows: A project funded by MBI Consortium to evaluate the utilization of a novel dried distillers grain co-product with much of the corn oil removed so the crude protein content is similar to soybean meal. Investigators are Ken Olson, Mindy Hubert, and Nikki Hojer.
- 5. Heterogeneity project: This study is evaluating the potential to utilize heavy winter Patch Grazing in mixed-grass prairie to enhance heterogeneity for wildlife habitat. Cattle behavior, cattle production, vegetation utilization, and bird and small mammal habitat attributes were measured to evaluate the benefits and/or consequences to livestock production and any benefits for bird habitat. Major participants: Pat Johnson, Kent Jensen, Ken Olson, Roger Gates, Christi Koehler, Janna Kincheloe.
- 6. Mixed-grass prairie root growth studies: This study is evaluating the production and decomposition of root biomass in mixed-grass prairie in an effort to evaluate root:shoot ratios and root turnover rates. Major participants: Sandy Smart, David Clay, Sharon Clay, Jiyul Chang, Pat Johnson.
- 7. Long-term range production and stocking rate study: A project that has continued for over 50 years to document differences in vegetation production and cattle gains associated with controlled stocking rates. The principal investigators are Pat Johnson, Roger Gates, Janna Kincheloe, and Ken Olson.
- 8. Cottonwood hosts three separate weather related stations:
	- A weather station for the SD State Climatological Office
	- A meteorological monitoring site for NOAA
	- A National Atmospheric Deposition Program collection site for precipitation chemical composition.
- 9. Hosted the research team for "Classification and Mapping of Riparian Forest along the White River in South Dakota". A project funded by a grant from South Dakota Game, Fish and Parks. The

principal investigators are Mark Dixon, Carter Johnson, Alex Cahlander-Mooers, and Alanna Robinson.

Antelope

- 1. Immunology project: Is response to a Bovine Viral Diarrhea Virus vaccine correlated with production and carcass traits in beef cattle? Investigators are Michael Gonda and Julie Walker.
- 2. Impact of programmed nutrition on offspring health, growth, performance and meat quality. A project funded by Alltech® that focuses on the influence of mineral bioavailability from conception to consumption on offspring health, growth feedlot performance and carcass characteristics. Investigators are A.D. Weaver (PI), K.R. Underwood, B. Holland, M. Gonda, C. Wright, R. Daly, and A.R. Taylor.
- 3. Influence of propionate salt levels on young cow reproductive performance: Funded through USDA Hatch Formula Grant. Investigators are J.A. Walker, G.A. Perry and K.C. Olson.
- 4. "Presynchronizing $PGF_{2\alpha}$ injection before the fixed-time artificial insemination (TAI) CO-Synch + CIDR program." This was part of a multi-state study with Kansas and Florida. Investigators: Jeff Stevenson at Kansas State University, Cliff Lamb at the University of Florida and George Perry at South Dakota State University.
- 5. Beef cattle systems-effects of early weaning and winter feeding strategy: A project funded by the Four-State Ruminant Consortium to evaluate the combined effects of weaning date (early or normal) and winter feeding strategy (limited or full) on livestock performance, rangeland forage utilization, and economic response. Investigators are Pat Johnson, Roger Gates, Ken Olson, Mary Beutler, Scott Fausti, Janna Kincheloe, Sandy Smart, George Perry, Robin Salverson, and Nikki Hojer-Kroupa.
- 6. Evaluating glycerin supplementation on reproductive performance of sheep: Funded through USDA Hatch Formula Grant. This project was collaborative effort between SDSU and NDSU, Hettinger Research Extension Center. Major participants were J.A. Walker, G.A. Perry, R. Salverson, P. Nester, C.S. Schauer, J.E. Held, and K.C. Olson.
- 7. Yellow-flowered alfalfa: A project funded by USDA-CSREES to evaluate the adaptation and value of yellow-flowered alfalfa for rangelands. Investigators include Roger Gates, Arvid Boe, Xu Lan, Pat Johnson, and Janna Kincheloe.
- 8. Antelope Station hosts two weather-related stations;
	- Meteorological Monitoring Site for NOAA
	- A weather station for the South Dakota State Climatological Office

In addition to these research activities, both stations have been used for Extension and teaching activities such as Tri-County Ag Day and Rangeland Days. Extension Field Staff training has been conducted at Antelope and Cottonwood has been used as a laboratory setting for Range 325, a course entitled Range Measurements.

- 1 Brookings: SDSU campus, Agricultural Experiment Station, Cooperative Extension Service
- 2 Beresford: Southeast South Dakota Research Farm Beef cattle nutrition Swine nutrition and management
- 3 Rapid City: West River Ag Center Professional research and Extension staff in Animal Science, Range Science, Agronomy, Horticulture, Community Development, Economics, 4-H, and Extension administration
- 4 Buffalo:Antelope Range Livestock Station Beef cattle breeding and range beef herd management Sheep nutrition, management, and breeding
- 5 Phillip: Range and Livestock Research Station Range beef nutrition and herd management Range management

These research and Extension units are geographically located in South Dakota to help solve problems, bring the results of livestock and range research to users, enhance the statewide teaching effectiveness of the Animal Science Department staff, and maintain a close and productive relationship with South Dakota producers and the agribusiness community.

The state of South Dakota is · our campus · our research lab · our classroom