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7-13-2005

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Theriogenology

Theriogenology 64 (2005) 657-678

www.journals.elsevierhealth.com/periodicals/the

## Fertility of yearling beef bulls during mating<sup>☆</sup>

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#### Abstract

Crossbred (Bos taurus) yearling beef bulls were assessed for breeding soundness and physical traits prior to multi-sire natural mating at pasture. Bulls (n = 60) were assigned to six groups of nine or 10 bulls and two bull-groups were rotated on 14-day intervals during a 63-day mating season in each breeding herd (n = 3) of 191–196 cows. The remaining bulls (n = 14) were maintained under similar environmental conditions without mating exposure. Bulls were observed during mating and assessed for breeding soundness and changes following mating. Bulls used for breeding (UFB) lost 77 kg of body weight and declined from body condition scores of 6 to 4.5, whereas bulls not used for breeding (NUB) maintained body condition scores of 6 and gained 27 kg. The UFB bulls incurred a 75% total injury rate with 63% incidence of lameness and 12% incidence of reproductive injuries, resulting in a 22% attrition rate. Only 45% were physically sound at the end of mating. Scrotal circumference declined in UFB bulls (-4.58%) and increased in NUB bulls (2.49%). From the 98% BSE-satisfactory rate (UFB) prior to breeding, only 61% were BSE-satisfactory post-breeding. The NUB bulls declined from 57 to 36% satisfactory. The BSE classification was influenced by significant increases in abnormal spermatozoa (primary and secondary), which was significantly associated with injuries incurred during mating. Group and breed differences in injury rates and BSE-status following mating were evident. Environmental conditions and mating activity influenced bull seminal quality and physical condition. Pregnancy rates in all three breeding herds (91-96%) were similar, with

<sup>&</sup>lt;sup>★</sup> A contribution of the University of Nebraska Agricultural Research Division, Lincoln, NE 68583. Journal Series No. 14511.

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insignificant differences between bull-groups; the effects of physical and reproductive changes on individual bull fertility were immeasurable.

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Keywords: Bull; Breeding soundness evaluation; Fertility; Breeding injuries

#### 1. Introduction

Yearling bulls are an important component of natural service mating in North American cattle operations and a Breeding Soundness Evaluation (BSE) is the principle method employed to assess the general reproductive potential of bulls. The BSE, when conducted within the guidelines of the Society for Theriogenology [1], assesses the current physical and reproductive status of bulls and allows for screening and selection of bulls with acceptable fertility-potential. Regarding yearling bulls, the BSE further serves to measure the pubertal advancement of the physical and reproductive systems and the attainment of characteristics considered sufficient to be classified as satisfactory potential-breeders [2]. A temporal limitation of the BSE involves the assessment of only the current status of physical, testicular and seminal characteristics; therefore, a satisfactory classification affords no implied guarantee of future reproductive performance of individual bulls [3].

Bulls from 12 to 24 months of age represent a progressive continuum of physical and sexual maturation and may be particularly vulnerable to factors that influence fertility. During natural service, the effects of extraneous influences on the fertility-associated parameters measured during a BSE, may contribute to alteration of reproductive capability and variability in reproductive performance [4]. Pregnancy rates achieved by bulls with satisfactory classification are consistently higher than untested bulls and generally higher than bulls not meeting satisfactory standards [5,6], however, the known limitations of the BSE [2,7] and the influence of environmental factors on fertility make the predictive value of the BSE towards reproductive performance of the individual bull unreliable [6,8].

Yearling beef bulls would be expected to grow in physical stature and improve in reproductive function from 12 to 24 months of age [9–13]. Testicular development is expected to continue [11,13] and seminal quality, particularly spermatozoal form and viability, is expected to improve through this period [4,11,14]. The presumption would be that fertility should improve. Three components of the BSE, i.e., scrotal circumference, spermatozoal motility and spermatozoal morphology are inter-related and positively correlated and positive changes in these parameters through puberty are associated with fertility-potential [15,16]. Young bulls attaining satisfactory-classification on a pre-breeding BSE would therefore presumptively be expected to advance in physical and reproductive capability. However, environmental and physiological stressors are known to unfavorably influence some physical and seminal characteristics [17–21]. Factors such as season, climatic temperatures, toxic principles, disease and injury, physiologic stress and nutritional imbalances have been linked with alterations in scrotal circumference and seminal quality [17,22]. The impacts appear to vary between individual bulls and generally

are temporary, although occasionally they may precipitate permanent infertility of the bull [22]. Accordingly, the reproductive soundness of bulls following a BSE should be considered to be a dynamic condition, with both favorable and unfavorable potential changes.

Previous studies have investigated the variability in fertility and reproductive performance of yearling beef bulls under natural mating conditions. Variability in fertility of bulls with satisfactory potential-breeder classification on BSE has been reported [8,23–26]. Bulls of diverse genotypes have had differences in physical and reproductive traits from 12 to 18 months of age and general improvements in fertility-potential through this period [10,27,28]. Environmental conditions can influence the differences according to breed and age of the bull. However, limited study has evaluated the physical and reproductive changes young bulls undergo through a natural mating season and the subsequent effects on fertility during mating.

Yearling beef bulls in groups of two, exposed to natural mating for 3 days, and estrussynchronized mating for 1 day, were evaluated for the effect of mating load on fertility [23]. On post-exposure observations, the bulls had body weight losses of 25–26 kg, with concurrent increases in scrotal circumference, however, seminal characteristics were not altered from pre-breeding BSE findings. In further work, mating performance of yearling beef bulls in groups of three were assessed with exposure to either natural or estrussynchronized mating for 44 days [24]. Although the bulls experienced BW losses of 30-32 kg, all bulls were classified as satisfactory on post-breeding BSE. An additional study evaluated yearling bulls of similar age, body weight, scrotal circumference and semen quality during single- and double-sire pasture matings [26]. After short, intense mating exposures and service capacity evaluations, post-breeding evaluations conducted 70-80 days following initial evaluations revealed normal increases in body weight and scrotal circumference, and no changes in seminal quality measurements. Mating activity levels did not influence physical or reproductive characteristics. Changes in physical condition of yearling bulls during mating have been confirmed, however, changes in testicular and seminal characteristics were not detected and variability in reproductive performance was not attributed to any alterations in reproductive capability.

To state that all yearling beef bulls with satisfactory classification on BSE are equally fertile throughout a mating season and will maintain reproductive soundness is tenuous and arguable. Realizing that fertility is an integration of multiple biological and behavioral components, the potential for deviation in the reproductive potential of the immature yearling bull affords possibilities for variability in performance. By observing and measuring the physical and reproductive characteristics of young beef bulls during a typical mating season, and the BSE-status at the completion of mating, knowledge regarding the potential changes in reproductive capability and subsequent variability in reproductive performance of bulls could be expanded. Furthermore, by monitoring environmental factors during mating and studying the associations of extraneous factors with the changes in physical and reproductive traits, additional information concerning the variance in reproductive performance of bulls could be provided. The objective of the study focuses on providing information applicable to explaining why all BSE-satisfactory yearling bulls do not perform equally in regards to reproductive outcomes during natural mating.

#### 2. Materials and methods

The observational study involved 74 crossbred yearling beef bulls, representing 7 *Bos taurus* sire-breed lines and either of two dam-breed lines, Hereford or Angus. The sire-breed lines (SBL) represented include: Angus, Charolais, Gelbvieh, Hereford, Limousin, Red Angus and Simmental. The bulls originated from a single beef cattle research operation and geographical location in south-central Nebraska, USA (U.S. MARC, USDA-ARS, Clay Center, NE, USA). The bulls were born in March and April of 2001, managed as a contemporary group following weaning (7–8 months of age) and through a traditional development period until the spring breeding season (June 2002). The bulls were defined as virgin yearling beef bulls at the initiation of the study (13–14 months of age). The females used for assessment of reproductive performance consisted of 2- and 3-year-old crossbred *Bos taurus* cows nursing spring-born calves (n = 581). The cows represented similar genetic compositions as the bulls and originated and were maintained as breeding populations of the research operation. Description of the female breeding populations and female fertility assessment has been previously reported [29].

