









# final report

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## Bullpower

# Delivery of adequate normal sperm to site of fertilisation

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## ABSTRACT

#### NAP3.117 Bullpower: Delivery of adequate sperm to the site of fertilisation

The project examined the effects of relocation on bull reproductive traits, sexual development in yearling bulls, the effect of yearling mating on pregnancy and calving rates, causes of preputial prolapse in bulls, the importance of sperm morphology as a reproductive trait and the effect of herd dispersion and bull mating percentages on pregnancy rates.

There was no effect of genotype, concentrate feeding or relocation per se, on semen traits.

Mating yearling *Bos indicus* bulls reduced pregnancy rates. Physical but not semen traits of yearling bulls were indicative of these traits at 2 years of age. Supplementing yearling bulls with concentrates did not affect semen traits.

Repeatability of normal sperm was high once bulls reached sexual maturity. Pregnancy rates were reduced using bulls with less than 50% normal sperm. The use of 2.5% reproductively-sound bulls appears adequate under most conditions.

Predisposition to preputial prolapse was equivocal.

Recommendations for research included further investigation on the effects of relocation; defining threshold levels for high energy diets for yearling bulls; methods to increase the proportion of yearling bulls passing a BBSE; determining heritability and genetic relationships of male reproductive traits; and researching cattle reproductive behaviour to explain differences in calf output of bulls.

## EXECUTIVE SUMMARY

#### NAP3.117 Bullpower: Delivery of adequate sperm to the site of fertilisation

Bulls are a major cost for commercial cattle breeders in northern Australia. The calf output of individual bulls in multiple-sire herds is extremely variable. Previous studies in northern Australian herds found that 14% of the bulls sired over 30% of calves whilst 58% of bulls individually sired 10% or less in their mating groups. A further 6% of bulls did not sire any calves.

The first Bullpower project (DAQ. 104) related pre-mating measures of physical, seminal and behavioural traits to calf output in *Bos indicus* derived bulls. Sheath and testicular traits, such as scrotal circumference and testicular tone, dominance hierarchy and sexual behaviour in serving capacity tests were generally not related to calf output. The strongest associations with calf output were with semen quality, mainly sperm morphology. However these traits only explained 35-57% of the variation in calf output. Other factors, such as bull behaviour, may affect calf output.

If sperm morphology is an important determinant of calf output, then delivery of adequate normal sperm to the site of fertilisation may be critical to maximise calf output of bulls of desired genotype. There are key reasons why bulls may not be able to provide adequate normal sperm to the site of fertilisation: viz

- Bulls have poor semen quality. This may be permanent or temporary. In bulls with permanently poor semen quality, the condition may be innate or acquired. In the majority of cases, the reason for this is not apparent but could be an inability to adapt to the environment. Temporary reductions in semen quality may be from stress, relocation, poor environmental adaptation, obesity and disease.
- Bulls have good semen quality but have suppressed mating ability through social dominance, lack of sexual motivation or impaired mating ability due to mechanical impediments such as lameness, serving ability or sheath problems.
- Herd dispersion effects may restrict bulls that are physically and reproductively sound having contact with females thus reducing conception rates and with more calves born later in the calving season.

The objectives of the project were to quantify the impact of relocation on subsequent bull fertility; identifying traits bulls can be selected on for fertility at an early age; identifying traits that predispose bulls to early breakdowns in their working life; developing knowledge of sexual behaviour of cattle; defining bull mating percentage on conception rates at different levels of herd dispersion; and delivering useful, practical information to industry

The main findings of the project were as follows:

- The detailed experimental studies found that there were minimal effects of genotype, concentrate feeding prior to relocation or relocation *per se*, on bull semen traits or sperm morphology either in the short term (1.5 3 months) or long term (12 months) post-relocation. In these experimental studies bulls were relocated under favourable conditions. Bulls were yarded overnight prior to relocation; the transport time was minimised, bulls were familiar with each other and then grazed good pasture post-relocation. However the observational studies on relocation effects on bull reproductive traits found that about 50% of bulls failed a bull breeding soundness examination (BBSE) up to 3 months after sale. The causes of these failures to pass a BBSE were not established but appeared unrelated to genotype, age, property of origin or prior nutrition. Our inference is that many bulls in the industry are being relocated and managed post-relocation under less than ideal conditions. This may have an adverse impact on the reproductive performance of cattle in the northern industry.
- Many yearling bulls had not reached puberty by 14 months of age thus few will pass a BBSE at this age. The average age and range of puberty in Brahman and Composite bulls was 17.4 (13.5 24.0) months and 15.4 (12.5-18.5) months respectively. At 14 months of age, 12 to 18% of *Bos indicus* bulls produced semen with at least 50% normal sperm whilst in Composites it was 59 to 89%. Producers can expect reduced pregnancy rates if *Bos indicus* bulls are mated as yearlings. However

limited data indicate that this reduction in pregnancy rates will not occur if yearling Composite bulls are used.

- Physical traits (liveweight, scrotal circumference, sheath depth, umbilical thickness and rosette), measured in yearling bulls reflected these traits when these bulls were 2-year-olds. However the repeatabilities for semen traits and sperm morphology were low, indicating that semen and sperm morphological assessments in yearling bulls are not accurate predictors of these traits in 2-year-old bulls.
- Supplementing yearling bulls with moderate levels of concentrates had a variable effect on scrotal circumference but there was no effect on semen quality or sperm morphology. However with concentrate feeding, there is a risk of lameness from acidosis, flight speeds increased and the advantages in liveweight and scrotal circumference were almost eroded 12 months later.
- The predisposing factors for preputial eversion may differ between *Bos indicus* derived breeds and *Bos taurus* breeds. There was no evidence of poorer preputial muscle development or more preputial eversion in polled than in horned Santa Gertrudis bulls. Preputial prolapse in polled Santa Gertrudis bulls could be due to the significantly reduced size and development of the preputial muscle complex. No obvious cause of prolapse in horned bulls could be determined from dissections. Preputial eversion in Droughtmaster bulls was related to the position of the penis within the sheath.
- Normal sperm was poorly repeatable in bulls that were still sexually maturing (14 through to 24 months of age). However the repeatability of normal sperm was high in Brahman and Composite 2-to 3-year old bulls, which were sexually mature.
- Single-sire studies demonstrated that pregnancy rates were reduced when bulls with less than 50% normal sperm were used, particularly under high mating loads.
- Herd dispersion studies showed that pregnancy rates were not reduced as a direct result of reducing bull percentages from high to moderate under high dispersion grazing management in north Australia, or from moderate to low under low or moderate herd dispersion. The data provided further support to the recommendation that using mating percentages of 2.5% reproductively-sound bulls are adequate under most north Australian cattle management conditions.

A number of recommendations for further research are made. Some of these included:

- Further research would more precisely quantify the effects of relocation on bull reproductive traits. Bulls exposed to larger insults such as relocation through sale yards should be measured.
- Further research on the effects of high-energy diets on physical, behavioural and semen traits would define thresholds for levels and duration of feeding.
- Further research may develop efficient ways to improve the proportion of *Bos indicus* and *Bos indicus* derived bulls that pass a BBSE at 14 months of age.
- Further research would allow the estimation of the heritability of male reproductive traits and the genetic relationships between these traits and other productive traits including female fertility.
- Herd dispersion effects need to be investigated using detailed behavioural research in controlled situations.
- Research into cattle reproductive behaviour may provide some valuable insights for developing more efficient and profitable mating management of bulls.

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## MAIN RESEARCH REPORT

## 1 Project team

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## 3 Background and Industry Context

Bulls are a major cost for commercial cattle breeders in northern Australia. The economic impact of bull management depends on two issues. The first is the capital cost per calf branded, incurred when buying the bull. This is a function of the purchase price of the bull, and the number of calves sired during its working life. The second is the genes contributed by the bull to its progeny. Major financial benefits are achieved by using bulls whose progeny have higher productivity through improved fertility, growth, temperament, survival and carcase attributes.

The calf output of individual bulls in multiple-sire herds is extremely variable. In a study of 235 bulls in 9 northern Australian herds from 37 multiple-sire mating groups, Holroyd *et al.* (2002a) found that 14% of the bulls sired over 30% of calves whilst 58% of bulls individually sired 10% or less in each of their mating groups. A further 6% of bulls did not sire any calves. All of these bulls had passed a breeding soundness examination. These findings on variability of calf output of bulls in multiple sire herds have been supported from other studies in Israel (Lehrer *et al.*, 1977), in Canada (Makarechian and Farid, 1985; Coulter and Kozub, 1989) and from the Northern Territory (McCosker *et al.*, 1989). The data of Holroyd *et al.* (2002a) also demonstrated the calf output of individual bulls was moderately repeatable across years.

Having obtained the appropriate genotype for particular markets through either investment in replacement bulls or breeding their own bulls, producers need to maximise the number of progeny from bulls that have been selected for superior genetic merit. Therefore producers and veterinarians need to know the physical, behavioural and reproductive traits that may limit a bull's ability to sire progeny or that can be used as predictors of calf output of individual bulls in multiple-sire herds.

In the study of Holroyd *et al.* (2002a), multiple regression models relating pre-mating measures of physical, seminal and behavioural traits to calf output were developed in 40 Santa Gertrudis, 24 5/8 Brahman and 74 Brahman bulls. Sheath and testicular traits, such as scrotal circumference and testicular tone, were generally not related to calf output. Dominance was only included in the 5/8 Brahman model but there was no significant relationship between dominance hierarchy and calf output. Some measures of sexual behaviour in the serving capacity test were related to calf output in Santa Gertrudis; these were for the number of displays of sexual interest and mounts but not number of serves, whilst in Brahman bulls, libido score was positively related to calf output. Semen motility was only related to calf output in 5/8 Brahmans. The strongest associations with calf output occurred with measures of semen quality based on spermatozoa morphology. However the models only explained 35% to 57% of the variation in calf output. Other factors, such as the behaviour of bulls in paddocks, may have been contributing to calf output.

If sperm morphology is an important determinant of calf output of individual bulls, then the delivery of adequate normal sperm to the site of fertilisation may be critical to maximise calf output of bulls of desired genotype. There are a number of reasons why bulls cannot provide adequate normal sperm at the site of fertilisation.

- Bulls may have poor semen quality. This may be permanent or temporary. In bulls with permanently poor semen quality, the condition may be innate or acquired. In the majority of cases, the reason for the condition is not apparent but could be an inability to adapt to the environment. Temporary reductions in semen quality may be from stress, relocation, lack of environmental adaptation, obesity and disease.
- Bulls with good semen quality may not be able to maximise their calf output. This may be from suppressed mating ability through behavioural influences such as social dominance or lack of sexual motivation or through impaired mating ability due to physical defects such as lameness, serving ability or sheath problems.
- *Herd dispersion effects* may limit bulls having contact with females in oestrus thus reducing conception rates and more calves tend to be born later in the calving season, despite using bulls that are physically and reproductively sound.

The Bullpower project addressed the issue of delivery of adequate normal sperm to the site of fertilisation by studying 6 areas concurrently.

*Investigating the effects of relocation on bull fertility*. Our studies defined the prevalence and severity of changes in various bull reproductive traits following relocation and factors associated with these changes.

*Increasing the working life of a bull.* Our studies concentrated on age of puberty, identifying criteria whereby bulls can be selected for reproductive traits at an early age, evaluating yearling mating and identifying traits that predispose bulls to preputial prolapse.

**Quantifying the role of sperm morphology as a criterion for bull selection and calf output**. Studies further investigated the repeatability of sperm morphology in both immature and mature bulls. Matings of bulls with low values of morphologically normal sperm were conducted, as single-sires, to provide further evidence of the detrimental effect on reproductive rates of mating bulls with low normal sperm counts.

*Herd dispersion effects and mating ratios on herd fertility*. Bull mating ratios were tested under a range of dispersion situations to extend earlier studies that reduced bull percentages without decreasing herd fertility rates.

A literature review of factors influencing beef cattle libido and paternity. Most information on sexual behaviour in cattle relates to animals in confined areas such as serving capacity tests, with limited information published on the sexual behaviour of rangeland cattle. The main benefit of this review was to identify gaps in our knowledge and, thus, provide a sound basis on which to recommend behavioural research in beef cattle.

**Delivery of useful, practical information.** Widespread and coordinated extension of accurate and consistent information was undertaken for all industry sectors, recognising the different requirements for producers, extension officers, veterinarians and other industry-support personnel.

## 4 **Project Objectives**

The objectives of the project were to develop management strategies that will maximise the calf output of bulls of desired genotypes in northern Australian herds by:

- Quantifying the impact of relocation on subsequent bull fertility;
- Identifying traits whereby bulls can be selected for fertility at an early age;
- Identifying traits that predispose bulls to early breakdowns in their working life;
- Developing knowledge of the sexual behaviour of cattle;
- Defining the effects of bull percentages on conception patterns and pregnancy rates at different levels of herd dispersion; and
- Delivering useful, practical information to industry on bull evaluation, selection and management.

## 5 Description of project and common materials and methods

The project ran from March 2000 to June 2004. Some prior activities in aspects of the work are included in this report to enhance activities included in the contract with MLA. The collection of field data was completed by November 2003. Completion of laboratory analyses of semen morphology was achieved by March 2004 and the statistical analyses by May 2004.

The components of the project were a survey, a series of experiments conducted on research stations and commercial properties throughout Queensland, a literature review, involvement of staff in field days, demonstrations, workshops and reporting on progress through conferences, press releases and journal articles. A breakdown of the surveys and experimental activities is listed in Table 1. Section 6.7 summarises activities associated with delivery of information.

#### 5.1 Project activities

Investigating the effects of relocation on bull fertility	
A survey of veterinary practitioners, insurance companies and pastoral companies on the Jan 01 – Dec 01 effects of relocation on bull fertility	l
Breeding soundness of sale bulls after relocation - Charters Towers, North Queensland Jun 00 – Jun 02	2
The effect of feeding and relocation on reproductive traits of Brahman bulls - Brigalow and Aug 00 – Nov 03 Toorak Research Stations	3
The effect of relocation on reproductive traits of Brahman and Composite bulls - Belmont Jul 01 – Nov 03 and Swan's Lagoon Research Stations	3
Increasing the working life of bulls - Identifying criteria to select bulls for fertility at a young age and evaluation of yearling mating	1
Sexual development in Brahman and Composite bulls - Belmont Research Station Nov 99 – Aug 00	)
Sexual development in Santa Gertrudis bulls – Gyranda Aug 00 – Aug 01	I
Sexual development in Droughtmaster bulls – Dilga Sep 00 – Aug 01	I
Fertility of 5/8 Brahman yearling bulls, Swan's Lagoon Research Station May 00 – Jun 01	I
Sexual development in Belmont Red bulls – Narayen Research Station Nov 95 – Aug 99	9
Increasing the working life of bulls – Traits that predispose bulls to preputial prolapse	
Survey of reasons for culling bulls Apr 98 – May 99	)
A quantitative anatomical study of the sheath and prepuce of Santa Gertrudis bulls in Mar 98 – Aug 98 relation to the occurrence of preputial eversion	3
An anatomical study of chronic preputial prolapse of Santa Gertrudis bulls Mar 99 – Aug 99	)
Relationship between position of the penis and preputial eversion in <i>Bos indicus</i> derived Mar 01 – Dec 01 bulls	I
Quantifying the role of sperm morphology as a criterion for bull selection and calf output	
Longitudinal study on semen and sperm traits of bulls - Belmont Research Station. Oct 99 – Nov 01	I
Pregnancy rates achieved by mating bulls with different percentages of morphologically Jan 99 – May 02 normal sperm – Swan's Lagoon Research Station	2
Herd dispersion effects and mating ratios on herd fertility	
Herd dispersion and mating ratio effects on conception patterns Oct 00 – Sep 02	2
Factors influencing beef cattle libido and paternity	
A literature review of factors influencing beef cattle libido and paternity Jun 01 – Jul 03	3

#### 5.2 Glossary of terms

AACV	Australian Association of Cattle Veterinarians
BBSE	Bull breeding soundness examination
Fertility	This is mainly used in the context of number of calves sired by a bull (i.e. calf output)
Libido	Sexual motivation as demonstrated through mate seeking, courtship and copulation
Serving capacity test	A test of sexual motivation. See section 5.3 for description
Sexual behaviour	Includes interest, mounts and serves as recorded in a serving capacity test

#### 5.3 Animal ethics committee approvals

The experiments had the following animal ethics approvals; TSV/06/99, TSV/23/00, TSV/31/00, TSV/48/01, TBC 115, TBC 118, TBC 127, CEN 7, STHLAEC/03/00 and FAS/467/97/MRC

#### 5.4 *Physical and reproductive examinations of bulls*

Bulls were subjected to a general physical and reproductive examination based on McGowan *et al.* (2002). Liveweight was measured as full weights. Body condition was visually scored using a 9-point scale (Holroyd 1985), where 1 is emaciated and 9 is overfat, with most bulls being in the range of 5-7.

Bulls were restrained in a veterinary crush to enable a general physical examination of the musculoskeletal system and a detailed physical examination on the external genitalia. If, after semen collection, there was a suspected problem with internal genitalia, then a rectal examination was conducted. The musculoskeletal system was visually assessed and abnormalities of the legs and feet such as straight hocks or scissor claws recorded as being either mild or marked. The scrotal contents were assessed visually and by manual palpation, and obvious abnormalities of the scrotum, testes or epididymes were recorded. Testicular tone was a manual subjective assessment based on Holroyd et al. (2002b) with a combined value for firmness and resilience. Scores were on a 5-point scale with 1 = very soft, 5 = very hard with a desirable tone in the 3-4 range.

The sheath and prepuce were assessed visually for abnormalities then measured. The penis was palpated through the sheath. Sheath and umbilical measurements were based on McGowan *et al.* (2002) and were measured in most but not all bulls. Measurements included:

- Sheath depth: vertical distance in cm from the ventral abdominal wall to the preputial orifice.
- Umbilical cord thickness: estimated diameter of the remnant of the umbilical cord midway along the vertical line for sheath depth. It was scored from 1 to 5 with 1, 0.5 cm or less, 3, 1.5 cm and 5, ≥ 2.5 cm.
- Navel or rosette score: This was an estimate of the size of the external umbilical scar including the surrounding fold of skin. Scoring was 0: absent, 1: small (<2 cm diameter), 2: medium (2-5 cm diameter) and 3: large (>5 cm diameter).
- Sheath score: this was the Breedplan score ranging from 1: very pendulous to 9: very tight and close to the ventral abdominal wall.

Semen was collected using standard electroejaculation techniques (Entwistle and Fordyce 2003). If a satisfactory sample could not be collected within several minutes, attempts were then made by massage of the ampullae. If a satisfactory sample could still not be collected, then a second attempt was made on some of these bulls later that day. Bulls were released from the crush and overall structural conformation assessed with the bull standing and then gait examined either at the walk or trot

Evaluation of semen was based on the procedures of Fitzpatrick *et al.* (2002) and Entwistle and Fordyce (2003) with the sample being assessed crush-side for mass activity, colour, density and progressive

individual forward motility (motility). Mass activity had a scale of 0-5 where 0 = no swirl, 3 = slow distinct swirl and 5 = fast distinct swirl with continuous dark waves. Colour had a scale of 1-5 where 1 = clear, 3 = milky and 5 = thick creamy to yellow. Density had a scale of 1-5 where 1 =  $<200 \times 10^6$  sperm/mL, 3 =  $500 - 1000 \times 10^6$  sperm/mL and 5 =  $1500 + \times 10^6$  sperm/mL. Motility was recorded as a percentage.

The morphology of 100 sperm was determined in the laboratory by examining a thin cover-slip preparation of semen preserved in 0.2% gluteraldehyde in phosphate buffered saline using phase contrast microscopy (x1000). Sperm were classified individually, then allocated into 1 of 5 categories, normal morphology, abnormal head, abnormal midpiece, abnormal tail or having a cytoplasmic (either proximal or distal) droplet. In the latter part of the project, some morphological assessments used the new AACV classification (Entwistle and Fordyce, 2003) based on functionality, *viz.* normal, proximal cytoplasmic droplets (PD), midpiece abnormalities (MP), tail defects and loose heads (T&H), pyriform heads (Py), knobbed acrosomes (KA), vacuoles and teratoids (V&T) and swollen acrosomes (SA).

Measurement of sperm concentration was done in a 1 in 60 dilution of semen prepared by adding  $50\mu$ L of fresh semen to 2.95 mL of 0.2% glutaraldehyde in phosphate buffered saline. The counting chamber of a Neubauer haemocytometer was loaded using a glass pasteur pipette and left to rest for 2 minutes to allow cells to settle before counting.

Bulls were serving capacity tested, based on the procedures of Bertram *et al.* (2002), in an approximately rectangular yard (dimensions varied from (12 x 7 m) over a 20-minute period using restrained females. Bulls had not been mated prior to the first test. Females were not synchronised for oestrus. During all serving capacity tests, bulls were pre-stimulated by allowing them to watch mounting behaviour of other bulls undergoing the test. The number of times each bull displayed interest, mounts and serves were recorded. Interest was defined as any form of sexual interest including flehmen, licking and false mounts. A mount was any combination of mounting with penile seeking including intromission and a serve was a mount, followed by intromission and an ejaculatory thrust. Sexual behaviours recorded in each test were transformed to a libido score of 0-10 (Table 2) based on the work of Chenoweth (1981).

Table 2. Method of calculating libido scores										
Libido score <sup>#</sup>	Interest	Mounts	Serves							
	n	n	n							
0	0	0	0							
1	1	0	0							
2	2-5	0	0							
3	>5	0	0							
4		1	0							
5		2	0							
6		>2	0							
7		0-5	1							
8		>5	1							
9		0-5	2							
10		>5	>2							

Table 2. Method of calculating libido scores

<sup>#</sup> eg. to qualify for a libido score of 7, bulls had up to 5 mounts and 1 serve

#### 5.5 Statistical analyses

Statistical analyses are described in each section. Levels of repeatabilities are described as low when <0.33, moderate, 0.33-0.66 and high when >0.66.

## 6 Results and Discussion

#### 6.1 The effects of relocation on bull reproductive traits

#### 6.1.1 Background

Many herd bulls used in the northern Australian cattle industry originate from bull breeding herds in environments different to the property of use. McCool and Holroyd (1993) reported that a proportion of bulls were sub-fertile in their first season after being relocated. There have been cases where up to 40% of large drafts of bulls have failed a standard bull breeding soundness examination (BBSE) within 3 months of delivery to Northern Territory properties after being purchased at central Queensland sales. This depression in fertility of bulls is generally not recognised until bulls have been resident on their new property for 12 months or more and it may be too late to remedy.

Relocation-induced depressed fertility (RDF) refers to the depression in fertility that has been observed in bulls after their relocation to a new environment. In the course of relocation, bulls may be transported many hundreds or even thousands of kilometres, through extremes of hot and cold weather. During transport, they may be subjected to irregular feed and water supply, crowding and physical trauma. On the property of destination, they often face a very different environment to their property of origin in terms of forage quantity and quality, water quality and climate. As well they may be faced with intense competition from existing bulls, different mustering and handling procedures and exposure to a range of pathogens new to the bulls (McCool and Holroyd, 1993).

Infertility or sub-fertility can operate via several mechanisms during the relocation process itself, or in the new environment soon after arrival. These mechanisms may be traumatic, physiological, adaptational or of disease origin. Barth (1991) reported that transport-related stress probably operates in a similar manner to that experienced from elevated testicular temperature and acts via endocrine mechanisms to cause testicular changes and thus influencing the proportion of different morphological abnormalities appearing in the ejaculate.

RDF is a topical industry issue because it may represent a significant cost to industry through inefficient bull use, reduced conception rates, delayed calving dates, depressed branding rates and accelerated bull wastage. However the prevalence and severity of the syndrome is not substantiated by comprehensive factual data. Fertility failure of relocated bulls in the first 12 months after purchase was one of the major issues in bull fertility identified by producers in a series of recent "Breeding Better Bulls' days in north Queensland (Beef Genetic Improvement Project report). However, RDF is not perceived as a significant problem in southern Queensland (J Bertram, pers com). RDF has been mainly associated with bulls that have been relocated to more stressful environments and accompanying long-distance transport (eg from central Queensland to north Queensland or Northern Territory) but recent feedback from a number of veterinary practitioners and breed societies would suggest that RDF is happening in relatively benign environments and after short-distance transport.

Our general aims were to examine several hypotheses relating to relocation viz:

- The prevalence and severity of RDF is a function of duration, distance and process of relocation as well as the environmental differences between the properties of origin and destination.
- RDF is only an issue within the first 12 months after relocation.
- Management strategies can be identified to prevent or minimise RDF.

Our research strategies involved 3 areas of work. Firstly a survey of veterinary practitioners and representatives from insurance and pastoral companies was undertaken to further clarify the perceptions of the effect of relocation on bull reproductive performance. Secondly a study was undertaken of bulls sold and then moved to north Queensland to obtain better data on perceived problems associated with relocation. Thirdly, two experiments where bulls were monitored prior to and post- relocation, were undertaken.

# 6.1.2 A survey of veterinary practitioners, insurance companies and pastoral companies on the effects of relocation on bull reproductive traits

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#### 6.1.2.1 Aim

The aim of the survey was to summarise the perceptions of a group of respondents to depressed fertility in bulls associated with relocation.

#### 6.1.2.2 Materials and methods

A survey was conducted by John Bertram who telephoned a total of 30 veterinarians, insurance and pastoral company representatives between January and February 2001. These people were closely involved in the veterinary examination of bulls in the northern Australian beef industry for fertility assessments, purchase or insurance. An introductory phone call discussed the intent of the survey, identified the respondent's level of cooperation and arranged for faxing of the survey prior to phoning back and completing the questionnaire (Appendix 1) over the telephone. Eleven questions were asked allowing for discrete responses in combination with subjective information.

The respondents by business and region are listed in Table 3. The results for each question were analysed using frequency tables or calculating means as appropriate with some cross-tabulations of questions against regions. Because of the small sample sizes within regions, caution should be taken in interpreting the results for cross-tabulations.

		U		
Region	Insurance Co.	Veterinarians	Pastoral Co.	Total
Kunnunura	0	1	1	2
North Queensland	0	7	2	9
Central Queensland	0	6	0	6
Western Queensland	0	3	0	3
Southern Queensland	1	8	0	9
Unknown	1	0	0	1
Total	2	25	3	30

Table 3. Distribution of respondents by business and region

#### 6.1.2.3 Results

Respondents readily provided their attitudes and perceptions regarding RDF. Many believed there was a problem but found it difficult to quantify in detail the extent and magnitude of the problem as the majority of bulls examined post-relocation were seen many months or years following the date of transport.

The main findings of the survey were as follows:

- 24 (80%) respondents believe reduced fertility sometimes followed relocation.
- The majority (63%) of respondents formed their opinions about the effects of relocation based partly on post relocation fertility evaluations.
- Reduced fertility was most frequently identified as occurring after relocating to a lower plane of nutrition (61%) or relocating to a hotter district (54%).
- 79% had encountered reduced fertility in either or both *Bos indicus* or *Bos indicus* cross whilst 54% identified *Bos taurus* breeds.
- 79% of respondents indicated that depressed fertility was more prevalent in bulls that had been supplementary fed grain in self-feeders and/or bulls raised in feedlots on predominately grain rations prior to relocation.
- 85% (22 out of 26) of respondents identified fat bulls as having a fertility problem.
- Scrotal circumference, testicular tone, skeletal structure and semen quality were recorded by at least 75% of those conducting BBSE.
- Many practitioners were not comfortable with examining bulls for structural soundness and left this aspect to the stud manager rather than commenting on the BBSE sheet.
- Attributes seen as important in influencing bull fertility were scrotal circumference (90% of respondents), normal sperm (79%), mass activity of semen, progressively motile sperm and mating ability (all 66%), skeletal structure (59%) and testicular tone (52%). Of these traits, mating ability is the one attribute that is currently not being recorded in a BBSE.
- Many practitioners used their own or variations of the AACV BBSE examination forms (either individual or multiples per page) leading to some inconsistency in reporting of fertility and producer understanding and interpretation.
- One respondent claimed he used a microscope infrequently for crush side examination of semen prior to sending a sample to a laboratory for analysis of sperm morphology.
- Both of the respondents from insurance companies were not familiar with the potential requirements and benefits of a full BBSE; and even less aware of the cost of a BBSE.

#### 6.1.2.4 Conclusion

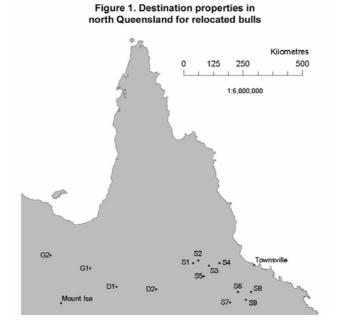
This survey indicated that veterinary practitioners have accepted some of the recommendations from Bullpower 1 (DAQ.104); e.g. 79% believe normal sperm is an important influence on bull fertility and record this in a BBSE. Conversely, 66% believe that mating ability is also important but only 21% record mating ability in a BBSE.

The results of this survey indicate there is an opportunity for the AACV to promote a better understanding by veterinary practitioners of the important bull reproductive traits that reduce branding rates.

- Relocation induces sub-fertility in bulls.
- The prevalence and severity of relocation-induced sub-fertility in bulls is primarily a function of body condition, genotype, age of bulls and the environmental and management differences between the properties of origin and destination.
- Relocation-induced temporary sub-fertility does not extend longer than 6 months.