Approximately 3 weeks prior to the breeding season, a Breeding Soundness Evaluation (BSE 1) was performed on all bulls, and repeated approximately 1 week prior to the breeding season on 14 individual bulls with deferred classification on the first exam. All BSE were performed and bulls classified according to the guidelines of the Society for Theriogenology [1]. Each bull was examined for soundness and health and individually identified. Examination of testicles, epdidymides, vesicle glands, ampullae, prostate and internal inguinal rings was accomplished by palpation and scrotal circumference was measured [30]. With the use of electroejaculation, the prepuce and penis were examined and semen samples were collected upon erection and ejaculation.

Semen samples were immediately evaluated for gross and progressive individual motility of spermatozoa under temperature-controlled conditions. Sperm morphology was assessed on eosin-nigrosin stained samples under phase contrast microscopy at  $\times 1000$  and proportions of normal sperm were categorized on counts of 100 spermatozoa. Further classification of abnormal spermatozoa into defects of head, acrosome, midpiece and tail [2,22] and evaluation for the presence of other abnormal seminal cells was performed. Classification of satisfactory potential-breeder was attained by achieving threshold measurements of: (1) >30 cm scrotal circumference, (2) >30% progressively motile spermatozoa, and (3) >70% morphologically normal spermatozoa [2]. In addition, bulls were to be free of faults of physical and reproductive systems. For purposes of this study, all bulls not meeting satisfactory standards were categorized as unsatisfactory. Two trained veterinarians performed the evaluations and a trained veterinary technician and one trained veterinarian performed the semen evaluations.

From the bull population, individuals (n = 57, UFB) were randomly selected by the cattle operation to meet the sire-breed line requirements of pre-established genetic mating plans and by BSE-satisfactory classification. Random, stratified assignments by sire-breed line were made into six bull-groups (GRP) of nine or 10 bulls with GRP identification as A, B, C, D, E and F. Each GRP was assigned to one of three cowherds, designated as HRD1 (n = 194), 2 (n = 191) or 3 (n = 196), at a GRP bull-to-female ratio of 1:20  $\pm$  2 (mean + S.E.M.). Each HRD had two GRP of bulls rotated on 14-day intervals through a

GRP	HRD	R	n	AN	СН	GB	НН	LM	AR	SM	BSE 1	Satis%
A	1	1	9	1	1	2	1	1	2	1	9/9	100
В	2	1	10	1	2	1	2	1	1	2	9/10	90
$C^{a}$	3	1	11	1	1	2	$1 + 1^{a}$	1	$2 + 1^{a}$	1	11/11	100
D	1	2	10	1	2	1	2	1	1	2	10/10	100
E	2	2	9	1	1	2	1	1	2	1	9/9	100
$F^a$	3	2	11	1	2	1	2	1	1	$2 + 1^{a}$	11/11	100
UFB			60	6	9	9	10	6	10	10	59/60	98
NUB			14	3	7	1	1	0	0	2	8/14	57
All bulls			74								67/74	90.5

Table 1 Bull breeding-groups (GRP) and sire-breed line (SBL) distribution

BSE 1 classification Herd and rotation assignments. GRP, bull-group; HRD, breeding herd; R, breeding rotation; Satis%, BSE satisfactory potential breeder classification.

standard 63-day mating season (early June to mid-August), thus each GRP had two distinct mating periods. Bull-groups A, B, and C in rotation 1 (R1) were exposed to females for 28 total days with a 14-day rest period, whereas bull-groups D, E, and F in rotation 2 (R2) were exposed to females for 35 total days with a 14-day rest period (Table 1). During the first mating periods for GRP C and F, 3 bulls were removed and replaced with like sire-breed bulls, therefore 60 UFB bulls were exposed to mating. The remaining bulls (n = 14, NUB) were maintained on grass pastures without mating exposure during the same time period. Mating pasture environments during the season varied between HRD and R, from native cool-season grasses, irrigated grasses and forages, and irrigated alfalfa, due to the extended low precipitation and higher than normal climatic temperatures for the season.

On Day 1 of each GRP mating period, bulls were individually weighed (non-shrunk BW), body condition score (BCS) estimated, hip height (HH) measured and scrotal circumference (SC) measured. Additionally, inspection for physical soundness was performed. Within 96-h following the last day of mating exposure for each GRP, the remaining UFB bulls (n = 57) were subjected to a BSE (BSE 2) and final BW, BCS, HH and SC were measured. BSE 2 was performed the same as previously outlined. The NUB bulls (n = 13) were similarly evaluated at the completion of the study.

The bulls were individually monitored by GRP throughout the mating season for aberrations of physical and reproductive soundness, i.e., injuries and illness. Injuries were diagnosed and categorized as musculoskeletal or reproductive by field observation and severity, duration, and prognosis determined the continuation or removal from mating exposure. A cumulative injury score was assigned as 1-point per injury incident and 3-points for removal from the mating pasture for each individual bull and the date of each injury incident plotted on a mating season time line.

Average daily climatic temperatures and daily peak temperatures, along with average relative humidity and precipitation were collected from the National Weather Service regional reporting center in Hastings, NE, USA.

Approximately 65 days following the completion of the mating season, the three HRD were palpated per rectum for pregnancy status and further definition of bull reproductive performance was assessed with calving dates and calving distributions of the females. A

<sup>&</sup>lt;sup>a</sup> Replacement bulls for injured bulls removed early during breeding season.

283-day average gestation length was utilized to estimate date of conception from calving dates.

Variables amendable to analysis of variance were subjected to a statistical software program, ANOVA-single factor test [31]. Further analysis of covariance, simple and rank correlation, and simple linear regression were performed to measure association and relationships [31]. All procedures were assessed at 0.05 critical values for the *F*-statistic. Least square means and standard errors of the mean (S.E.M.) are reported for data sets and sample comparisons. Variables with non-normal distributions and heterogeneous variances were converted to categorical data where appropriate and Chi square tests of independence and trend analysis were performed. Wilcoxon–Mann–Whitney nonparametric tests were used to compare distributions of ordinal-scale variables and Kruskal–Wallis H tests were utilized to determine significant differences of rank-transformed sample means of semen quality statistics. The software program PEPI, Version 4.0x [32] was utilized to perform nonparametric analyses. Limitations were assumed with sample groups of small and unequal sizes, such as GRP and SBL, however, mean and S.E.M. values for the variables are reported and conservative analysis of trends and variance procedures (Tukey's Studentized Range Test) were utilized.

#### 3. Results

#### 3.1. Pre-breeding observations

The BSE 1 was performed 17 and 31 days prior to the initiation of the breeding season for mating rotations R1 and R2, respectively. Final satisfactory classification was given to 67 of the 74 bulls (90.5%). To meet the sire-breed requirements for each GRP, one Limousin-sired bull with unsatisfactory classification was selected as a UFB candidate, therefore, 59 of the 60 UFB bulls (98%) began the breeding season with satisfactory potential-breeder classification. This individual was assigned to GRP B in R1; bull B6. For the NUB bulls, 8 of 14 (57%) were classified as satisfactory potential-breeders.

Age of the 74 crossbred yearling beef bulls at BSE 1 was  $428 \pm 0.8$  days (mean  $\pm$  S.E.M.) with a range of 28 days (414–442 days of age)—approximately 13.5–14.5 months of age. No differences in age were revealed between any of the bull sample-groups or sire-breed groups (P > 0.10). No significant correlations were demonstrated for age at BSE 1 with other BSE observations or physical characteristics.

Scrotal circumference (SC) for all bulls was  $36.3 \pm 0.2$  cm (range, 32.5–45.0 cm). All bulls exceeded the minimum required SC of 30 cm for bulls 12–15 months of age. No differences in SC were revealed between any of the bull sample-groups or sire-breed groups (P > 0.10) or between bulls classified as satisfactory or unsatisfactory. Examination of testicles and epididymides revealed no gross abnormalities, except for bull B6 (GRP B) who was detected to have unequal testicle size due to one testicle being smaller and softer in tone. No association of age with SC was revealed (P > 0.10).