#### Study design

Between mid-2000 and mid-2002, bulls introduced onto properties in north Queensland were classified according to genotype, history, source and destination (Figure 1, Table 4). Table 5 lists the 13 properties of the 11 beef producers who co-operated in the study. At all assessments, a BBSE excluding serving assessment was carried out. Bulls were assessed as soon as possible after sale and relocation. Where possible, they were reassessed at various times in the 18 months after relocation.



	Abbreviation	Description
Genotype	Bosl	Bos indicus
	Trln	Bos taurus x Bos indicus
	BosT	Bos taurus
History	GrnF	>20 months of age; partial or full concentrate feeding
	PdkF	>20 months of age; paddock reared
Source	Temp	Temperate region
	SubT	Sub-tropical region
	Trop	Tropical region
Destination	Gulf	Savannas of the Gulf region
	SpGr	Savannas predominated by spear grass (Heteropogon contortus)
	Downs	Black-soil plains with few trees, south of the Gulf region

Genotype		Bosl	Bosl	Bosl	Bosl	Trln	Trln	Trln	Trln	BosT	BosT	BosT	
History		GrnF	GrnF	PdkF	PdkF	GrnF	GrnF	PdkF	PdkF	GrnF	GrnF	PdkF	
Source		SubT	Trop	SubT	Trop	SubT	Trop	SubT	Trop	Temp	SubT	SubT	
Destination	Date	#											Total
G1	08-Sep-00								44				44
	02-Nov-00								135				135
G2	05-Dec-00	22	28	57									107
	03-Jul-01	18	23	46									87
D1	07-Jun-00	30	11		1	2	9						53
	07-Dec-00	23	5			1	4						33
D2	07-Jun-00					1	8	1			18	5	33
	07-Dec-00					1	6	1			17	4	29
S1	22-Aug-00				25					56			81
S2	26-Oct-00	2	4										6
	21-Jan-01	2	4										6
S3	26-Oct-00	3											3
S4	26-Oct-00	2									7		9
	21-Jan-01	2									7		9
	28-May-01	2									7		9
S5	28-Sep-00				9		2						11
	22-Dec-00				10		2						12
	03-Aug-01				9								9
	04-Jan-02				9		1						10
	04-Jan-02		14		35								49
S6	29-Oct-00		3										3
S7	29-Oct-00	14											14
S8	05-Jan-00											12	12
	03-Feb-00											11	11
	02-Mar-00											11	11
	11-Apr-00											12	12
	17-May-00											11	11
	23-Jun-00											12	12
	19-Jul-00											12	12
	29-Aug-00											12	12
	12-Nov-00											12	12
	14-Jan-01											12	12
	06-Sep-01											10	10
S9	03-Feb-01	13	1								2		16
	03-Aug-01	13	2								2		17
	24-Jan-02	13	1								1		15
	Total	159	96	103	98	5	32	2	179	56	61	136	927

#### Table 5. Numbers of bulls and observations

# See Table 4 for explanation of abbreviations

#### Nutrition effects on bull breeding soundness following relocation

On property S5 (Table 5), 10 Brahmans and 2 Charbrays, all aged 2 years and purchased in July 2000, were assessed 2, 5, 13, and 18 months after purchase in the late dry season (September 2000), early wet season (December 2000), mid-dry season (August 2001), and early wet season (January 2002) respectively. BBSEs were conducted on property S9 (Table 5) on a group of 15 Brahman and 2 Limousin bulls at similar times (February 2001, August 2001, and January 2002). The S9 bulls were between 2 and 7 years of age in February 2001; 10 bulls had been resident on the property for at least 1 year, and 3 and 4 bulls had been purchased 1 and 8 months earlier, respectively.

Both S5 and S9 are in the dry tropics. Following good rains in the 2000/01 wet season, an early, cold (frosts reduce pasture protein content) winter heralded the onset of a long dry season with extended very poor nutrition from early May to November-December 2001 inclusive.

#### Statistical analyses

The primary response variable in analyses was pass/fail BBSE. A major secondary variable was sperm morphology presented as either being continuous (normal sperm), or discontinuous (above/below a threshold value of either 50% or 70% normal sperm, as indicated by Entwistle and Fordyce (2003).

In most cases there was no BBSE report available prior to relocation. Therefore the observation primarily developed a regional perspective on bull soundness post-sale and relocation. At site G1, a BBSE had been conducted on a proportion of the bulls prior to relocation. At site S8, bulls had been part of another sexual development study and a full BBSE prior to relocation was available. Except where further explanation below outlines more detailed analyses, the data collected from each site was simply summarised.

Site G1: Analysis of variance with the bulls as blocks was used to assess the impact of relocation on various traits. A chi-squared test was used to compare the proportion of bulls that passed the BBSE pre- and post-relocation.

Site G2: To assess main effects on the non-binomial variables measured, a mixed-model analysis was used with bull as a random effect and date of measurement, arrival, history, source and birth date as fixed effects. Non-significant fixed effects (except for date of measurement) were removed from the model so the final model contained only significant fixed effects. A generalised linear model (using the same factors as for the mixed-model analyses) with binomial distribution and logit link was used to test the effects of source, date, and the interaction between these on BBSE result (0 = fail, 1 = pass) and the proportion of bulls whose normal sperm values were either above or below the 50% or 70% thresholds.

Sites D1 and D2: The bulls were initially relocated to the one site south of D1 and D2; soon after BBSE they were relocated to either D1 (*Bos taurus* and some Charbray bulls) or D2 (Brahmans and some Charbray bulls). Genotype/source (confounded) differences for non-binomial measurements made at the first assessment post-relocation, were analysed using one way analysis of variance. The effect of genotype on binomial parameters was analysed as for Site G2 data. Analysis of differences between the first and subsequent measurement of Brahmans and of *Bos taurus* bulls used a split-plot analysis of variance with blocking structure of bulls as the whole plots and dates as the subplots. A similar analysis was performed on Charbray bull data, but with property of destination at the whole plot level. For all bulls, a generalised linear model with binomial distribution and logit link was used to analyse BBSE result (0 = fail, 1 = pass) and the proportion of bulls whose normal sperm values were either above or below the 50% or 70% thresholds.

Site S1: The effect of age/relocation date (confounded) on traits of the Angus bulls was tested by oneway analysis of variance. A chi-squared test was used to compare the BBSE of the 3 cohorts. A 2 x 2 chisquared test was applied to test the difference in BBSE between the youngest Angus cohort and the Brahmans that were of the same age.

Sites S2 and S4: An analyses of variance using bull as the blocking term compared measurements made on the 3 post-relocation dates.

Sites S5 and S9: A one-sample t-test was used to compare the mean reduction in variables between the wet season (December 2000 at S5 and February 2001 at S9) and the subsequent dry season (August 2001 at both sites) with a hypothesized value of 0 to assess whether the reduction was significant. A similar test was applied to the increase in variables between the dry season and next wet season (January 2002 at both sites). Repeatability (intra-class correlation) of scrotal circumference and normal sperm was calculated after adjusting for the fixed effects of length of time on property and genotype. Spearman rank correlations were calculated between successive times of measurement for scrotal circumference and normal sperm.

#### 6.1.3.4 Results

#### **Relocation to the tropics**

Results indicate that approximately half of the bulls relocated to commercial herds in north Queensland failed a BBSE in the 6 months after relocation, primarily because their ejaculates contained less than 50% normal sperm. This occurred irrespective of genotype or whether they had been previously fed concentrates (Figure 2, Table 6).

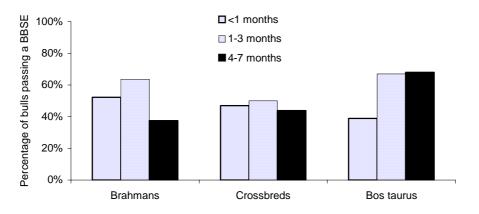


Figure 2. Percentage of bulls passing a BBSE at <1 month, 1-3 months and 4-7 months after relocation

#### Brahman bulls post-relocation

A group of 107 Brahman bulls from multiple sources across Queensland was assembled over 2 months and relocated to property G2 in late 2000. The bulls were evaluated at 1 week and 87 were evaluated 7 months later (Table 7). The bulls were aged 2 and 3 years (60% and 40%, respectively) at relocation. Average weights after relocation of 2-year-old paddock-reared bulls, 2-year-old lot-fed bulls, and 3-year-old lot-fed bulls were 490, 590 and 645 kg, respectively. Scrotal circumference of 3-year-old bulls was larger than for 2-year-old bulls (34.9 v 32.4 cm; P<0.001). Between the 2 assessments, these bulls had been mated as one group. Seasonal conditions were poor. In the 7 months after relocation, the bulls lost 2 units of body conditions score (Table 7). Scrotal circumference was reduced by 0.6 cm. At relocation, a little over half the bulls passed a BBSE, and this reduced to a quarter by the end of mating. There were no significant effects on BBSE parameters due to bull age, previous diet or source.

Gtype	Source	History	Age	n	Body	Testes	Scrot Sperm		N	lormal Spe	mal Sperm	
					condition	tone	circ.	motility	Av.	>50%	>70%	Pass
			yrs		1-9	1-5	cm	%	%	%	%	%
Relocate	ed within p	oast monti	h									
Bosl	SubT	GrnF	1.5	31	6.8	3.3	30.9	12	36	27	7	27
Bosl	SubT	GrnF	2	21	7.7	3.8	34.9	49	50	62	29	57
Bosl	SubT	GrnF	3	21	7.6	3.7	35.7	52	53	65	35	65
Bosl	SubT	PdkF	2	40	6.8	3.5	32.1	45	51	58	20	58
Bosl	Trop	GrnF	3	22	7.8	3.6	35.3	40	49	59	14	64
Bosl	Trop	GrnF	2	13	7.7	3.5	34.3	28	52	46	23	46
Trin	Trop	GrnF	1.5	17	6.7	3.4	34.3	15	49	47	35	47
BosT	SubT	GrnF	1.5	18	6.6	3.3	32.6	13	44	40	13	33
BosT	SubT	PdkF	1.5	5	5.4	3.0	31.9	27	56	60	40	60
Relocate	ed 1-3 mor	nths ago										
Bosl	SubT	PdkF	2	11	7.2	3.7	33.1	46	49	45	18	45
Bosl	SubT	PdkF	3	6	7.5	3.3	35.1	55	46	33	33	33
Bosl	Trop	GrnF	2	7	7.1	3.7	35.1	23	54	67	50	57
Bosl	Trop	PdkF	2	35	5.8	3.4	31.6	50	60	76	32	76
Trin	SubT	PdkF	2	12	4.3	3.7	31.8	50	41	27	9	27
Trin	Trop	PdkF	2	135	5.1	3.3	34.2	44	50	54	21	52
BosT	Temp	GrnF	2	21	4.8	3.2	34.4	32	58	67	24	67
Relocate	ed 3-7 mor	nths ago										
Bosl	SubT	GrnF	2	25	6.8	3.4	34.1	61	60	68	52	68
Bosl	SubT	GrnF	4	17	5.4	3.4	34.6	28	37	8	8	8
Bosl	SubT	GrnF	3	7	6.3	3.3	35.3	33	44	33	33	33
Bosl	SubT	PdkF	3	40	5.7	3.1	31.8	40	43	35	12	35
Bosl	Trop	GrnF	4	20	5.5	3.5	34.1	47	44	29	7	21
Bosl	Trop	GrnF	2	16	5.5	3.3	31.9	53	40	44	6	38
Bosl	Trop	GrnF	3	11	5.6	3.3	32.7	48	39	18	0	27
Bosl	Trop	PdkF	3	29	5.1	3.4	34.9	57	41	35	5	34
Bosl	Trop	PdkF	2	16	6.4	3.6	32.8	58	52	63	19	63
Trin	SubT	PdkF	2	13	4.0	3.7	32.6	40	37	15	15	15
Trin	Trop	GrnF	2	12	6.3	3.5	34.4	55	58	75	33	75
BosT	SubT	GrnF	2	31	6.1	3.5	36.1	51	56	72	38	68
Relocate	ed 8-12 mc	onths ago										
Bosl	SubT	PdkF	3	11	5.3	3.2	31.8	24	30	0	0	0
Bosl	SubT	PdkF	4	6	5.7	3.7	35.5	40	40	33	0	33
BIxT	SubT	PdkF	3	12	2.8	3.9	28.4	29	33	14	0	10
BosT	SubT	GrnF	3	7	4.1	3.7	36.3	44	34	0	0	0
BosT	Temp	GrnF	3	8	4.1	3.6	35.2	34	54	63	25	63
Relocate	ed >1 year	ago										
Bosl	Trop	PdkF	3	20	5.4	3.4	32.4	50	41	33	11	33
BosT	Temp	GrnF	3	27	4.9	3.7	37.5	39	65	78	57	67

#### Table 6. Averages values for physical and reproductive traits of relocated bulls

A group of 27 well-grown, fat Brahman bulls aged 2-3 years and weighing approximately 600-800 kg was relocated to 5 properties (S2, S3, S4, S6 and S7) within 200 km of each other in the northern spear grass region. Only half passed a BBSE immediately after relocation (Table 8). Subsequent access to only 8 of these was possible and this was at the end of the dry season going into mating in January, 3 months later. At this time, only two of these eight passed a BBSE.