Progressive, individual spermatozoal motility (MOT%) for all bulls was  $57.8 \pm 1.6\%$  (range, 25-80%). All UFB bulls exceeded the >30% threshold for progressive motility and 49 of 60 (82%) had motility percentages greater than 50%. Four NUB bulls had MOT%

<30% and 9 of 14 (64%) had motility >50%. No differences in spermatozoal motility were detected between bull sample-groups or sire-breed groups (P > 0.10). MOT% was not significantly associated with SC or age although a negative correlation was demonstrated with primary abnormal sperm (PRM%), r = -0.28 (P < 0.05) and secondary abnormal sperm (SEC%), r = -0.33 (P < 0.05). Bulls classified as unsatisfactory potential-breeders (n = 7) had a lower MOT% than the satisfactory bulls, 38.4% compared to 59.9%, (P < 0.05).

Evaluation for morphological characteristics revealed the percentage of normal spermatozoa (MRPH%) was  $80.4 \pm 1.5\%$  for all bulls. The range of distribution for normal spermatozoa was 28-96%. All UFB bulls, except bull B6 (28% normal sperm), exceeded the >70% threshold for morphologically normal spermatozoa. Nine of 14 NUB bulls (64%) had >70% MRPH%. There were no differences revealed in percentage of normal morphology for any of the bull sample-groups or sire-breed groups (P > 0.10). Bulls classified as unsatisfactory potential-breeders (n = 7) had lower percentages of morphologically normal spermatozoa,  $52.3 \pm 8.5\%$ , than bulls classified as satisfactory (n = 67),  $83.3 \pm 0.9\%$  (P < 0.05). MRPH% was weakly and negatively correlated to SC, p = -0.20 (p = 0.08) and positively correlated to MOT%, p = 0.37 (p < 0.01).

Spermatozoa with abnormal morphological features were classified according to defect-type and were categorized as primary (PRM%) or secondary abnormalities (SEC%). Overall, the percentage of primary abnormal spermatozoa was  $13.3 \pm 1.8\%$ , which included specific abnormalities of the spermatozoal head, acrosome, midpiece and/ or the presence of proximal protoplasmic droplets. The range of distribution for primary abnormal sperm was 2-54%. The bulls classified as unsatisfactory had higher percentages of primary abnormal spermatozoa, 29.1% compared to 12.1% (P < 0.01) for the bulls classified as satisfactory (P < 0.05). The percentage of secondary abnormal spermatozoa was  $6.3 \pm 0.7\%$  (range, 0–37%). Defects categorized as secondary included specific tail abnormalities, normal-appearing detached sperm heads, and distal protoplasmic droplets. The bulls classified as unsatisfactory had higher percentages of secondary abnormal sperm than the satisfactory bulls (P < 0.01), 18.6 versus 4.5%, respectively No differences in primary or secondary morphologically abnormal spermatozoa were revealed between UFB bull sample-groups or sire-breed groups (P > 0.10). The LM sirebreed group had numerically higher PRM% (21.2  $\pm$  6.8%) due to the inclusion of bull B6 with 54% primary abnormal spermatozoa. No differences in specific classes of morphology defects (primary or secondary) were detected between sample-groups (P > 0.10). Midpiece defects (5.2%), detached, normal sperm heads (3.6%) and proximal protoplasmic droplets (3.6%) were numerically the most prevalent for bulls classified as satisfactory. No significant correlations were demonstrated for PRM% with SC and age. However, SEC% was positively associated with SC, r = 0.26 (P < 0.05); refer to Tables 2 and 3 for BSE 1 results.

Physical and reproductive examination of the bulls revealed several individuals with abnormalities. Abnormalities (enlargement, increased tone, tenderness) of the vesicular glands (seminal vesiculitis) were detected by palpation in three NUB bulls with concomitant white blood cells in the semen. A bull with an injured foot and one bull with a persistent penile frenulum were placed in the NUB group and one bull, which failed to adequately extend the penis during electroejaculation, was also placed as a NUB.

Table 2 Reproductive characteristics

	n	Least squar	re means (S.E.	M.) for bull p	opulation and sa	ample groups	
		Age BSE (days)	SC BSE (cm)	MOT% (%live)	MRPH% (% normal)	PRM% (% primary)	Satis% (satisfactory)
BSE 1							
All Bulls	74	428 (0.8)	36.3 (0.2)	57.8 (1.6)	80.4 (1.5)	13.3 (1.8)	90.50
UFB	60	428 (0.9)	36.5 (0.3)	59.4 (1.7)	81.8 (1.3)	13.5 (1.1)	98
NUB	14	430 (1.6)	36.2 (0.6)	51.0 (4.4)	74.4 (5.7)	14.7 (3.1)	57
Herd 1	19	429 (2.0)	35.9 (0.4)	56.3 (3.3)	84.2 (1.3)	10.6 (1.6)	100
Herd 2	19	428 (2.0)	36.9 (0.4)	60.5 (3.3)	80.6 (3.2)	14.4 (2.8)	95
Herd 3	22	427 (1.0)	36.3 (0.6)	61.1 (2.3)	80.8 (2.0)	15.0 (2.3)	100
R1—ABC	30	427 (1.3)	36.6 (0.4)	55.8 (2.5)	81.1 (2.2)	14.4 (2.8)	97
R2—DEF	30	428 (1.1)	36.1 (0.3)	63.0 (2.0)	82.4 (1.4)	12.6 (1.7)	100
GRP (UFB on	ly)						
Α	9	428	35.7 (0.5)	51.1 (5.1)	85.7 (1.2)	9.6 (1.4)	100
В	10	428	37.3 (0.6)	58.0 (4.4)	76.6 (5.8)	17.3 (4.4)	90
C	11	426	36.7 (0.9)	57.7 (3.8)	81.5 (2.7)	15.2 (2.0)	100
D	10	430	36.1 (0.6)	61.0 (3.8)	82.8 (2.1)	11.7 (1.7)	100
E	9	428	36.4 (0.5)	63.3 (5.0)	85.0 (1.5)	10.8 (1.3)	100
F	11	428	35.9 (0.6)	64.5 (2.1)	80.0 (3.2)	14.9 (2.6)	100
SBL (UFB onl	y)						
#1—AN	6	433	36.4 (2.6)	68.3 (1.7)	86.7 (2.4)	7.7 (1.0)	100
#2—CH	9	434	35.4 (0.6)	55.5 (5.4)	84.4 (1.9)	11.3 (1.0)	100
#3—GB	9	427	36.9 (0.5)	56.7 (5.5)	83.3 (2.6)	12.6 (2.7)	100
#4—HH	10	426	37.2 (1.0)	67.0 (2.6)	79.3 (2.2)	16.2 (1.7)	100
#5—LM	6	425	35.8 (0.5)	54.2 (7.6)	73.2 (9.3)	21.2 (6.8)	83
#6—AR	10	426	35.5 (0.5)	54.0 (3.1)	81.4 (2.4)	12.8 (2.0)	100
#7—SM	10	425	37.1 (0.6)	61.0 (3.1)	82.9 (3.2)	13.1 (2.7)	100
Range		414-442	32.5-45.0	25-80	28-96	2-54	

BSE 1: breeding soundness evaluation.

Analysis of least square means via ANOVA-single factor test, Kruskal-Wallis test.

Analysis of rates and proportions via Chi square; trend analysis, multiple comparisons.

Rows with different letters are different (P < 0.05 or less).

At the beginning of the breeding season, the non-shrunk BW of the UFB bulls was  $549 \pm 6$  kg, BCS was estimated at 6.1, and hip height averaged 52.1 in. For the NUB bulls, BW was interpolated at 555 kg, BCS was estimated at 6.1 and hip height averaged 52.6 in. No differences were revealed between UFB sample-groups or sire-breed groups (P > 0.10) or between bulls classified satisfactory or unsatisfactory. BW was positively correlated with BCS (r = 0.41, P < 0.001), HH (r = 0.61, P < 0.001), and SC (r = 0.46, P < 0.001); refer to Table 4.