Time from relocation	Body condition	Testes tone	Scrotal circumference	Sperm motility	Mean normal sperm	>50% normal sperm	>70% normal sperm	BBSE pass
	(1-9)*	(1-5)**	(cm)	(%)	%	%	%	%
G1. Crossbred	<b>ls 2 years</b> (r	n=135)						
2 m	5.1	3.3	34.2	44	50	52	19	52
G1a. Subset 2	(n=38)							
-0.5 m		3.2	36.2 a		55#	61	37	58
2 m	5.1	3.2	35.1 b	41	49	52	20	50
G2. Brahmans 2-3 years (n=107)								
0.8 m	7.4	3.5	33.9 a	43	50 a	57 a	22 a	57 a
7 m	5.6	3.3	33.3 b	38	41 b	26 b	9 b	25 b

#### Table 7. BBSE parameters for bulls relocated to Queensland's Gulf country

\* 1 (Very poor) to 9 (Over-fat) scale; \*\* 1 (Very soft) to 5 (Very hard) scale

Means followed by a letter in common are not significantly different (P=0.05): # P=0.07 for the difference

#### Table 8. BBSE parameters for bulls relocated to the northern spear grass region

Time from relocation	Body condition	Testes tone	Scrotal circumference	Sperm motility	Normal sperm	>50% normal sperm	>70% normal sperm	BBSE pass
	(1-9)*	(1-5)**	(cm)	(%)	%	%	%	%
S1. Brahma	ns 2 years (	n=25)						
3 m	5.5 x#	3.3	31.0 x	42	56	72	24	72
S1. Angus 2	<b>? years</b> (n=2	1)						
3 m	4.8 ay	3.2 a	34.4 ay	32	58	67	24	67
S1. Angus 3	<b>3 years</b> (n=9	)						
15 m	4.1 b	3.6 b	35.2 b	34	54	63	25	63
S1. Angus 4	<b>i years</b> (n=2	27)						
27 m	4.9 a	3.7 b	37.5 c	39	65	78	57	67
S4. Red Ang	gus 2 years	(n=7)						
4 m	7.0	3.3	39.6	31	50	67	33	57
7 m	7.1	3.4	39.6	45	43	57	14	57
11 m	4.1	3.7	36.3	44	34	0	0	0
S2, S3, S4, S	S6, S7. Bral	nmans 2-3 yea	<b>ars - Fat</b> (n=27)					
0.8 m	7.6	3.9	35.7	49	51	54	29	54
S2,S4. Subs	set of Brahn	nans 2-3 years	<b>s <i>- Fat</i></b> (n=8)					
0.8 m	7.3	4.0	34.9	53	47	50	25	50
3 m	6.8	3.8	34.3	60	47	25	25	25
S5. Brahma	ns 2-3 year:	s - Average co	ondition (n=49)					
5-7 m	5.2	3.3	33.5	57	41	40	6	40

\* 1 (Very poor) to 9 (Over-fat) scale; \*\*1 (Very soft) to 5 (Very hard) scale

# Means for S1 bulls followed by a letter in common within the range of a-c or x-y are not significantly different (P=0.05)

A group of 25 eighteen-month-old, paddock-reared Brahman bulls was relocated to property S1 (Table 8) in the northern spear grass region and evaluated 12 weeks later when their average weight was 437 kg. Three-quarters of these bulls passed a BBSE.

A mix of 50 paddock-reared and grain-fed Brahman bulls aged 2-3 years was relocated to property S5 on northern spear grass and evaluated 6 months later in the early wet season (Table 8). These bulls had experienced a severe dry season and were recovering body condition. Less than half passed a BBSE.

A group of 18-month-old Brahman bulls was relocated to property D2 via a property immediately south in mid-2000. These bulls had been sourced from both sub-tropical and tropical areas. They were evaluated 9 days and 6 months (only 2/3rds mustered) after relocation (Table 9). Immediately after relocation, bulls from the local region, and which had been relocated 2 months earlier than ex sub-tropics bulls, had larger scrotal circumferences (P=0.006), and half passed a BBSE, twice as many as ex sub-tropics bulls. Six months later, two-thirds of these bulls passed a BBSE, and in that period, scrotal circumference had increased by 4 cm (P<0.001).

Genotype#	Body condition	Testes tone	Scrotal circumference	Sperm motility	Mean normal sperm	>50% normal sperm	>70% normal sperm	BBSE pass
	(1-9)*	(1-5)**	(cm)	(%)	%	%	%	%
0.3 m after r	elocation							
Bs (n=30)	6.8 a##	3.3	30.6 a	12 a	36	28	7	28
CBs (n=17	6.3 b	3.3	31.2 ab	28 bc	42	33	0	33
CBt (n=3)	6.7 b	3.4	34.3 b	15 ab	50	47	35	47
C (n=18)	6.6 b	3.3	32.6 b	13 ab	44	40	13	33
RA (n=6)	5.3 a	3.0	32.3 ab	34 c	57	67	33	67
Brahman 0.3	3 (ex sub-tr	opics) and 2.3	(ex tropics) m	after relo	cation			
Bs (n=30)	6.8	3.3	30.6 a	12 a	36 a	28	7 a	28
Bt (n=10)	7.1	3.6	34.4 b	25 b	52 b	56	44 b	50
Brahman 0.3	3/2.3 and 6.	0 m after reloc	<i>ation</i> (n=27)					
B (0.3/2.3)	6.8	3.3	30.7 a	9 a	26 a	4	0 a	4
B (6.0)	6.8	3.3	34.4 b	56 b	58 b	71	48 b	66
Bos taurus	0.3 and 6.0	m after reloca	<i>tion</i> (n=22)					
C&RA (0.3)	6.3 a	3.2 a	32.5	18 a	50	53	21	45
C&RA (6.0)	5.1 b	3.7 b	33.1	58 b	61	72	43	68
Charbray 0.	3 and 6.0 m	after relocatio	<b>on</b> (n=12)					
CB (0.3)	6.7	3.4	33.1 a	17 a	43 a	25	17	25
CB (6.0)	6.2	3.3	34.2 b	59 b	57 b	83	25	83

Table 9. BBSE parameters for 2-year-old bulls relocated to northern downs (D1 & D2) in Queensland

\* 1 (Very poor) to 9 (Over-fat) scale; \*\* 1 (Very soft) to 5 (Very hard) scale

# B=Brahman, C=Charolais, CB=Charbray, RA=Red Angus, s & t suffix=ex Sub-tropics & ex Tropics

## Means for bulls followed by a letter in common are not significantly different (P=0.05)

## Relocation change in crossbred bulls from central Queensland to the Gulf and northern downs

A group of 135 paddock-reared Brahman x Red Angus bulls was relocated from central Queensland to property G1. Two months later, half of them passed a BBSE (Table 7). A subgroup of 38 of the bulls had undergone a BBSE prior to relocation. These bulls averaged 543 (430-630) kg prior to relocation (29 August 2000). Testes tone remained constant. Scrotal circumference reduced during the relocation period (P<0.001), as did average normal sperm (P=0.07); but the proportion of bulls passing a BBSE did not change ( $\chi^2$ =0.21; P=0.65; Table 7).

A group of 18-month-old Charbray bulls was relocated from central Queensland to a property south of D1 in mid-2000, and soon after relocated again to either property D1 or D2. These bulls were evaluated 9 days and 6 months after relocation (Table 9). Source did not significantly influence BBSE parameters. Over 80% of the bulls (10 of 12 tested) passed a BBSE after 6 months, with less than half achieving this immediately after relocation. Some of this increase could have been due to growth and maturity changes, and some due to recovery from sub-fertility associated with relocation.

#### Bos taurus bulls post-relocation in north Queensland

A group of 7 Red Angus bulls in prime condition and aged about 20 months was relocated to property S4. The bulls were segregated until mating between January and May 2001. They were assessed 4 months after arrival, and then at the start and end of mating. The bulls did not receive preferential management. All bulls lost substantial body condition over mating during the wet season, and all became sub-fertile with average normal sperm of 34% (Table 8) eleven months after relocation.

A group of 18-month-old Charolais and yearling Red Angus bulls was relocated from central Queensland to property D1 in mid-2000. These bulls were evaluated 9 days and 6 months after relocation (Table 9). 68% of the bulls passed a BBSE after 6 months whilst only 45% achieved this immediately after relocation. Some of this increase could have been due to growth and maturity changes, and some possibly due to recovery from fertility depression following relocation.

Angus bulls aged 2 to 4 years were relocated from temperate areas to property S1 when 18 months of age. The youngest of these had been relocated 3 months prior to BBSE. The average weights of the bulls aged 2 and 3 years were 467 and 459 kg, respectively; the bulls aged 4 years appeared heavier (scales were unavailable). Despite these bulls being in condition score 4-5, two-thirds of them passed a BBSE (Table 8). Body condition of the Brahmans was significantly better than that in the youngest Angus (P<0.001).

#### Sanga (Bos taurus) bulls relocated to a harsh environment

Twelve 2-year-old Belmont Red bulls, all having at least 50% normal sperm and an average of 66% normal, were relocated in September 2000 about 850 km from central Queensland to property S8 in north Queensland. Nutritional conditions were poorer than average in the year following relocation. The bulls dropped from condition score 5.8 at relocation in September 2000 to score 4.3 by November 2000 after relocation (Figure 3). Their average condition score was 3.9 by the start of mating in January 2001. After a wet season with very low rainfall, the bulls experienced a harsh dry season, and their average condition score in September 2001 was 2.8, when 2 were missing, presumed dead.

Scrotal circumference continually declined after relocation (Figure 3). The scrotal circumference of 4 of the bulls dropped to about 25 cm by September 2001 with one having almost no testicular tone. Overall, scrotal circumference reduced 5 cm after relocation, 4 cm of which was between January and September 2001. The severe loss of body condition, scrotal circumference and testicular tone was reflected in reductions in the normal sperm produced by the bulls. Two, 4 and 12 months after relocation, only 27%, 17% and 10% of bulls respectively had at least 50% normal sperm (Figure 4). No semen was taken from poor-condition bulls in September 2001. Only one bull had normal sperm of at least 50% at that time; it was the only bull with a scrotal circumference greater than 32 cm.

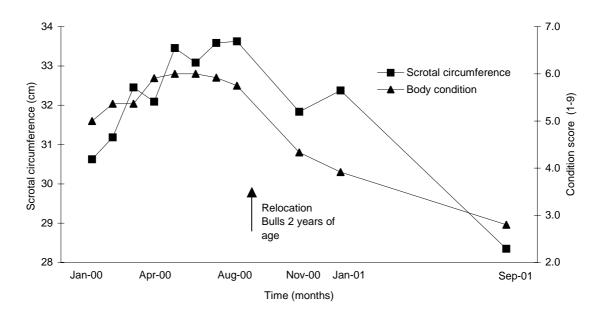
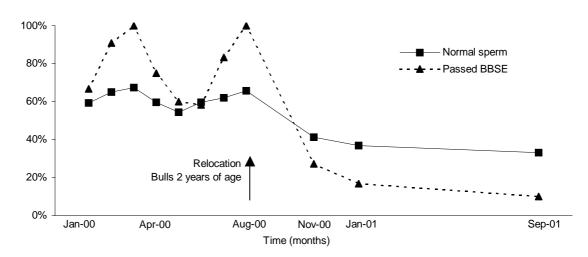


Figure 3. Change in body condition and scrotal circumference of Belmont Red bulls preand post-relocation

Figure 4. Change in normal sperm and percentage of Belmont Red bulls passing a breeding soundness examination pre- and post-relocation



#### Nutritional effects on bull breeding soundness subsequent to relocation

At both S5 and S9, all but 2 bulls passed a BBSE in the months after purchase. In the 2001 dry season, bulls, with the exception of one at S5, lost an average of 1.6 (S5) and 2.8 (S9) units of body condition (P<0.001); i.e., they went from condition score 7 to between score 4 and 5. Scrotal circumference was reduced by 2.5 cm (P<0.001) over this dry season. Bulls produced an average of 30% fewer normal sperm (P<0.01) reflecting body condition and scrotal circumference loss (Figures 5 and 6). Only one bull across both properties had at least 50% normal sperm in the mid-dry season. These effects were in

reverse in the early wet season of 2001/2002 when 1/3 (S5) to 3/4 (S9) of the scrotal circumference loss (P<0.001) and 61% (S5) and 91% (S9) of the average loss in normal sperm had been recovered (P<0.01). The ranking of bulls on scrotal circumference was consistent with repeatabilities of 0.8 (S5) and 0.7 (S9) (se: 0.1). Spearman rank correlations for normal sperm of 0.5 (S5) and 0.6 (S9; P<0.05) between August 2001 and January 2002 indicated moderately consistent ranking. However repeatabilities over all testing times were not high at  $0.49\pm0.18$  (S5) and  $0.36\pm0.16$  (S9). There was no apparent effect of time on the property or genotype on any trait in these bulls.

#### Figure 5. Seasonal effects on scrotal circumference of relocated bulls

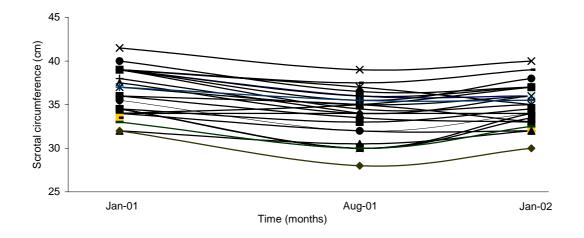
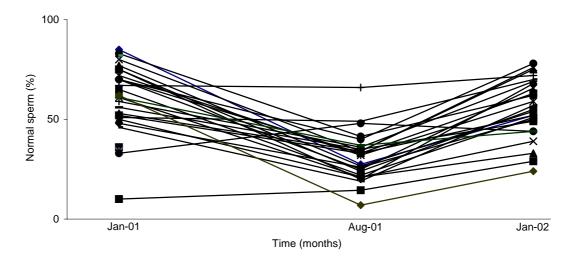


Figure 6. Seasonal effects on normal sperm of relocated bulls



#### 6.1.3.5 Discussion and conclusions

The observations conducted clearly show that a very high proportion of bulls (~50%) fail a BBSE in the period immediately after sale and relocation. This occurs independently of genotype, age, and region of source. About 80% of these bulls recover breeding soundness which is similar to previous observations for acclimatised bulls (LA Fitzpatrick, pers comm).

The recovery of breeding soundness can be within six months under favourable nutritional and management conditions. This appeared more likely on northern downs country than on either spear grass or Gulf country. However, our observations have shown that many bulls experiencing prolonged poor nutritional conditions after relocation may even suffer further depression of fertility. Our observations occurred during a period of below-average seasonal conditions and this created extended nutritional stress for many bulls.

The effect of poor nutrition depressing breeding soundness was often, but not always, greatest in *Bos taurus* bulls. Such bulls on good nutrition may recover levels of fertility equivalent to that for adapted cattle as shown in the observations at D1 and S1. The S9 observation clearly showed the disastrous consequences of introducing poorly-adapted cattle into a harsh tropical environment.

We have presented data for a range of genotypes showing that nutritional stress and significant loss of body condition by bulls is accompanied by reduced scrotal circumference and a high proportion of bulls producing semen with <50% normal sperm. This effect is reversed with a return of good nutrition. The major implication is that a breeding soundness evaluation of bulls in backward condition, and/or under nutritional stress will not be directly indicative of breeding soundness when in forward body condition. Also, it demonstrates that bulls should be managed to have them in good condition leading into mating.

If bulls in backward body condition are evaluated, the evidence from our observations at properties S5 and S9 is that ranking of bulls on scrotal circumference and sperm morphology will be similar to that when body condition is recovered, thus giving some indication of future relative breeding soundness. This is of importance when using these traits as parameters in selection of bulls for mating when seasonal conditions are unfavourable.

## 6.1.4 The effect of feeding and relocation on reproductive traits of Brahman bulls

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#### 6.1.4.1 Summary

At Brigalow Research Station in central Queensland, 80 yearling Brahman bulls (~ 18 m of age and mean liveweight of 355 kg) were allocated to 2 treatments, G - grazing pasture only or F – pasture and supplemented with a commercial grain and concentrate pellet mix through self-feeders at a rate of 1-1.5% body weight (about 6 kg/bull/day) for 150 days. At the end of feeding, within each feeding treatment, bulls were allocated to 4 relocation treatments: - NRG (not relocated, grazing pasture only prior to relocation), NRF (not relocated, grazing pasture and supplemented prior to relocation), RG (relocated, grazing pasture only prior to relocation) and RF (relocated, grazing pasture and supplemented with pellets prior to relocation). Relocated bulls were then transported by truck 1100 km (16 h) to Toorak Research Station in north-west Queensland. Bulls were subjected to a physical and reproductive examination and semen collected on a 6-8 weekly basis from the commencement of the experiment (16 May 2001) until 12 months after relocation (04 November 2002).