Scrotal circumference, re-measured just prior to the first mating exposures for all UFB bulls, was  $35.9 \pm 0.3$  cm, with a range from 32.0 to 44.0 cm. No differences were demonstrated between bull sample-groups or sire-breed groups (P > 0.10). SC just prior to mating was 0.6 cm less than the SC measured at BSE 1, corresponding to a time period of

Table 3						
Reproductive	characteristics	for	BSE	1:	UFB	bulls

BSE variables	Least square means and	l S.E.M.		
	BSE 1 satisfactory	BSE 1 unsatisfactory	BSE 1 UFB bulls	
n	67	7	60	
Age (days)	428 (0.8)	429 (1.5)	428 (0.9)	
SC (cm)	36.2 (0.3)	37.1 (0.8)	36.5 (0.3)	
MOT%	59.9 (1.5) a	38.4 (4.5) b	59.4 (1.7)	
MRPH%	83.3 (0.9) a	52.3 (8.5) b	81.8 (1.3)	
PRM% (primary)	12.1 (0.8) a	29.1 (8.3) b	13.5 (1.1)	
SEC% (secondary)	4.5 (0.4) a	18.6 (4.7) b	4.7 (1)	
Head defects (%)	3.3	4.1	3.3	
Midpiece defects (%)	5.2	8.9	5.2	
Tail defects (%)	1.0 a	5.6 b	1.0	
Detached heads (%)	3.6 a	14.7 b	3.6	
Prox. droplets (%)	3.6 a	13.0 b	3.6	
Acrosome defects (%)	0.8	1.1	0.8	
Nuclear defects (%)	0.2	0.7	0.7	

Comparison of satisfactory and unsatisfactory bulls. Analysis of least square means and variances via ANOVA-single factor test. Columns with different letters (a and b) are different (P < 0.05). Classification of primary (PRM%) and secondary (SEC%) spermatozoal abnormalities and specific classification of spermatozoal defects according to reference sources [2,22].

17–31 days. Thirty-two UFB bulls (53%) had declines in SC from 0.5 to 2.5 cm, 13 UFB bulls (22%) had increases in SC from 0.5 to 2.0 cm, and 15 UFB bulls (25%) had no change in SC. Overall, the change in SC for UFB bulls represented a 1.67% decline in SC. No differences were revealed for percentage-change of SC between UFB sample-groups or sire-breed groups (P > 0.05); refer to Table 4.

SC was positively correlated with starting BW, r = 0.46 (P < 0.001), BCS, r = 0.27 (P < 0.05), and hip height, r = 0.38 (P < 0.001). In addition, SC was positively correlated with MOT% from BSE 1, r = 0.22 (P = 0.08) although revealing a weak trend for negative correlation with MRPH%, r = -0.12 (P = 0.25).

Physical inspection of the UFB bulls just prior to the first mating exposures revealed no detectable abnormalities or injuries.

Table 4
Pre-breeding physical characteristics for bull population and sample groups

	Least squa	Least square means and S.E.M.									
	UFB	NUB	Herd 1	Herd 2	Herd 3	R1 (ABC)	R2 (DEF)	Range			
$\overline{n}$	60	14	19	19	22	30	30	60			
Age (days)	454 (1)		453 (3)	454 (2)	455 (2)	446 (1)	461 (1)	431 to 473			
BW (kg)	549 (6)	555 <sup>a</sup>	534 (10)	558 (12)	554 (9)	553 (10)	545 (8)	445 to 643			
Hip ht. (in)	52.1 (0.1)	52.6	51.6 (0.3)	52.3 (0.3)	51.9 (0.3)	52.1 (0.3)	51.8 (0.3)	48.0 to 54.5			
BCS	6.11 (0.1)	6.10	6.11 (0.1)	6.10 (0.1)	6.16 (0.1)	6.11 (0.1)	6.12 (0.1)	5.5 to 7.0			
SC (cm)	35.9 (0.3)		35.2 (0.4)	36.2 (0.3)	36.1 (0.5)	35.9 (0.4)	35.8 (0.3)	32.0 to 44.0			
SC chg (cm)	0.6 (0.13)		0.7	0.7	0.2	0.7	0.4	2.0  to  -2.5			
SC chg (%)	-1.67		-1.99	-1.93	-0.55	-1.95	-0.84	5.8  to  -6.9			

<sup>&</sup>lt;sup>a</sup> BW interpolated from ave.-daily-gain from yearling BW of UFB bulls.

#### 3.2. Post-breeding observations

Within 3 days following removal of each GRP from their respective HRD, a Breeding Soundness Evaluation (BSE 2) was performed on the remaining UFB bulls (n = 57). The elapsed time between BSE 1 and BSE 2 was an average of 72 days for the UFB bulls, and ranged from 61 to 87 days by GRP. R1 averaged 64 days and R2 averaged 84 days, the difference between R corresponding to the 21-day duration of the last mating period. The NUB bulls were similarly subjected to a follow-up BSE on Day 67 following BSE 1.

The SC for the UFB bulls was  $34.9\pm0.2$  cm, a 2.87% decline from the SC of  $35.9\pm0.3$  cm at the start of the breeding season and an overall decline of 4.58% from the  $36.5\pm0.3$  cm SC at BSE 1 (P<0.01; Fig. 1). The range of percentage of decline for 54 UFB bulls was 0-12.5% (4.0 cm) with 3 UFB bulls having SC increases up to 2.9% (1.0 cm). The range of SC measurements was 31.0-41.0 cm. Conversely, 12 NUB bulls had an increase in SC from BSE 1 to BSE 2 of 2.49%,  $36.2\pm0.6$  to  $37.1\pm0.6$  cm respectively, with one individual declining in SC and one bull diagnosed with orchitis. The decline in SC for UFB bulls was decidedly different than the increase of SC for NUB bulls (P<0.001; Table 5).

The SC at the end of breeding was strongly correlated to SC at the start of breeding (r=0.79, P<0.01). The percentage-change in SC from BSE 1 to BSE 2 was positively correlated to SC at BSE 1 (r=0.39, P=0.01) with bulls having SC greater than 36 cm at BSE 1 more likely to have negative and greater SC changes (P<0.05). The change in SC was weakly associated with BW loss during the first mating period (r=0.22, P=0.11) and modestly associated with BW loss during the second mating period (r=0.35, P=0.05) and with total BW loss (r=0.24, P=0.07). No differences in change of SC or mean SC at the end of breeding were demonstrated between UFB sample-groups or sire-breed groups (P>0.10). The change in SC measured during BSE 2 revealed no significant relationships with R, HRD, GRP or SBL (P>0.10). The change in SC at BSE 2 had minimal and weak associations with semen quality parameters. Rank correlation with MOT% was r=0.13 (P>0.10) and with MRPH% was r=0.10 (P>0.10). No association was detected with PRM% or SEC%.

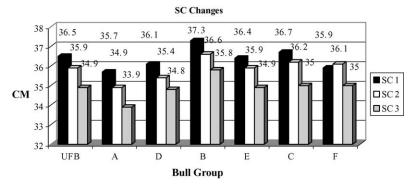


Fig. 1. SC Changes from BSE 1, to start of breeding, to BSE 2. SC1—scrotal circumference at BSE 1, SC2—scrotal circumference at start of breeding season, SC3—scrotal circumference at BSE 2.