During the feeding period, G bulls lost 4 kg (-0.03 kg/day) whilst the F bulls gained 143 kg (1.03 kg/day). Final liveweights were 362 kg and 517 kg for G and F bulls respectively. Mean scrotal circumference at the end of the feeding period was 31.8 cm for G bulls and 36.1 cm for F bulls. There was no effect of feeding on mass activity or motility of semen or on either normal sperm or on any of the sperm abnormality categories although both semen and sperm traits improved over time. All semen and sperm traits were either moderately to highly repeatable over the feeding period.

Gains during the post-relocation period and final liveweights were respectively NRG 149 and 526 kg, NRF 52 and 565 kg, RG 225 and 593 kg and RF 128 and 633 kg. For scrotal circumference, changes during the post-relocation period and final sizes were respectively NRG 3.4 and 36.6 cm, NRF 1.2 and 37.1 cm, RG 4.4 and 37.3 cm and RF 2.2 and 37.7 cm. For semen traits, there was a significant treatment x time (P<0.001) interaction for mass activity with R bulls generally higher than in NR bulls. There was only a significant (P<0.001) effect of time on motility with motility decreasing until August 2002 and then increasing back to its original value at time of relocation. Overall relocation had minimal effect on sperm morphology. For normal sperm, there was a significant (P=0.014) relocation x time interaction with NR bulls being significantly higher than R bulls (60.6% v 50.2%) in September 2002 but not at other times. There was no effect of either feeding or relocation treatment on any of the sperm abnormality categories. Normal sperm was highly repeatable with semen traits and sperm abnormalities being at least moderately repeatable.

Biochemical and haematological parameters indicated that the relocation process was somewhat stressful with significant elevations in L-lactate, non-esterified fatty acids (NEFA) and packed cell volume (PCV) on arrival at Toorak but there were no elevations in cortisol, creatine phosphokinase (CPK) and aspartate aminotransferase (AST). Bulls were relocated under conditions better than those experienced by many industry bulls. Thus, our conclusion from this study is that providing bulls are transported under favourable conditions, there should be minimal effect of feeding or relocation on semen traits or sperm morphology.

## 6.1.4.2 Aim

The aim of this experiment was to measure the effect of feeding and relocation on reproductive traits of bulls relocated from central Queensland to north-west Queensland.

## 6.1.4.3 Materials and methods

#### **Experimental design**

The experiment was conducted in 2 phases, the feeding phase prior to relocation and then the relocation phase.

- a) Feeding phase. An unreplicated design of 2 treatments with 40 bulls per treatment was used.
  - G grazing pasture only.
  - F grazing pasture and supplemented with a commercial grain and concentrate pellet mix.
- *b) Relocation phase.* The design was a 2 x 2 factorial of 4 treatments each with 19 Brahman bulls per treatment with 76 bulls in the experiment.
  - NRG not relocated, grazing pasture only prior to relocation,
  - NRF not relocated, grazing pasture and supplemented with pellets prior to relocation,
  - RG relocated, grazing pasture only prior to relocation, and
  - RF relocated, grazing pasture and supplemented with pellets prior to relocation.

#### Experimental sites and animals

The experiment was conducted at both Brigalow Research Station  $(24^{\circ}S, 149^{\circ}E)$  in the brigalow region of central Queensland and at Toorak Research Station  $(21^{\circ}S, 141^{\circ}E)$  in the Mitchell grass downs country of north-west Queensland. Brigalow Research Station has a sub-humid sub-tropical environment with mean minimum and maximum temperatures ranging from between  $21^{\circ}$  and  $33^{\circ}C$  for January to between  $4^{\circ}$  and  $22^{\circ}C$  for July. The average rainfall for the station is 730 mm with about 50% of it occurring in the November to February period. Toorak is an arid tropical environment with mean minimum and maximum temperatures ranging between  $24^{\circ}$  and  $41^{\circ}C$  for January to between  $4^{\circ}$  and  $27^{\circ}C$  for July. The average rainfall for the station is 480 mm with about 70% of it occurring in the November to February period.

The bulls were high grade Brahman born at Fletcherview Research Station, Charters Towers, north Queensland. They were born from November 1999 to February 2000 and weaned in June 2000. On 03 August 2000, 80 weaner prepuberal bulls were trucked 920 km to Brigalow Research Station. On arrival the bulls grazed as a common group for 10 months on improved pasture of a mixture of buffel grass (*Cenchrus ciliaris*), bluegrass (*Dicanthium sericeum*), Rhodes grass (*Chloris gayana*), green panic grass (*Panicum maximum*) and seca stylo (*Stylosanthes scabra*). During this acclimatisation period, bulls were mustered 8 times at 3-8 weekly intervals for weighing, physical and reproductive examinations and semen was collected twice, on 4 April 2001 and 16 May 2001. Bulls were vaccinated against the tick fevers, the common clostridial diseases and bovine ephemeral fever. Stocking rate was about 1 adult equivalent (AE) to 3 ha.

#### Procedures

On 13 June 2001, bulls were randomly allocated to 2 treatments, G and F on liveweight on 16 May 2001 and balanced for morphologically normal sperm. Both groups grazed separate native pasture paddocks and the F treatment were fed, commencing 29 June 2001, a commercial grain and concentrate pellet mix (CP 15%, ME 10.4 MJ/kg) through self-feeders at a rate of 1-1.5% body weight (about 6 kg/bull/day) for 150 days until 26 November 2001. During the feeding period, bulls were mustered 5 times at 4-6 weekly intervals for weighing, recording flight speed and conducting physical and reproductive examinations. Semen was collected at 3 of these examinations.

On 26 November 2001, within each feeding treatment, bulls were allocated to the 4 relocation treatments, NRG, NRF, RG and RF using a stratified randomisation of average of normal sperm from the 2 previous assessments with the proviso that the liveweights within each feeding treatment were comparable. The non-relocated bulls were then returned to the paddock and run as a common group on pasture. The relocated groups were kept as separate groups overnight in the yard on water, loaded next day at 0830 with 19 RG bulls on the top deck and 18 RF bulls on the bottom deck and transported 1100 km to Toorak Research Station in north-west Queensland, arriving at 0030 the next day. On arrival bulls were kept in the yards as one group with access to grassy hay and water. Within 10 hours of their arrival, bulls were bled from the coccygeal vein and then paddocked at a stocking rate of 1AE to about 10-15 ha. The topography is flat, with little shade and pasture mainly Mitchell and Flinders grasses.

After relocation, bulls were mustered 8 times at 5-7 weekly intervals, at both properties, for weighing, recording flight speeds, and conducting physical and reproductive examinations with the final examinations in November 2002. The examinations on each property were conducted within 2–8 days of each other, with all of the physical and reproductive assessments being done by the same operator throughout the experimental period. At both locations bulls were mustered on the morning of the examinations and yarded about 0700-0800, weighed and examinations completed by about 1600, then returned to their paddock that day.

Flight speed was recorded using the method of Burrow *et al.* (1988) with the time taken for an animal to travel 1.6 m on release from the weighing crate being measured. This value was converted to metres per second for analysis

A blood sample for haematology and biochemistry was taken from the coccygeal vein from each bull on 15 November 2001 (prior to relocation), within 10 hours of arrival at Toorak on 28 November 2001 then again at each location at the January and March examinations. Blood was drawn into 10mL EDTA evacuated tube (Vacutainers, Becton Dickinson) and stored in a cool-box until all samples had been collected. Total erythrocytes, leucocytes and haemoglobin concentration were determined using a Sysmex electronic counter. The sample was then centrifuged (1000g, 20 min) and five 1mL aliquots of plasma were aspirated and placed into 1mL cryotubes. Plasma samples were stored at –20°C awaiting biochemical analysis. For biochemical analysis, a 1mL plasma aliquot from each animal was thawed and centrifuged (1000g, 20 min) to remove fibrin precipitate. Using an Olympus Reply Chemistry Analyser, each sample aliquot was tested for the following: - aspartate aminotransferase (AST), creatine phosphokinase (CPK), L-lactate and non-esterified fatty acids (NEFA). Plasma cortisol levels were determined using the EIA Cortisol ELISA kit (DSL Laboratories).

After relocation, at each property a faecal sample was collected at each examination, at the time of insertion of the electroejaculation probe, from about 50% of bulls. This sample was bulked, sub-sampled then oven-dried at 60°C for determination of dietary crude protein (CP), in vitro digestibility of dry matter (IVDMD) and grass/non-grass content by near-infrared reflectance spectroscopy (NIRS) (Lyons and Stuth 1992; Coates 1998)

#### Statistical analyses

All statistical analyses were performed using GenStat for Windows 6<sup>th</sup> Edition

#### Feeding period

A random coefficients regression approach was used to analyse liveweight and scrotal circumference. The average growth pattern was approximately linear over the time span considered for the majority of bulls. However, the individual bulls have their own pathway of growth so that there is potentially variation in intercepts and slopes across bulls.

The full model fitted was

• Fixed effects: *feeding* + *time* + *feeding.time* 

- Random effects: *bull* + *bull.time* + *time\_factor* + *feeding.time\_factor*
- Random intercepts and slopes within bulls are correlated.

#### Where

- feeding has 2 levels, G (pasture) and F (supplement),
- *time* is a covariate, days since the start of feeding on 29 June 2001,
- bull is the variance component for random intercepts for bulls,
- *bull.time* is the variance component for random slopes for bulls,
- time\_factor is a random lack of fit term for additional fluctuations about the linear trend, and
- *feeding.time\_factor* is a random lack of fit term for additional fluctuations about the linear trend for each feeding treatment.

Random terms with zero variance components and non-significant fixed terms (P>0.05) were removed to arrive at the final model.

A linear mixed model was used to analyse all other variables measured during the feeding period. A log transformation was applied to flight speed data and an angular transformation applied to abnormal midpieces, abnormal tails and cytoplasmic droplets prior to analysis to stabilise the variance. The full model fitted was:

- Fixed effects: feeding+time+feeding.time+age+feeding.age
- Random effects: *bull+bull.time*

#### Where

- feeding has 2 levels, G (pasture) and F (supplement),
- time is a factor generally with 3 levels 15 August 2001, 4 October 2001 and 15 November 2001,
- age is a covariate, age in days at the start of feeding (29 June 2001),
- bull represents the variance component for individual bulls, and
- *bull.time* is the residual variance.

Random terms with zero variance components and non-significant fixed terms (P>0.05) were sequentially removed to arrive at the final model for each trait. Approximate least significant differences were calculated for significant (P<0.05) fixed effects.

Repeatability (intraclass correlation) was estimated as the ratio of variance components:

Repeatability = bull/(bull + bull.time).

#### **Relocation period**

A random coefficients regression approach was used to analyse liveweight and scrotal circumference during the relocation period. The average growth pattern was approximately linear for scrotal circumference and quadratic for liveweight over the time span considered for the majority of bulls.

The full model for liveweight was as follows.

- Fixed effects: feeding + treatment + time + time<sup>2</sup> + feeding.treatment + feeding.time + treatment.time + feeding.treatment.time<sup>2</sup> + treatment.time<sup>2</sup> + feeding.treatment.time<sup>2</sup>
- Random effects: *bull* + *bull.time* + *time\_factor* + *feeding.time\_factor* + *treatment.time\_factor* + *feeding.treatment.time\_factor*
- Random intercepts and slopes within bulls are correlated.

#### Where

- feeding has 2 levels, G (pasture) and F (supplement),
- treatment has 2 levels, relocated (R) and non-relocated (NR),
- time is days since the start of relocation on 27 November 2001,
- *time*<sup>2</sup> is the square of time representing the quadratic response,
- *bull* is the variance component for random intercepts for bulls,
- bull.time is the variance component for random slopes for bulls,
- time\_factor is a random lack of fit term for additional fluctuations about the linear trend, and
- feeding.time\_factor, treatment.time\_factor and feeding.treatment.time\_factor are random lack of fit term for additional fluctuations about the quadratic trend

The full model for scrotal circumference was as follows.

- Fixed effects: feeding + treatment + time + feeding.treatment + feeding.time + treatment.time + feeding.treatment.time
- Random effects: *bull* + *bull.time* + *time\_factor* + *feeding.time\_factor* + *treatment.time\_factor* + *feeding.treatment.time\_factor*
- Random intercepts and slopes within bulls are correlated.

Random terms with zero variance components and non-significant fixed terms (P>0.05) were removed to arrive at the final model.

The following linear mixed model was used to analyse condition score, testicular tone, flight speed, motility, mass activity and haematological and biochemical traits measured during the relocation period. A log transformation was applied to cortisol and CPK data prior to analysis to stabilise the variance. The full model fitted was:

- Fixed model: feeding + treatment + time + feeding.treatment + feeding.time + treatment.time + feeding.treatment.age + feeding.treatment.age + feeding.treatment.age
- Random model: *bull* + *bull.time*

#### Where

• feeding has 2 levels, G (pasture) and F (supplement),

- treatment has 2 levels, relocated (R) and non-relocated (NR),
- time is a factor generally with 9 levels, 15 November 2001, 14 January 2002, 26 February 2002, 4 April 2002, 13 May 2002, 4 July 2002, 5 August 2002, 17 September 2002 and 4 November 2002,
- age is a covariate, bull age (days) at the time of relocation (27 November 2001),
- *bull* represents the variance component for individual bulls, and
- *bull.time* is the residual variance,

Sperm morphology variables were analysed 2 ways:

- Including all samples with 100 sperm counted in the spermiogram (all-bulls), and
- Including samples from bulls with 50%+ normal sperm in November 2001 and with 100 sperm counted (50%+ bulls).

An angular transformation was applied to abnormal midpieces, abnormal tails and cytoplasmic droplets data prior to analysis to stabilise the variance. Analysis of sperm morphology traits was similar to that for condition score above, but with the addition of an extra random term *block*. The full random model was:

• Random model: *block* + *block.bull* + *bull.time* 

Where

• *block* represents the blocks formed by stratified allocation of bulls to relocation treatments based on normal sperm.

In all linear mixed models, random terms with zero variance components and non-significant fixed terms (P>0.05) were sequentially removed to arrive at the final model for each trait. Approximate least significant differences were calculated for significant (P<0.05) fixed effects.

Repeatability of traits during the relocation period was calculated using the same method as outlined for traits during the feeding period.