Table 5
Reproductive characteristics of bulls

	Lea	st square	means (S.E	E.M.) for bull	population an	d sample grou	ıps	
	n	Time (days)	MOT% (%live)	MRPH% (%normal)	PRM% (%primary)	SEC% (%second)	Satis% (satisfactory)	SC BSE 2 (cm)
BSE 2 results All bulls	70	72	52.7 (2.5)	63.0 (3.1)	23.8 (2)	13.2 (1.5)	58.60	35.4 (0.2)
UFB NUB	57 13	73 67	54.5 (2.5) 48.9 (6.5)	65.2 (3.2) 51.3 (8.2)	24.7 (2.4) 21.7 (4.4)	9.7 (1.6) 23.0 (6.0)	63.00 a 36.00 b	34.9 a (0.2) 37.1 b (0.6)
Herd 1 Herd 2 Herd 3	17 19 21	72 74 76	57.6 (6) 59.5 (4) 47.4 (7)	70.2 (4) 66.7 (8) 60.9 (7)	19.9 (4) 24.5 (6) 28.8 (5)	9.8 (3.1) 8.8 (2.4) 10.3 (2.9)	76.00 63.00 52.00	35.9 a (0.4) 36.9 ab (0.4) 36.3 b (0.6)
R1—ABC R2—DEF	30 27	64 84	52.7 (3.9) 56.5 (3.3)	61.1 (4.8) 70.6 (3.9)	26.4 (5) 22.7 (5)	12.4 (2.7) 6.6 (1.3)	57.00 70.00	35.0 a (0.4) 34.9 b (0.3)
GRP (UFB or	nly)							
A B C	9 10 11	61 63 66	55.6 (6) 60.0 (4) 43.6 (8)	59.9 (10) 69.9 (7) 54.2 (9)	26.5 a (7) 20.6 ab (5) 31.6 a (6)	13.6 ab (6) 9.5 ab (4) 14.2 a (3)	56.00 a 70.00 ab 46.00 a	33.9 ab (0.5) 35.8 b (0.5) 35.0 a (0.8)
D E F	8 9 10	82 84 87	60.0 (7) 58.9 (4) 51.5 (6)	82.1 (2) 63.1 (9) 68.2 (6)	12.5 b (2) 28.7 ab (8) 25.7 ab (5)	5.4 b (1) 8.1 ab (3) 6.1 ab (4)	100.00 b 56.00 ab 60.00 b	34.8 c (0.5) 34.9 ab (0.3) 35.0 ab (0.6)
SBL (UFB on	ly)							
#1—AN #2—CH	6		43.3 54.4	46.3 72.6	38.3 a 19.9 b	15.4 a 7.5 b	50.00 a 78.00 ab	36.4 a (2.6) 35.4 b (0.6)
#3—GB #4—HH	9 9		56.7 46.1	75.0 59.3	19.3 b 27.9 ab	5.7 b 12.8 ab	78.00 b 44.00 ab	36.9 b (0.5) 37.2 a (1.0)
#5—LM #6—AR	6		50.0 58.9	55.0 69.3	25.2 ab 23.4 ab	19.8 ab 7.3 ab	50.00 a 67.00 ab	35.8 a (0.5) 35.5 b (0.5)
#7—SM Range (%)	9		66.7 0–80	71.9 0–94	25.6 b 3–87	2.5 ab 0–82	67.00 b	37.1 b (0.6) 31.0–41.0

Breeding soundness evaluation (BSE 2).

Analysis of least square means and variance via ANOVA-single factor test.

Analysis of rates and proportions via Chi square test of association; trend analysis, multiple comparisons.

Columns with different letters (a and b) are different (P < 0.05 or less).

Time = days between BSE 1 and BSE 2.

Estimation of progressive individual spermatozoal motility (MOT%) for the UFB bulls was  $54.5 \pm 2.5\%$  and for the NUB bulls was  $48.9 \pm 6.5\%$  (P > 0.10). MOT% for BSE 2 was not different than MOT% for BSE 1 (P > 0.10), for either UFB or NUB bulls. Individually, 56% (n = 32) of the UFB had increases in MOT% and 44% (n = 25) had declines. The range of MOT% was 0-80% progressively motile sperm. No differences were detected between R, HRD, GRP or SBL (P > 0.10); refer to Tables 5 and 6. MOT% was associated with BSE 2 classification (P < 0.01) as 16 of the 21 bulls classified as unsatisfactory had motility percentages less than 30% and 14 of 25 UFB bulls with MOT% declines were classified as unsatisfactory. UFB bulls classified as unsatisfactory (n = 21)

had MOT% of 40.4% compared to the 62.8% for the bulls classified as satisfactory (P < 0.01). No association was revealed between SC at the end of the breeding season and MOT%, and no correlation between the change in SC from BSE 1 to BSE 2 with MOT% was revealed, r = 0.13 (P > 0.10). A strong, positive correlation of MOT% with MRPH% existed (r = 0.55, P < 0.01), indicating that as motility percentage decreased, percentage of morphologically normal spermatozoa decreased. MOT% on BSE 2 demonstrated no significant associations with body weight losses or with injury scores (P > 0.10).

Percentage of normal spermatozoa (MRPH%) declined from  $80.4 \pm 1.5\%$  on BSE 1 to  $63.0 \pm 3.1\%$  for the 70 bulls completing BSE 2 (P < 0.01). For both the UFB and NUB bulls, MRPH% declined 7–32% for all bull-groups, with the single exception of GRP D, whose 8 remaining bulls maintained a high percentage of normal spermatozoa on both evaluations (82%). MRPH% declined for the UFB bulls from 81.8 to 65.2% (P < 0.05) and for the NUB bulls from 74.4 to 51.3% (P < 0.05). The declines in MRPH% were not different between UFB and NUB (P > 0.10). Individually, 18 UFB bulls had increased MRPH% and 39 had declines (P < 0.01). The range of MRPH% was from 0 to 94% morphologically normal spermatozoa. No differences in MRPH% or the level of decline of MRPH from BSE 1 to BSE 2 were demonstrated for R or HRD; however, GRP and SBL differences in MRPH% were evident (P < 0.05). GRP A and C demonstrated lower percentages of normal spermatozoa (54–59%) than the other GRP, particularly in comparison to GRP D (82%), and the AN- (46%) and the LM-SBL (55%) had considerably reduced MRPH% than the other SBL (P < 0.05); refer to Tables 5 and 6. The degree of change in sperm morphology was influential on BSE 2 classification, as bulls with improved MRPH% (n = 19) or low levels of declines (n = 10) were highly likely (8.7 to 1) to be classified as satisfactory, whereas bulls with greater than 10% declines (n = 28) were 1.8-times more likely to be classified as unsatisfactory (P < 0.001). The AN-SBL revealed a 40% decline in MRPH% overall which resulted in 50% of this SBL being classified as unsatisfactory. Bulls classified as satisfactory had MRPH% of  $80.6 \pm 1\%$  compared to  $40.0 \pm 5\%$  for the bulls classified as unsatisfactory (P < 0.01).

The percentage of primary abnormal spermatozoa (PRM%) increased from levels revealed at BSE 1,  $23.8 \pm 2\%$  compared to 13.3 + 2%, respectively (P < 0.01). Increases of PRM% were larger for the UFB bulls (11.2% increase) compared to the NUB (7% increase). Differences in PRM% were demonstrated between GRP and SBL, with GRP C (31.6%) and the AN-SBL (38.3%) having higher PRM% than the other groups (P < 0.05); refer to Fig. 2. Differences in PRM% were not demonstrated for R or HRD, although HRD3 had a trend for higher primary abnormal spermatozoa (P = 0.09). R1 had numerically more PRM% than R2 (P = 0.12). All classes of primary abnormal defects increased from BSE 1, with proximal protoplasmic droplets, abnormal head shape and nuclear defects (diadem craters) being the predominant and most significant increases (P < 0.05). The bulls classified as unsatisfactory had nearly double the levels of acrosome, abnormal head shape and midpiece defects, 5-times more protoplasmic droplets and nearly 10-times the percentages of nuclei with diadem craters (9.8 versus 1.0%) than the satisfactory bulls (P < 0.05); refer to Table 6.

Secondary spermatozoal abnormalities (SEC%) increased at BSE 2 to  $13.2 \pm 1.5\%$  for all bulls, compared to  $6.3 \pm 1\%$  on BSE 1 (P < 0.05), however, the NUB bulls revealed a marked increase in SEC% (23.0% compared to 10.9%) and the UFB bulls had a modest

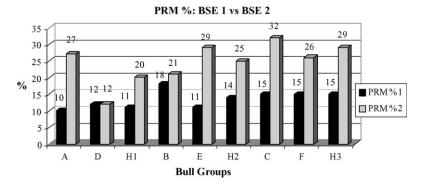


Fig. 2. Changes in percent primary abnormal spermatozoa (PRM%) in bulls from BSE 1 to BSE 2. HRD and GRP. PRM% (least square means).

increase (9.7% compared to 4.7%). No differences were found for HRD or R, although R1 had numerically higher SEC% than R2 (P = 0.09), similar to findings with the PRM% differences. Differences were detected between GRP as well as SBL, with GRP A and C demonstrating higher SEC% and the AN-, HH-, and LM-SBL having higher SEC% (P < 0.05). The principle secondary defects revealed were detached, normal sperm heads (8.8%); refer to Table 6.

Final classification for the 70 bulls available for BSE 2 revealed a significantly lower percentage of bulls achieving satisfactory classification (58.6%) compared to BSE 1

Table 6 Semen characteristics of bulls (UFB only)

BSE variables	Least square means (S.	E.)	
	BSE 2 satisfactory	BSE 2 unsatisfactory	BSE 2 UFB bulls
n	36	21	57
MOT% BSE 2	62.8 (2) a	40.4 (4) b	54.5 (2.5)
MOT% BSE 1	59.0 (2)	59.0 (3)	59.4 (1.7)
MRPH% BSE 2	80.6 (1) a	40.0 (5) b	66.2 (3.2)
MRPH% BSE 1	83.3 (1)	78.7 (3)	81.8 (1.3)
PRM% BSE 2	13.8 (1) a	43.4 (3.6) b	24.7 (2.4)
PRM% BSE 1	12.1 (1)	16.6 (3.8)	13.5 (1.1)
SEC% BSE 2	5.6 (1) a	16.8 (4) b	9.7 (1.6)
SEC% BSE 1	4.7 (1)	4.6 (1)	4.7 (1)
Head defects (%)	4.0 (0.5) a	9.5 (2.0) b	6.0 (0.3)
Midpiece defects (%)	6.2 (0.7) a	11.3 (1.7) b	8.1 (0.4)
Tail defects (%)	1.1 (0.2)	0.5 (0.1)	0.8 (0.1)
Detached heads (%)	4.5 (0.6) a	16.3 (3.9) b	8.8 (0.5)
Prox. droplets (%)	2.1 (0.3) a	10.8 (2.2) b	5.3 (0.2)
Acrosome defects (%)	0.6 (0.2)	2.0 (0.8)	1.1 (0.1)
Nuclear defects (%)	1.0 (0.5) a	9.8 (3.5) b	4.2 (0.5)

Comparison of satisfactory and unsatisfactory bulls. Columns with different letters (a and b) are different (P < 0.05).

Analysis of least square means and variances via ANOVA-single factor test.

Analysis of rates and proportions via Chi square test of association; trend analysis, multiple comparisons.

(90.5%), (P < 0.01). Bulls D4, D6, F4 and one NUB were not available for testing due to injuries that conditionally would result in unsatisfactory classification, reducing the satisfactory-percentage to 55% for all 74 original bulls. For all 60 UFB bulls, 36 (60%) were classified as satisfactory and 24 (40%) were classified as unsatisfactory on BSE 2, including bull B6 who was originally unsatisfactory on BSE 1; refer to Table 5. Only 5 of 14 NUB bulls were classified as satisfactory (36%) and all NUB bulls unsatisfactory on BSE 1 (n = 6) remained unsatisfactory on BSE 2. R1 had less satisfactory bulls than R2, 57% compared to 70%, respectively (P < 0.05). HRD3 had 52% satisfactory, which was different from HRD1 (76%), (P < 0.05), and HRD2 had 63% satisfactory. By GRP, GRP C had only 44% satisfactory, GRPA, E, F, and B had 56–70%, and GRP D had 100% achieve satisfactory classification (P < 0.05); refer to Fig. 3.

No differences in pre-breeding physical characteristics (BW, BCS, HH, Age, SC) were found between bulls classified as satisfactory or unsatisfactory on BSE 2. The satisfactory bulls did have less SC decline (2.6%) compared to the unsatisfactory (3.3%) (P < 0.05) however, no differences in body weight loss during either mating period or total BW loss were evident; refer to Table 7.

Through the breeding season, UFB bulls had body weight losses of  $58 \pm 2$  kg during the first mating period (range of 29–91 kg) and  $36 \pm 2$  kg during the second mating period (range of 7–75 kg). The UFB bulls completing both mating periods (n = 47) incurred a total weight loss of  $77 \pm 3$  kg (range of 25–121 kg). The total weight loss was equivalent to  $13.9 \pm 0.4\%$  of the BW at the start of the breeding season (5.1–20.2%). Bulls in R1 had higher total weight loss than bulls in R2, during less days of mating exposure (P < 0.05). A significant relationship between rotation (breeding season time-period) and BW loss was demonstrated, r = 0.40 (P < 0.05) and total BW loss was positively associated with BW at the start of breeding, r = 0.62 (P < 0.01). The 13 UFB bulls removed from mating due to injuries, although only exposed to mating for 5–24 days, incurred total weight loss of  $50 \pm 5$  kg or 9.2% loss of body weight from the starting BW. During the 14-day rest period between mating exposures, 82% (n = 45) of the UFB bulls gained BW of  $15 \pm 2$  kg (range of -18–42 kg). BW at the end of the breeding season was  $478 \pm 5$  kg (range of 381–548 kg), which was less than the non-adjusted average yearling weight for the entire bull population. BCS declined in conjunction with weight loss to  $4.6 \pm 0.1$  (range of 4–5 units).

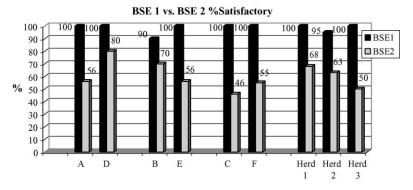


Fig. 3. Bulls classified as satisfactory for BSE 2 and BSE 1. HRD and GRP percentages.

Table 7
Comparison (physical characteristics) of bulls classified as satisfactory and unsatisfactory for BSE 2

BSE variables	Least square means (S.E.)	
	BSE 2 satisfactory	BSE 2 unsatisfactory
n	36	21
BW start (kg)	548 (8)	556 (10)
HH (in.)	52.0 (0.3)	51.8 (0.3)
SC BSE 1 (cm)	36.5 (0.3)	36.4 (0.5)
SC start (cm)	35.9 (0.3)	36.0 (0.5)
SC end (cm)	35.0 (0.2)	34.8 (0.5)
SC chg (%)	2.60 (0.5) a	3.30 (0.8) b
BWL1 (kg)	58 (2)	59 (3)
BWL2 (kg)	35 (2)	35 (1)
TWL (kg)	75 (2)	78 (3)
Bull-days	29.0 (1)	26.9 (2)
Lame (%)	58 a	81 b
Removed (%)	8 a	33 b

Columns with different letters are different (P < 0.05).

Analysis of least square means and variances via ANOVA-single factor test.

Analysis of rates and proportions via Chi square test of association; trend analysis, multiple comparisons.

During the same time period, the NUB bulls gained  $27 \pm 8$  kg with a final BW of 585 + 8 kg and BCS of 6.2 units; refer to Table 8.

An unexpected high incidence of lameness and reproductive injuries occurred in the UFB bulls during the breeding season. Total observed cases of lameness were 52 bulls, with

Table 8
Changes in body weight and BCS of bulls during the breeding season UFB and sample groups

	Least square	e means (S.E.)	and percenta	ges			
	UFB	HRD1	HRD2	HRD3	R1	R2	Range
n	57	19	19	19	28	29	
Bull-days	1651	530	581	540	698	953	5–35
BWL1	58 (2)	54 (2) a	69 (3) b	51 (3) a	63 (2) a	52 (2) b	29-91
BW chg (%)	10.5 (0.3)	10.1 (0.5)	12.4 (6)	9.2 (0.5)	11.4 (0.3)	9.6 (0.5)	5.5-14.6
n	48	14	18	16	22	26	
BWL2	36 (2)	42 (3) a	33 (2) b	38 (5) a	35 (3)	37 (3)	7–75
TWL	77 (5)	74 (5) a	74 (4) a	84 (6) b	82 (5) a	73 (4) b	25-121
BW chg (%)	13.9 (0.4)	13.9 (0.9)	13.2 (0.6)	14.7 (0.9)	14.7 (6)	13.3 (6)	5.1-20.2
BW2	478 (12)	458 (12)	484 (10)	480 (7)	473 (10)	476 (7)	382-548
BCS2	4.60 (0.1)	4.66 (0.1)	4.58 (0.1)	4.55 (0.1)	4.65 (0.1)	4.58 (0.1)	4-5

Bull days = total days of bull exposure by bulls/sample. Columns with different letters (a and b) are different (P < 0.05 or less). Bull-days, days of bull exposure for bull-group; BWL1, body weight loss during first mating period (kg); BW chg. (%), percentage BW change from starting BW; BWL2, body weight loss during second mating period (kg); TWL, total body weight loss through breeding season (kg); BW2, non-shrunk BW at end of breeding season (kg); BCS2, body condition score at end of breeding season.