For haematological and biochemical traits a second analysis was conducted including relocated bulls only to incorporate the measurement made on relocated bulls only, immediately after relocation. The full model was:

- Fixed effects: *feeding* + *time* + *feeding.time*
- Random effects: *bull* + *bull.time*

Where

- feeding has 2 levels, G (pasture) and F (supplement),
- *time* is a factor with 4 levels, 15 November 2001, 28 November 2001, 14 January 2002 and 4 April 2002,
- *bull* represents the variance component for individual bulls, and
- *bull.time* represents the residual variance.

#### 6.1.4.4 Results Pre-feeding period

At allocation there were no significant differences between the groups for any of the physical or reproductive traits (Table 10).

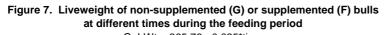
#### 6.1.4.4 Results Feeding period

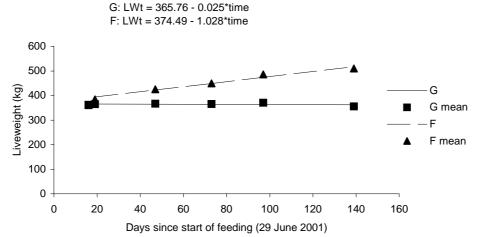
#### Liveweight

The average growth pattern was approximately linear over time for both treatments (Figure 7). For the 139-day period (29 June to 15 November 2001), G bulls lost 4 kg (0.03 kg/day) whilst the F bulls gained 143 kg (1.03 kg/day). Final liveweights were 362 kg and 517 kg for G and F bulls respectively. Three bulls developed lameness from acidosis within a month of the start of feeding. One bull was then removed from the experiment as lameness persisted.

supplemented (F) at anocation			
Trait	G	F	sd
Liveweight (kg)	355.4	355.1	32.8
Condition score (1-9)	5.4	5.3	0.5
Flight speed (m/sec)	0.68	0.55	0.3
Scrotal circumference (cm)	30	31	3.5
Testicular tone (1-5)	3.9	4	0.3
Mass activity (1-5)	1.6	1.6	1.3
Motility (%)	57.9	58.6	21.5
Normal sperm (%)	44.7	38.7	21.7
Abnormal heads (%)	32.7	32.5	16.1
Abnormal midpieces (%)	10.9	12	10.3
Abnormal tails (%)	2.6	5	8.9
Cytoplasmic droplets (%)	9.2	11.8	13.9

Table 10. Means and standard deviations (sd) for non-supplemented (G) and supplemented (F) at allocation



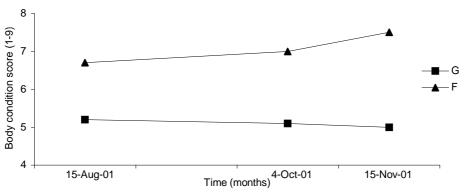


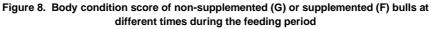
#### Flight speed

Feeding had a significant (P=0.003) effect on flight speed with F bulls having consistently higher flight speeds than G bulls (1.90 v 1.45 m/sec). Flight speed did not change over time (P=0.229). Repeatability of flight speed was estimated as 0.76 ± 0.04

#### **Condition score**

There was a significant feeding x time interaction (P>0.001) on condition score. Values for F bulls significantly increased over time and were significantly higher than that of G bulls at each time (Figure 8).



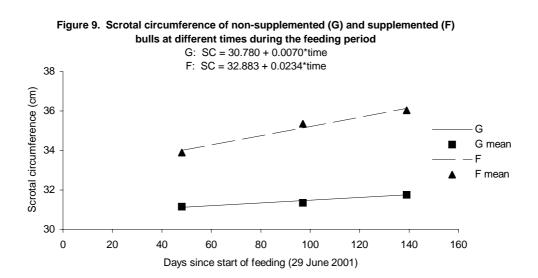


#### Testicular tone

There was almost no variation in testicular tone over time with the majority of bulls having values of 4.

#### Scrotal circumference

Figure 9 illustrates the increase in scrotal circumference with the average growth pattern for both treatments being approximately linear over time. There was a significant (P<0.001) difference for both the intercept and slope with G being greater than F. Mean scrotal circumference of G and F bulls at the end of the feeding period was 31.8 and 36.1 cm respectively. Repeatability of scrotal circumference was estimated as  $0.96 \pm 0.01$ .



### Mass activity and motility

There were no significant treatment effects with both traits significantly (P<0.001) increasing over time (Table 11).

### Sperm morphology

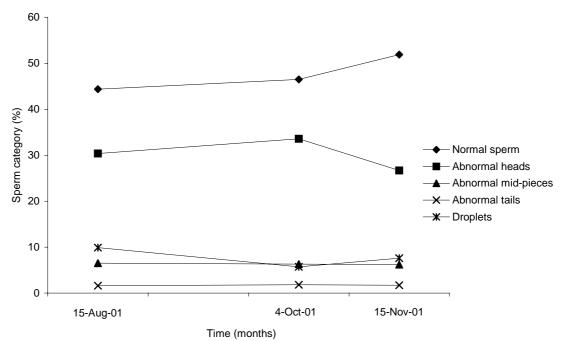
Normal sperm significantly (P<0.001) increased over time with no significant differences between G and F bulls (Table 11; Figure 10). There were no significant differences between G and F bulls for the various categories of abnormalities. Abnormal heads significantly (P<0.001) decreased with time whilst abnormal midpieces and abnormal tails remained relatively constant. Cytoplasmic droplets were highest in August 2001 and lowest in October 2001.

15-Aug-01	4-Oct-01	15-Nov-01	lsd
2.0a	2.7b	2.8b	0.3
64.9a	70.9b	72.0b	3.5
44.4a	46.5a	51.9b	3.6
30.4b	33.6c	26.7a	3.1
14.8(6.5)a#	14.6(6.3)a	14.5(6.2)a	
7.4(1.6)a	7.7(1.8)a	7.6(1.7)a	
18.3(9.9)b#	13.8(5.7)a	16.1(7.6)ab	2.5
	2.0a 64.9a 44.4a 30.4b 14.8(6.5)a# 7.4(1.6)a	2.0a         2.7b           64.9a         70.9b           44.4a         46.5a           30.4b         33.6c           14.8(6.5)a#         14.6(6.3)a           7.4(1.6)a         7.7(1.8)a	2.0a         2.7b         2.8b           64.9a         70.9b         72.0b           44.4a         46.5a         51.9b           30.4b         33.6c         26.7a           14.8(6.5)a#         14.6(6.3)a         14.5(6.2)a           7.4(1.6)a         7.7(1.8)a         7.6(1.7)a

#### Table 11. Effects of feeding on semen and sperm characteristics

Means within rows not followed by a common letter differ significantly (P<0.05)

#Both angular transformed and back transformed means presented



### Figure 10. Changes in sperm abnormality categories during feeding period

### Repeatability of semen and sperm parameters

Results are presented in Table 12. All of the traits were moderately to highly repeatable with the value for normal sperm being  $0.75 \pm 0.04$ .

,	· , · ·	
Trait	Repeatability (± se)	Comment
Mass activity	$0.41 \pm 0.07$	Moderate
Motility	$0.58 \pm 0.06$	Moderate
Normal sperm	$0.75 \pm 0.04$	High
Abnormal heads	$0.51 \pm 0.07$	Moderate
Abnormal mid-pieces	$0.67 \pm 0.05$	High
Abnormal tails	$0.67 \pm 0.05$	High
Cytoplasmic droplets	$0.47 \pm 0.07$	Moderate

Table 12. Repeatability (± se) for semen and sperm traits

# 6.1.4.5 Relocation period

### Liveweight

The response during the relocation period was approximately quadratic (Figure 11). There were significant differences in the slope for both relocation treatment and previous feeding (P<0.001). As well there was a difference in the curvature for previous feeding (P<0.001) but the curvature was similar for Gains during the 12-month relocation period and final liveweights were relocation treatments. respectively NRG 149 and 526 kg, NRF 52 and 565 kg, RG 225 and 593 kg and RF 128 and 633 kg.

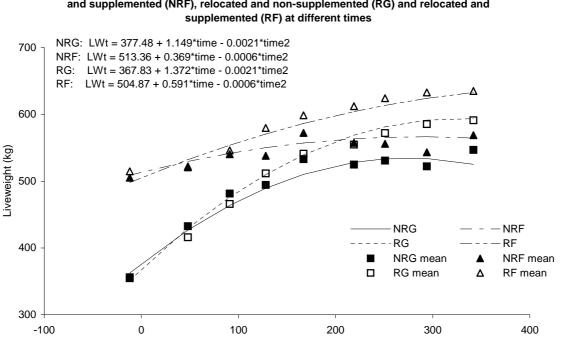
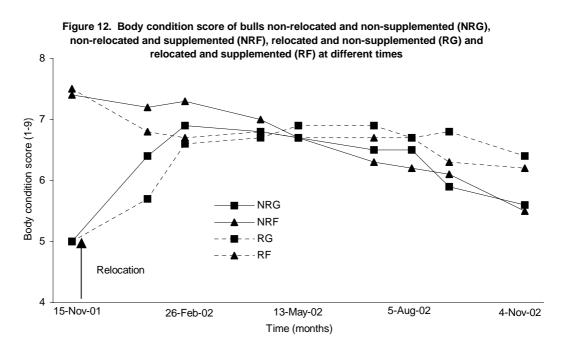


Figure 11. Liveweights of bulls non-relocated and non-supplemented (NRG), non-relocated and supplemented (NRF), relocated and non-supplemented (RG) and relocated and

Days since relocation (27 November 2001)

# Body condition score (Figure 12)

There was no consistent pattern over time with a significant (P=0.006) feeding x relocation x time interaction. F bulls were in significantly (P<0.05) better body condition than G bulls at time of relocation. The condition of F bulls then gradually decreased over time with the decrease being greater in NR bulls. This contrasted with the G bulls where condition of the NR increased sharply then slowly decreased while the R bulls increased in condition then generally maintained this condition.

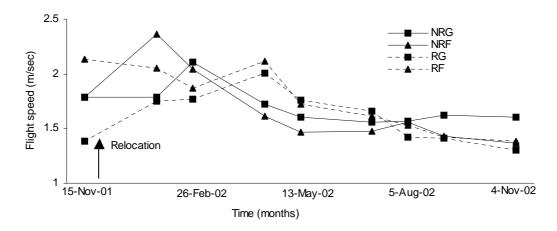


# Flight speed (Figure 13)

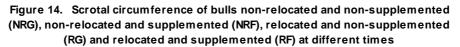
There was a significant (P=0.048) feeding x treatment x time interaction of flight speed. Overall, many bulls had reasonably consistent flight speeds over time but some bulls had marked fluctuations. Apart from RF bulls which decreased, flight speeds increased in groups until 26 February 2002 then tended to decrease over time with less change in NR bulls than in R bulls.

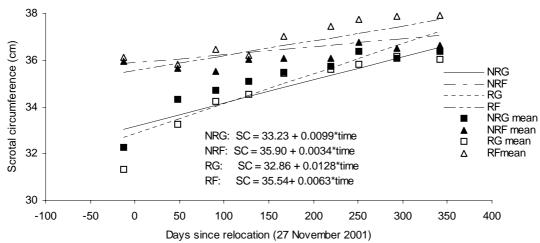
### Scrotal circumference

The response in all 4 treatments was approximately linear during the relocation phase although there was a difference between relocation treatments (P=0.009) and previous feeding (P=0.002) for the slope (Figure 14). The initial differences between the G and F groups had narrowed by the end of the experiment with gains during the 12-month relocation period and final scrotal circumferences being respectively NRG 3.4 and 36.6 cm, NRF 1.2 and 37.1 cm, RG 4.4 and 37.3 cm and RF 2.2 and 37.7 cm.









### **Testicular tone**

There was a significant (P=0.003) treatment x time interaction but overall there was no consistent pattern. Mean values were 4.0 at time of relocation and 3.6 at the end of the relocation period, 12 months later.

### Mass activity

There was no significant effect of feeding with means of  $2.6 \pm 0.1$  and  $2.7 \pm 0.1$  for G and F bulls. There was a significant (P<0.001) relocation x time interaction. Mass activity was generally higher in R than NR bulls with these differences being significant at 14 January, 4 July, 17 September and 4 November 2002 (Table 13; Figure 15).

### Motility

There was considerable variation between and amongst bulls in motility. There was only a significant (P<0.001) effect of time on motility with motility decreasing until August 2002 and then increasing back to its original value at time of relocation (Table 13).

Trait	Treatment	15-Nov-01	14-Jan-02	26-Feb-02	4-Apr-02	13-May-02	4-Jul-02	5-Aug-02	17-Sep-02	4-Nov-02	lsd
Mass activity (0-5)	NR	2.7a	3.1b	2.8a	2.5a	2.6a	2.8b	2.3a	3.1b	3.2b	
	R	2.8a	2.5a	2.7a	2.6a	2.7a	2.0a	2.1a	2.6a	2.6a	
	Isd within times	4.4									
Motility (%)	All bulls	72.0c	68.6c	67.8c	63.1b	60.0ab	56.7a	56.3a	68.5c	69.1c	4.18
Normal sperm (%)	NR	51.4a	51.4a	53.9a	56.5a	56.8a	58.6a	54.8a	60.6b	54.9a	
	R	52.4a	51.6a	51.9a	55.4a	52.2a	54.2a	52.7a	50.2a	55.2a	
	Isd within times	6.57									
Abnormal heads (%)	NR	27.7a	25.4a	27.2a	25.9a	25.6a	24.4a	29.1a	23.7a	26.5a	
	R	25.3a	29.0a	29.0a	24.4a	23.6a	24.8a	26.4a	28.7a	27.2a	
	Isd within times	5.29									
Abnormal midpieces (%)	All bulls	14.9bc(6.6) <sup>#</sup>	12.9a(5.0)	13.7ab(5.6)	14.2abc(6.0)	15.8bcd(7.4)	16.8d(8.4)	14.3abc(6.1)	14.0abc(5.9)	14.0ab(5.8)	1.79
Abnormal tails (%)	NR	5.5a(0.9) <sup>#</sup>	4.7a(0.7)	6.3a(1.2)	7.2a(1.6)	3.7a(0.4)	5.8a(1.0)	5.1a(0.8)	5.0a(0.7)	5.8a(1.0)	
	R	9.9b(2.9)	5.5a(0.9)	7.9a(1.9)	8.7a(2.3)	8.7b(2.3)	10.3b(3.2)	7.0a(1.5)	4.4a(0.6)	6.8a(1.4)	
	Isd within times	3.73									
Cytoplasmic droplets (%)	NR	17.3a(8.8) <sup>#</sup>	19.7a(11.3)	15.3a(6.9)	13.5a(5.4)	12.9a(4.9)	10.2a(3.1)	12.6a(4.8)	12.6a(4.8)	13.0a(5.0)	
	R	14.5a(6.2)	16.7a(8.2)	14.7a(6.4)	13.2a(5.2)	15.6a(7.2)	9.7a(2.8)	13.8a(5.7)	15.2a(6.8)	11.8a(4.2)	
	Isd within times	3.61									
	G	14.5a(6.2) <sup>#</sup>	18.5a(10.0)	13.6a(5.5)	14.4a(6.1)	12.8a(4.9)	9.3a(2.6)	14.4a(6.2)	13.7a(5.6)	12.9a(5.0)	
	F	17.3a(8.8)	17.9a(9.4)	16.4a(8.0)	12.3a(4.6)	15.7a(7.3)	10.5a(3.3)	12.1a(4.4)	14.1a(5.9)	11.9a(4.3)	
	lsd within times	3.61									

Table 13. Effects of relocation and feeding on semen and sperm traits of all bulls

For traits, means within rows or columns not followed by a common letter differ significantly; # angular transformed and back-transformed means presented

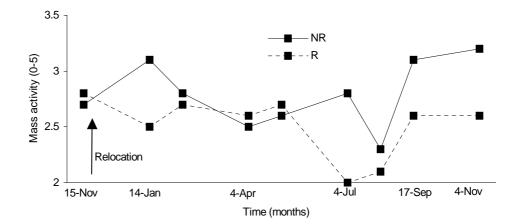
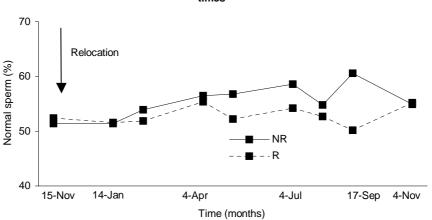


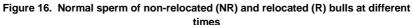
Figure 15. Semen mass activity of non-relocated (NR) and relocated (R) bulls at different times

### Sperm morphology

### Normal sperm

With all bulls (n=75), means for NR and R bulls were  $55.4 \pm 3.9$  and  $52.9 \pm 3.9$  and G and F bulls were  $56.6 \pm 3.9$  and  $51.7 \pm 3.9$  respectively. There was a significant (P=0.014) relocation x time interaction on normal sperm with NR bulls being significantly higher than R bulls (60.6% *v* 50.2%) in September 2002 but not at other times (Table 13; Figure 16).