Analysis of least square means and variance via ANOVA-single factor test.

Analysis of frequencies via Chi square test of association; trend analysis, adj residuals.

38 of the bulls observed lame at least once (63%) and 14 bulls lame more than once (23%). As for reproductive injuries, 7 of the 60 bulls (12%) incurred prepucial or penile injuries, of which three cases were detected during the mating exposures and four were detected following the breeding season. Overall, 45 bulls (75%) incurred injuries and 13 (22%) were removed prior to the completion of the season. Three bulls were added as replacements during the first mating period only. Of the 60 UFB bulls, 47 (78%) remained at the completion of the breeding season, although 16 (34%) were lame and four had existing reproductive injuries. No injuries causing lameness were observed in the NUB bulls and one bull had testicular abnormalities. The incidence rates of lameness were 53% for R1 and 73% for R2 (P = 0.11), however more of the cases of lameness in R2 were considered minor and occurred late in the season. Herd incidence rates ranged from 47% in HRD2, to 68% in HRD3 and 74% in HRD1 (P = 0.20). Differences in injury rates were present between bull sample-groups and sire-breed lines (P < 0.05). No significant associations were evident for injuries or injury rates with any physical or pre-breeding reproductive traits. However, bulls used in HRD1 (GRP A and D), having the highest overall injury rate (74%), revealed a significant rank correlation for injury score with hip height, r = 0.62 and body weight, r = 0.54 (P < 0.05), whereas similar associations were not detected in the other two herd-groups of bulls.

The bulls classified as unsatisfactory on BSE 2 had a disproportionate level of injuries and bulls removed due to injuries, with 81% of these bulls incurring injuries and 33% being removed prior to the end of the mating season, compared to 58% injury rate and 8% removal-rate for the bulls classified as satisfactory (P < 0.01). Including the three injured individuals not available for BSE 2 would widen this difference; refer to Table 7.

Pregnancy rates were not significantly different between herds: HRD1 = 93.3%, HRD2 = 91.6% and HRD3 = 95.4 (P = 0.32). Similarly, no differences in calving rates were revealed between herds: 91.2, 89.5 and 93.4%, respectively. No differences in calving rates during the first, second or third 21-day periods of the calving season were detected between herds; refer to Table 9a. Utilizing calving dates and estimated date of conception, calves sired by GRP were estimated. Although numerical differences in percentage of calves born per exposed female were evident, no differences (P > 0.05) were demonstrated

Table 9a		
Pregnancy rates of HRD	and calving rates by mating	period for each herd

	Percentages and fi	Percentages and frequency counts						
	HRD1	HRD2	HRD3	P				
n	194	191	196					
Pregnancy rate	93.30%	91.60%	95.40%	0.32				
Calving percentage <sup>a</sup>	(% of total herd)							
Period 1	80 (41.2)	64 (33.5)	71 (36.2)	0.28				
Period 2	50 (25.8)	54 (28.3)	68 (34.7)	0.14				
Period 3	28 (14.4)	25 (13.1)	30 (15.3)	0.82				
Period 4	18 (9.30) a	28 (14.7) b	14 (7.10) a	0.04				

Analysis performed via Chi square test of association. Values in parentheses are percentages. Columns with different letters (a and b) are different (P < 0.05).

<sup>&</sup>lt;sup>a</sup> Calves born per period per total females exposed for breeding.

Table 9b Estimated calving rates by mating periods (*P*) for GRP Calving rates for females available to become pregnant during corresponding mating periods

HRD1		Females calves		HRD2 GRP	Females calves			HRD3	Females calves			
	GRP	Available	Born	%		Available	Born	%	GRP	Available	Born	%
P1	A	194	80	41	В	191	64	34	C	196	71	36 <sup>a</sup>
P2	D	114	50	44	E	127	54	43	F	125	68	54
P3	A	64	28	44	В	73	25	34	C	57	30	53
P4	D	36	18	50	E	48	28	58 <sup>a</sup>	F	27	14	52

Analysis via Chi square test of association and trend analysis of proportions.

Females available to conceive during each subsequent mating period.

Calves born during corresponding calving period.

for bull-groups during the first three mating periods, however, GRP E did have higher numbers of calves born during the last 21 days of the calving season when compared to GRP D and F (P < 0.05); refer to Table 9b. More concise and specific information on bull reproductive performance will be elucidated with DNA-genotyping of the offspring, sires and dams to assign parentage to each individual calf.

#### 3.3. Environmental conditions prior to and during the breeding season

At the time of BSE 1 (May 16, 2002), average daily temperatures had been below normal for 13 of the prior 16 days (range from 46 to 69 °F). Mean daily-temperature for the month of May was 59 °F, 2.7 °F below normal with the average maximum-temperature of 70 °F. At the end of the month and the first 2 days of June, five continuous days of temperatures 8–18 °F higher than normal occurred, with average daily high-temperatures of 90–99 °F and average daily-temperatures of 74–86 °F. Another 4 days of higher than normal daily-temperatures occurred during the turnout time of R1 bulls, with >90 °F average daily-highs and >75 °F average daily-temperatures. During the turnout period for R2 bulls, average daily high-temperatures exceeded 90 °F for 10 days and average daily-temperatures exceeded 78 °F. The average daily-temperature for June was 77 °F and average maximum-temperature was 90 °F, with 19 days exceeding 90 °F. Precipitation during the month of May 2002 was 1.12 in. below normal and during June was 2.64 in. below normal.

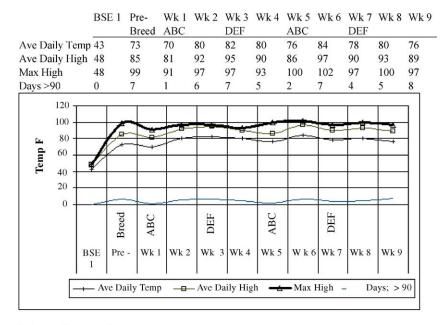
The average daily temperatures, average daily high temperatures, and maximum high temperatures are charted in Fig. 4. The reference climate period was from 1971 to 2000.

During the first mating period of R1, 7 of the 14 days for each of GRP A, B and C had daily high temperatures greater than 90 °F and daily low temperatures greater than 70 °F, with 11 days of maximum high temperatures from 90 to 97 °F. Average daily temperature rose from 70 °F during Week-1 to 80 °F during Week-2. Only a trace of precipitation was recorded and relative humidity ranged from 30 to 58%.

During the first mating period for R2, 10 of the 14 days for each of GRP D, E and F had daily high temperatures greater than 90 °F and daily low temperatures greater than 70 °F,

<sup>%,</sup> percent of calves born per available cow to conceive per period (calving rate).

<sup>&</sup>lt;sup>a</sup> Denotes percentage differences between mating periods within herd only.



Wk = week of breeding season
Recorded at National Weather Service Regionla Center, Hastings, NE, USA.
ABC = R1 DEF = R2

Fig. 4. Weekly average air temperature readings for bull breeding season. (temperatures in °F).

with 12 days of maximum high temperatures from 90 to  $101\,^{\circ}$ F. Average daily temperatures for the two-week period was  $80-82\,^{\circ}$ F. No precipitation was recorded and relative humidity ranged from 30 to 50%.