When the analysis included only bulls that had 50+% normal sperm at time of relocation, there was a significant effect of feeding (P=0.012) and time (P=0.044) but not relocation treatment (P=0.607) on normal sperm. Means for NR and R bulls were  $62.3 \pm 1.8\%$  and  $63.4 \pm 1.8\%$  and G and F bulls were  $65.6 \pm 1.8\%$  and  $60.1 \pm 1.8\%$  respectively.

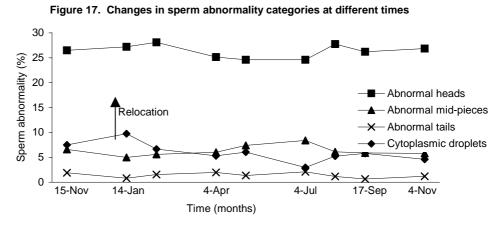
### Sperm abnormalities

The levels of significance of main effects and interactions of different categories of abnormalities for either all-bulls or 50+% normal bulls are presented in Table 14. Overall there were no significant main effects of relocation treatment or feeding on the various classes of abnormalities. There were significant main effects of time on all classes of abnormalities except abnormal heads of all bulls and abnormal midpieces of 50+% normal bulls.

Abnormality	Class	Feed	Relocation	Time	Feed x time Relocation x time	Age
Abnormal heads	All bulls	ns (P=0.752	2) ns (P=0.866)	* (P=0.038)	* (P=0.025)	
	50+%	ns (P=0.080	)) ns (P=0.486)	ns (P=0.306)		
Abnormal midpieces	All bulls	ns (P=0.068	3) ns (P=0.473)	*** (P=0.001)		
	50+%	ns (P=0.294	l) ns (P=0.482)	ns (P=0.365)		
Abnormal tails	All bulls	ns (P=0.063	3) ns (P=0.145)	*** (P<0.001)	** (P=0.003)	
	50+%	ns (P=0.180	)) ns (P=0.224)	**(P=0.004)		
Cytoplasmic droplets	All bulls	ns (P=0.694	l) ns (P=0.856)	*** (P<0.001)	* (P=0.036) * (P=0.044)	
	50+%	ns (P=0.740	)) ns (P=0.480)	*** (P<0.001)	** (P=0.002)	*(P=0.011)

Table 14. Main effects and interactions for categories of sperm abnormalities

For simplicity, changes for the various categories of sperm abnormalities are presented for the main effects of the all-bull category (Figure 17). The mean levels of abnormal heads were reasonably consistent ranging from 28.1% on 26 February 2002 to 24.6% on 4 April and 13 May 2002. There was no significant difference in abnormal heads between relocation treatments but the significant (P=0.025) relocation x time interaction was caused by different patterns over time, eg there was a significant increase in abnormal heads in R bulls but not NR bulls after relocation. Mean abnormal midpieces were in the range of less than 5.0 to 8.4% with no significant relocation effects (Table 13). Although there was a significant (P=0.003) relocation x time interaction with abnormal tails (Table 13), mean values were low overall ranging from less than 1 to 2.9. There were significant (P=0.044) relocation x time and feed x time (P=0.036) interactions with cytoplasmic droplets (Table 13). Overall means for cytoplasmic droplets were 5.9 and 5.8% for NR and R and 6.0 and 5.7% for G and F bulls respectively. There was also a significant (P=0.011) age effect of droplets in 50+% bulls with the coefficient for the covariate, age at time of relocation, being -0.0674  $\pm$  0.0264 indicating that droplets were more prevalent in younger bulls.



Repeatability of traits

Scrotal circumference was highly repeatable yet testicular tone was only lowly repeatable (Table15). Flight speed was moderately repeatable. Repeatability of the semen traits, mass activity and motility, was moderate. With all bulls, repeatability of sperm morphology traits was high for normal sperm and at least moderate for the various abnormality categories. Repeatability of morphology traits declined within the subsets of bulls having 50+% normal sperm with normal sperm, abnormal heads and abnormal midpieces being moderate and the other 2 categories being lowly repeatable.

Attribute	All bulls	Comments	Bull with 50+% normal sperm	Comments
Flight speed	$0.66 \pm 0.04$	Moderate		
Testicular tone	$0.24 \pm 0.04$	Low		
Scrotal circumference	$0.93 \pm 0.01$	High		
Mass activity	$0.42 \pm 0.05$	Moderate		
Motility	$0.48 \pm 0.05$	Moderate		
Normal sperm	$0.78 \pm 0.03$	High	$0.42 \pm 0.06$	Moderate
Abnormal heads	$0.57 \pm 0.05$	Moderate	$0.38 \pm 0.06$	Moderate
Abnormal midpieces	$0.65 \pm 0.04$	Moderate	$0.45 \pm 0.06$	Moderate
Abnormal tails	$0.69 \pm 0.04$	High	$0.23 \pm 0.05$	Low
Cytoplasmic droplets	$0.48 \pm 0.05$	Moderate	$0.32 \pm 0.06$	Low

Table 15.	Repeatability	(± se	) of various pl	hvsical, semen	and sperm traits

### Correlations between times for normal sperm

All correlations were significant (Table 16). The majority of these were at the P<0.001 level except for a few correlations between April 2001 and other dates with these being significant at least at the P<0.05 level. The r values increased as the bulls became older. The high levels of significance agree with the high repeatability of normal sperm for the feeding phase (r = 0.75) and the relocation phase (r = 0.78).

Table 16. Correlations between times for normal spe	erm
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	Apr 01	May 01	Aug 01	Oct 01	Nov 01	Jan 02	Feb 02	Apr 02	May 02	Jul 02	Aug 02	Sep 02
May 01	0.542***											
Aug 01	0.467***	0.690***										
Oct 01	0.475***	0.547***	0.695***									
Nov 01	0.469**	0.558***	0.729***	0.809***								
Jan 02	0.548***	0.541***	0.615***	0.750***	0.794***							
Feb 02	0.402**	0.470***	0.619***	0.584***	0.705***	0.708***						
Apr 02	0.359*	0.587***	0.567***	0.641***	0.747***	0.736***	0.807***					
May 02	0.396**	0.595***	0.658***	0.627***	0.656***	0.709***	0.788***	0.842***				
Jul 02	0.421**	0.621***	0.631***	0.670***	0.759***	0.753***	0.780***	0.808***	0.871***			
Aug 02	0.342*	0.593***	0.660***	0.635***	0.697***	0.671***	0.727***	0.775***	0.758***	0.806***		
Sep 02	0.461**	0.606***	0.679***	0.635***	0.733***	0.709***	0.807***	0.778***	0.824***	0.831***	0.764***	
Nov 02	0.332*	0.464***	0.500***	0.577***	0.613***	0.713***	0.729***	0.740***	0.761***	0.688***	0.751***	0.758***

\*P<0.05: \*\*P<0.01: \*\*\*P<0.001

### Haematology

There were significant feed x time (P<0.001), relocation x time (P<0.001) and feed x relocation (P=0.023) interactions for WBC (Table 17). Initially WBC was higher in F than in G bulls but then the reverse occurred in January 2002 and April 2002 (Figure 18). R bulls had significantly less WBC than NR bulls in January 2002 but the reverse occurred in April 2002 (Figure 19). With the subset of relocated bulls only, there was a significantly higher in RG (9.7 to  $10.8 \times 10^9$ /L) but in RF bulls there was no change in

WBC values immediately post-relocation (12.1 x  $10^{9}$ /L) compared with values prior to relocation.

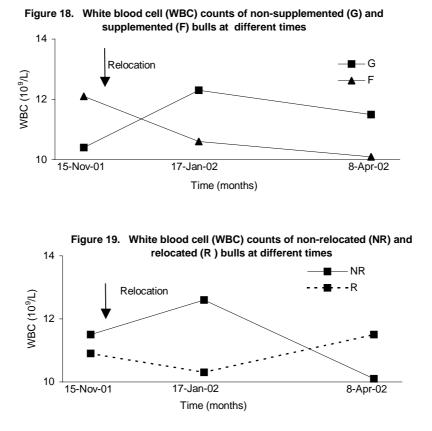
With RBC, there was a significant (P=0.036) feed x relocation x time interaction (Table 17). In November 2001, NRF and RF bulls were significantly greater than NRG and RG bulls but by January 2002, RF bulls were significantly greater than NRF and RG bulls but not NRG bulls. With the subset of relocated bulls only there was a significant (P<0.001) increase in RBC immediately post-relocation compared with values prior to relocation (9.8 to 11.5 x  $10^{12}$ /L).

Parameter	Treatment	15-Nov-01	17-Jan-02	8-Apr-02
White blood cell (10 <sup>9</sup> /L)	G	10.4a	12.3b	11.5b
	F	12.1a	10.6a	10.1a
	lsd within times	1.04		
	NR	11.5a	12.6b	10.1a
	R	10.9a	10.3a	11.5b
	lsd within times	1.04		
Red blood cell (10 <sup>12</sup> /L)	NRG	8.5a	10.2ab	8.7a
	NRF	10.8b	9.9a	9.3a
	RG	8.8a	9.3a	9.2a
	RF	10.8b	11.3b	9.5a
	lsd within times	1.3		
Packed cell volume (%)	G	41a	43a	40a
	F	51b	46a	43a
	lsd within times	4.0		
Haemoglobin (g/dL)	G	11.5a	12.1a	12.9a
	F	13.3b	13.4b	14.1b
	lsd within times	0.47		
	NR	12.5a	12.8a	12.9a
	R	12.2a	12.7a	14.2b
	lsd within times	0.47		

Table 17	Effects of	feeding and	relocation o	on haematologica	I parameters
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Means for each trait not followed by a common letter differ significantly (P<0.05)

There was a significant (P=0.003) feed x time interaction for PCV (Table 17). PCV was significantly higher in F bulls than G bulls in November 2001 and then onwards declined with no difference between treatments. With the subset of relocated bulls only, there was a significant (P<0.001) increase in PCV immediately post-relocation compared with values prior to relocation (46 to 53 % respectively).



There were significant feed x time (P=0.012) and relocation x time (P<0.001) interactions on Hb (Figures 20 and 21). There was a gradual increase of Hb in both G and F bulls with the increase greater in F. Overall means for G and F bulls were  $12.2 \pm 0.1$  and  $13.6 \pm 0.2$  g/dL respectively. Hb of R bulls increased with time whilst that of NR bulls remained relatively constant. Overall means for NR and R bulls were  $12.7 \pm 0.1$  and  $13.0 \pm 0.1$  g/dL respectively. With the subset of relocated bulls only, there was a significant (P=0.025) feeding x date interaction. Values of Hb for RG increased from 11.4 to 12.9 g/dL from pre-relocation to immediately post-relocation whilst the corresponding increase was from 13.1 to 15.3 g/dL for RF bulls.

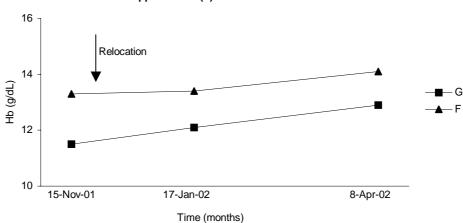
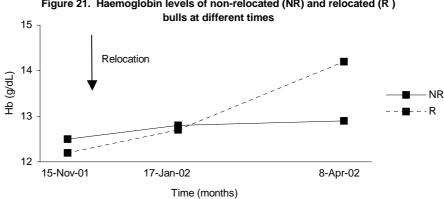


Figure 20. Haemoglobin (Hb) levels of non-supplemented (G) and supplemented (F) bulls at different times



# Figure 21. Haemoglobin levels of non-relocated (NR) and relocated (R)

### **Biochemistry** (Table 18)

With cortisol, there were no relocation effects but there was a significant (P=0.044) feed x time interaction. Cortisol levels were significantly higher in G bulls in January 2002 but there was no significant difference at other times (Figure 22). Within the subset of relocated bulls, there was no significant change in cortisol levels immediately following relocation.

CPK levels increased with time. There was a significant (P=0.030) effect of relocation with R bulls having higher CPK levels overall than NR bulls (206 v 171 nmol/L). There was a significant (P=0.017) interaction of feed x time on CPK levels with F bulls having higher CPK levels than G bulls in November 2001 but no difference at subsequent times (Figure 23). Within the subset of relocated bulls, there was no significant change in CPK levels immediately following relocation.