During the second mating period for R1, 8 of the 14 days had daily high temperatures greater than 90  $^{\circ}$ F and daily low temperatures greater than 70  $^{\circ}$ F, with 10 days of maximum high temperatures from 90 to 104  $^{\circ}$ F. Average daily temperature for week 5 was 76 and 84  $^{\circ}$ F for week 6. Only a trace of precipitation was recorded and average relative humidity ranged from 25 to 60%.

During the second mating period for R2, 10 of the 21 days had daily high temperatures greater than 90 °F and daily low temperatures greater than 70 °F, with 15 days of maximum high temperatures from 90 to 104 °F. Average daily temperatures were near 80 °F for each week. Precipitation was recorded on 6 days, totaling 1.9 in. of rain and relative humidity ranged from 30 to 60% on the days not recording precipitation.

Overall, the 63-days breeding season had 13 days with daily temperatures greater than 10  $^{\circ}$  above normal and 37 days with daily high temperatures greater than 90  $^{\circ}$ F. Day 7 through 24 was an extended period of higher than normal ambient temperatures, with daily-highs above 88  $^{\circ}$ F (88–101  $^{\circ}$ F) and minimal nighttime cooling (65–75  $^{\circ}$ F). From Days 32–35, a 3-day period of marked above normal temperatures occurred consecutively as well as a 6-day period from Days 47–52. Precipitation was 6 in. below normal for the entire breeding season.

As a subjective observation, native-grass and cool-season grass pasture conditions deteriorated through the breeding season and forage availability was reduced due to the extended dry-period commencing the previous year. Herds on irrigated pastures (HRD1 and 2) had access to more readily available forage of potentially higher nutritive quality. Bulls in HRD3 had access to native forages only throughout the breeding season. The forage quality of the pastures where the NUB bulls were maintained was similarly influenced by the climatic conditions, however, supplemental feed was provided late in the season.

#### 4. Discussion

Results from this study reveal that yearling beef bulls of uniform age and similar prebreeding physical and reproductive attributes experience considerable changes during natural mating and within seasonal environmental conditions. Loss of body weight and condition during mating is a consistent finding in other studies and was attributed to such factors as; pre-breeding physical condition, excessive physical activity during mating and reduced nutritional intakes, particularly during the early season when the mating loads are heavier [12,29,33]. Exposure of the UFB bulls to mating and the associated increased physical activity contributed to marked physical changes, whereas, the NUB bulls, under similar environmental conditions, maintained physical condition as expected. UFB bulls incurred higher BW losses during the early portions of mating, demonstrating rapid physical changes when mating activity would be expected to be highest. Environmental conditions and available nutrition from the breeding pastures may have contributed to the physical changes.

In conjunction with physical changes, changes in reproductive characteristics in both the UFB and NUB bulls were revealed. The declines in SC in UFB bulls, from the initial BSE and prior to any mating exposure, have been similarly reported in yearling bulls following performance testing [34]. Changes of this nature have been attributed to seasonal effects of increasing ambient temperatures [35]. Loss of scrotal fat depositions associated with body condition declines and stress-induced depression of testicular function have also been incriminated as contributing factors for SC declines following environmental and management changes [17,19]. The positive association of SC declines with BW losses, in conjunction with the finding of increases in SC in the NUB bulls with BW gains, demonstrates the linkage of SC to body condition. Although bulls with heavier prebreeding body weights lost higher percentages of BW and SC, no assessment of differences in scrotal fat was undertaken to evaluate the association of body weight with scrotal fat content and possible relationships with SC changes. Body weight losses in young bulls would be indicative of physiological stress and insufficient nutritional intakes to meet growth and activity demands, both of which could lead to stress-induced testicular changes. Seasonal effects, scrotal fat declines during periods of BW declines, and stress-induced effects are all potential explanations for SC changes as demonstrated in this study.

The magnitude of physical and reproductive injury rates in UFB bulls was unexpected. Prevalence rates of injuries to bulls during mating are undocumented, however, the risks and occurrence of injury to yearling bulls are not unexpected. Reasons for the high

incidence in this study are unclear and only limited evidence of association to prebreeding physical characteristics was revealed. Overall, no single phenotypic or genotypic trait was demonstrated to be associated with injury occurence, however, small and unequal sample groups limited expression of significant findings. Breeding herd, bullgroup and sire-breed line differences were evident. The severity of injury was higher during the early mating periods and visual observations would suggest inexperienced mating ability and over-aggressive sexual and social behaviors as contributing to injury events. Injuries during the later mating periods, although occurring at higher rates, were less severe and could have been related to physical fatigue and stress. No clear explanation for the phenomenon was revealed. Injuries incurred during the mating season did influence the availability of physically sound bulls within mating periods, however, no significant differences in bull-group pregnancy rates were demonstrated. Pregnancy rate similarities were probably sustained with the expanded bull-to-female mating ratios afforded by the rotational mating design and less than heavy mating loads. Individual bull reproductive performance was undoubtedly altered, specifically in bulls removed early from the mating season.

The negative changes in seminal quality, revealed in individual bulls following the mating season were primarily associated with increases in abnormal spermatozoa. Sperm motility changes were highly associated to morphological abnormalities. The demonstration of these seminal changes illustrates the potential for change in fertility of yearling bulls during mating conditions. Similar changes in seminal quality have been reported in newly purchased bulls following acquisition and environmental and management changes [19] and in bulls with induced temperature and stress conditions [18,21]. In this study, declining reproductive potential of nearly 50% of the individual UFB bulls was demonstrated with findings of reduced spermatozoal motility and marked increases in both primary and secondary abnormal spermatozoa. Loss of body weight and declines in SC were not significantly associated with the seminal changes. However, UFB bulls classified as unsatisfactory had significantly larger declines of SC, which suggests testicular changes contributing to reduced semen quality. The demonstration of similar seminal quality changes in NUB bulls with prior satisfactory classification, in conjunction with BW and SC increases, suggests a common factor other than physical condition changes as influencing aberrant sperm production in both UFB and NUB bulls. High climatic temperatures, particularly during the early periods of the mating season, potentially inflicted adverse effects on spermatozoal production. This was the principal factor common for bulls used for mating and those not exposed. The effect of climate on nutritional conditions of pastures was not assessed to determine any added effects. Both UFB and NUB bulls demonstrated spermatozoal abnormalities consistent with climatic temperature-induced testicular dysfunction [21].

In summary, although body weight and condition declines were not different between UFB bulls classified as satisfactory or unsatisfactory, the occurrence of weight loss during mating activity was indicative of physiologic stress. Rapid weight loss during the early mating periods, in association with high mating activity and elevated climatic temperatures imposed a compounding of stressors upon testicular function and sperm production. Furthermore, the significant negative influence of injury on seminal quality parameters and BSE-classification indicates the potential effects associated with pain and stress related to

the injuries. Bulls with induced stress conditions and injuries [17,18,21] have demonstrated similar seminal changes as revealed in this study. Individual bulls demonstrated variable responses to physiological and environmental stressors and seminal characteristics were influenced in similarly variable degrees.

Fertility-potential of yearling beef bulls is dependent on the interaction and relationships of multiple physical, reproductive and behavioral factors. In addition, the environmental and management conditions imposed upon young bulls may favorably or unfavorably influence reproductive capability. Within the conditions of this study, yearling beef bulls of satisfactory fertility-potential prior to a mating season incurred marked physical and reproductive changes from the physiologic stress of mating activity, adverse environmental conditions and effects of injuries. The breeding soundness status of nearly 50% of the bulls declined through the mating season to standards considered less than satisfactory for expected mating performance. However, reproductive performance within the breeding herd was sustained at acceptable levels due to sufficient fertility of individual bulls through the course of the mating season and moderate mating loads. Undoubtedly, individual bulls contributed differently to the herd reproductive results due to the altered physical and reproductive characteristics and variable responses to the factors identified as affecting fertility. Further investigation will elucidate individual bull reproductive performance under the conditions imposed by this study and individual bull responses to the environmental impacts.

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