Parameter	Treatment	15-Nov-01	17-Jan-02	8-Apr-02	lsd
Cortisol	G	4.98a(144.7) <sup>#</sup>	5.24b(188.1)	4.80a(121.4)	
(nmol/L)	F	5.02a(151.4)	4.80a(121.3)	4.63a(102.3)	
	Isd within dates	0.30			
CPK	G	4.36a(78.5) <sup>#</sup>	5.43a(229.1)	5.83a(341.4)	
(IU/L)	F	4.71b(111.5)	5.26a(192.9)	5.80a(331.6)	
	Isd within dates	0.27			
AST	G	55.1a	90.3b	98.3a	
(IU/L)	F	68.1b	82.0a	102.5a	
	Isd within dates	7.9			
L-lactate	NR	5.9a	8.2a	6.9a	
(nmol/L)	R	6.0a	8.1a	9.7b	
	lsd within dates	1.35			
NEFA	All bulls	0.81b	0.36a	0.41a	0.06
(nmol/L)					

Table 18. Effects of feeding and relocation over time on biochemical parameters

For traits, means within rows or columns not followed by a common letter differ significantly

#both log transformed and back-transformed means shown

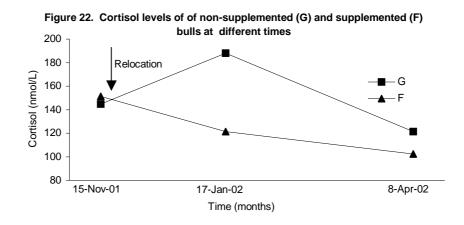
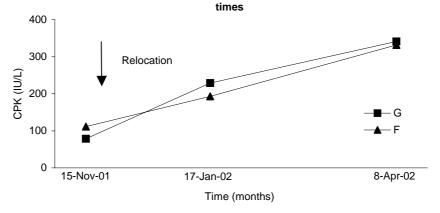
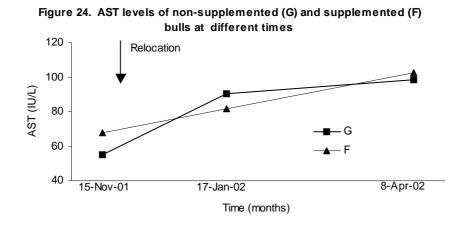


Figure 23. CPK levels of non-supplemented (G) and supplemented (F) bulls at different

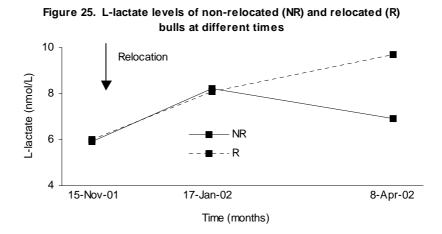


AST levels increased over time with a significant (P<0.001) feed x time interaction. F bulls had significantly higher AST levels than G bulls in November 2001, the reverse in January 2002 but there was no significant difference in April 2002 (Figure 24). Within the subset of relocated bulls, RF bulls had significantly higher AST levels than RG bulls prior to relocation (70.2 v 57.1 IU/L respectively) but not immediately after relocation (75.2 v 62.5 IU/L respectively).

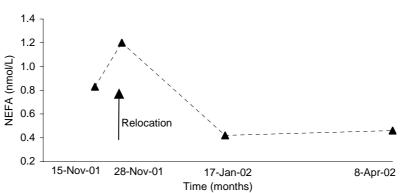


With L-lactate, there was a significant feeding (P=0.015) effect with F bulls having higher L-lactate levels than G bulls (8.1 v 6.9 nmol/L). As well there was a significant (P<0.001) relocation x time interaction

when R bulls had higher levels of L-lactate than NR bulls in April 2002, but not at other times (Figure 25). Within the subset of relocated bulls only, L-lactate levels steadily increased after relocation with values of 6.0, 7.7, 8.1 and 9.7 nmol/L respectively on 15 November 2001, 28 November 2001, 17January 2002, and 08 April 2002.



With NEFA, there were significant effects of feeding (P=0.002), relocation (P=0.031) and time (P<0.001). NEFA was significantly higher in F bulls than G bulls ( $0.59 \pm 0.03 \vee 0.46 \pm 0.03 \text{ nmol/L}$ ); in R bulls than NR bulls ( $0.57 \pm 0.03 \vee 0.48 \pm 0.03 \text{ nmol/L}$ ) and was significantly higher prior to relocation than after relocation (0.81, 0.36 and 0.41 nmol/L respectively for 15 November 2001, 17January 2002, and 08 April 2002. Within the subset of relocated bulls only, NEFA significantly increased immediately after relocation, then decreased and stayed constant (Figure 26).

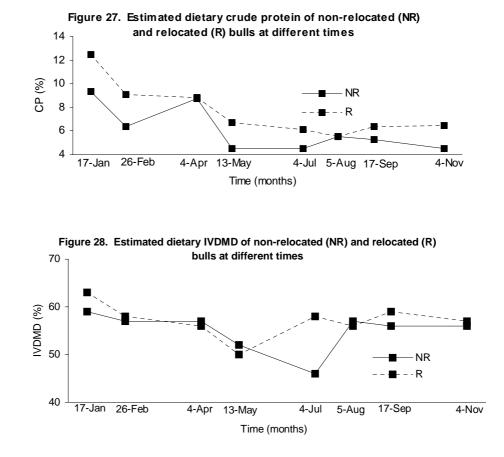


#### Figure 26. NEFA levels of relocated bulls at different times

### **Dietary intake**

Dietary measurements of CP and IVDMD by faecal NIRS were not statistically tested and thus the data gave a trend of pasture conditions at each site. Overall CP tended to be higher at Toorak (R) than at Brigalow Research Station (NR) (Figure 27). Values at Toorak steadily declined until August 2002 then plateaued. CP values at Brigalow decreased to February 2002, then increased to April 2002 then declined to May 02 then also plateaued but at a lower level than Brigalow.

Overall there was a steady decrease in IVDMD until May 2002 with values at both locations being similar (Figure 28). Then values for Toorak (R) increased, Brigalow (NR) decreased then both converged and plateaued.



# 6.1.4.6 Discussion

The study demonstrated that, at this level of feeding (about 6kg/day of pellet mix of CP 15% and 10.4 MJ/kg) there was no significant effect on semen or sperm characteristics. This is despite fed bulls gained 143 kg more, were about 2 units better in body condition and had scrotal circumferences larger by 4.3 cm than bulls grazing pasture alone. The feeding period of 150 days in many cases is considerably less than that experienced by industry bulls and the final liveweight of 521 kg of fed bulls is at the lower range of bulls presented for sale. Longer and higher levels of feeding which occur particularly in the stud industry need to be explored, as at these levels there may well be a detrimental effect on fertility.

Even at this level of feeding there were some detrimental effects on bulls. Lameness from laminitis occurred in 3 of the 40 fed bulls with one animal having to be removed from the fed group. Flight speeds were higher in F than in G bulls and this is important for temperament as animals with faster flight speeds have poorer temperament. During the feeding period F bulls were more difficult to handle in the yards and this has implications for animal welfare and for safety of handlers.

Feeding prior to relocation had no short term or long term effect on mass activity and motility of semen. With sperm morphology, there were some minor differences in some of the categories of sperm. There was no effect on normal sperm with all bulls but in the subset of bulls with 50+% normal sperm, F bulls were 5.5% units less than G bulls.

At the end of the feeding period, there were differences in both haematological and biochemical parameters but the average differences still were within normal ranges suggesting that these indices were really not good indicators of liveweight changes. WBC counts were higher in F bulls although none of these values were in the pathological range. Similarly RBC, PCV and Hb values were higher in F bulls. The biochemical parameters, AST and CPK were significantly higher in F than in G bulls at the end of the

feeding period but the overall values were mostly within normal ranges. The haematological and biochemical parameters of the 2 bulls that had laminitis were within normal ranges.

Relocation had some effect on mass activity of semen with values being generally higher in NR bulls than R bulls. This difference was pronounced at the first collection after relocation in January 2002, then in the latter part of the experiment. However there was no significant effect of relocation on semen motility and the gradual decrease in time at both sites from November 2001 to August 2002 may be related to seasonal conditions. Changes in both mass activity and motility may be of little consequence in terms of herd fertility as both of these crush-side tests are poor indicators of calf output of bulls in multiple-sire herds (Holroyd *et al.* 2002a).

Overall, relocation had minimal effect on sperm morphology. There was certainly no short term effect on normal sperm and the significantly lower levels in R bulls that occurred in September 2002, some 10 months after relocation, may well have occurred by chance. Similar trends occurred with the various classes of abnormalities in that there was generally little effect of relocation. Whilst there were minor fluctuations over time, values for normal sperm, abnormal heads, abnormal midpieces, abnormal tails and cytoplasmic droplets were the same 12 months after relocation as they were prior to relocation.

There were mixed signals from the biochemical parameters on the effect of feeding prior to relocation of bulls on their performance post relocation. Both L-lactate and NEFA were higher in F than G bulls yet cortisol and AST was higher in G than F bulls in January 2002. Biochemical and haematological parameters indicated that the relocation process was somewhat stressful with significant elevations in L-lactate (an indicator of stress), NEFA (associated with glucose depletion) and PCV (indicative of dehydration) on arrival at Toorak but interestingly there were no elevations in cortisol, CPK and AST, the latter 2 associated with muscle damage and physical exertion. However the relocated bulls may not have been coping with the Toorak environment, as there were gradual increases in CPK, AST, L-lactate and NEFA through to April 2002.

Whilst biochemical parameters indicated that R bulls may not have been coping as well with the Toorak environment, this was not reflected in the liveweight performance of the R bulls as R bulls gained more weight, were in better condition and had larger scrotal circumferences than NR bulls. The better performance of R bulls at Toorak was a probably a reflection of better dietary intakes, particularly CP, as determined by faecal NIRS. However having better liveweight, body condition and scrotal circumference should have little influence on semen and sperm morphology as these traits in 3-year-old Brahman bulls are poorly correlated with normal sperm (Fitzpatrick *et al.* 2002)

Benefits of feeding were gradually eroded with time although some advantages were retained. By November 2002 the 147 kg advantage of F bulls in November 2001 was reduced to 39 kg, there were no differences in body condition and the 4.7 cm scrotal circumference advantage was reduced to 0.4 cm.

In this study, bulls were relocated under conditions better than those experienced by many industry bulls. Bulls were not subjected to the sale yard process. Bulls had been kept as one group up to the commencement of feeding, and then as 2 separate groups during the feeding period. Prior to relocation, bulls were mustered in their separate nutritional treatments the previous afternoon at 1500 and kept on water overnight, again in their separate treatments. The next day they were loaded with bulls from the same nutritional treatment in the same truck compartment. The relocation process was relatively short with commencement at 0830 and arrival at Toorak at 0030 the following day. There they were immediately unloaded and provided access to feed and water with bulls from both treatments in the same yard. There appeared to be little fighting during relocation or on arrival although this was based on limited observations. That morning after blood sampling, bulls were then paddocked and maintained as one group throughout the rest of the experiment.

# 6.1.4.7 Conclusions

Our conclusion from this study is that providing bulls are relocated under favourable conditions, there will be minimal effect of feeding or treatment (relocation) on semen traits or sperm morphology. Any depressions in bull fertility associated with relocation are therefore likely to be from factors other than changes in semen quality, or that occur in bulls that undergo a more stressful relocation process than that experienced by the bulls in this study.

# 6.1.5 The effect of relocation on reproductive traits of Brahman and Composite bulls

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# 6.1.5.1 Summary

At Belmont Research Station in central Queensland, 38 Brahman (B) and 42 Composite (C) bulls (~24 m, and averaging 381 kg liveweight) were allocated to 2 relocation treatments: Non–relocated (NR) with bulls remaining at Belmont Research Station and bulls relocated (R) to Swan's Lagoon Research Station in north Queensland. Bulls were subjected to a physical and reproductive examination and semen collected on a 6 weekly basis from the commencement of the experiment (7 November 2001) until 12 months after relocation (28 October 2002).

There was a general increase in liveweight over time in both breeds. The pattern of liveweight gain for B and C bulls was similar in NR bulls but in the R bulls, liveweights plateaued after 6 months with B bulls heavier than C bulls in the latter part of the experiment. Twelve-month gains and final liveweights for the treatment and breed classes were 239.5 kg and 619.6 kg for NRB; 237.5 kg and 618.1 kg for NRC; 144.1 kg and 527.6 kg for RB and 108.1 kg and 486.2 kg for RC. There was no significant breed effects on scrotal circumference, whilst there was a more rapid increase of scrotal circumference of NR bulls than R bulls.

Relocation had only minor effects on mass activity and motility of semen. There was no effect of relocation or breed on normal sperm or on any of the various classes of sperm abnormalities, either in the immediate period post-relocation or over the ensuing 12-month period. However there were changes over time with normal sperm being lowest in February 2002 (57%) and highest in August 2002 (69%).

Whilst there were some differences in various haematological and biochemical parameters associated with both breed and relocation treatment, none could be attributed to relocation *per se* apart from an elevation in L-lactate on arrival at Swan's Lagoon. Bulls were relocated under conditions better than those experienced by many industry bulls. Thus, our conclusion from this study is that providing bulls are relocated under optimum conditions, there should be minimal effect of breed or relocation on semen traits or sperm morphology.

# 6.1.5.2 Aim

The aim of this experiment was to measure the effect of relocation on reproductive traits of Brahman and Composite bulls relocated from central Queensland to north Queensland.

# 6.1.5.3 Materials and methods

### Experimental design

The design was a 2-relocation treatment by 2-breed factorial. Treatments were:

- NR Non-relocated with bulls remaining at Belmont Research Station in central Queensland,
- R Bulls were relocated to Swan's Lagoon Research Station in north Queensland.
- B Brahman bulls
- C Composite bulls that were ¼ Africander ¼ Brahman ¼ Hereford ¼ Shorthorn,

There were 17 B and 20 C bulls per relocation treatment with 74 bulls in total in the experiment.

### Experimental sites and animals

The experiment was conducted at both Belmont Research Station (23<sup>o</sup>S, 150<sup>o</sup>E) in the sub-coastal central Queensland and at Swan's Lagoon Research Station (20<sup>o</sup>S, 147<sup>o</sup>E) in the sub-coastal spear grass region of north Queensland. The paddocks at Belmont were undulating open savannah woodland and consisted of a mix of improved (buffel, Rhodes, green panic oversown with seca stylo) and native (predominately black spear grass) pastures with stocking rates of 1 AE to 2.5 ha. At Swan's Lagoon, the paddocks were native pastures, with the principal species being black spear grass, tropical tall grasses such as cane grass and sorghums, cockatoo and blue grasses forming a flat savannah woodland. Stocking rates were at 1 AE to 4 ha.

The bulls were born on Belmont from October – December 1999 and had been run on pasture at Belmont until the commencement of the experiment in November 2001.

### Procedures

On 12 July 2001, 38 B and 42 C bulls were selected on the basis of scrotal circumference (minimum of 28 cm) good temperament and free of obvious physical defects from 44 B and 47 C bulls. On the 26 and 27 September 2001, these selected bulls were weighed (full), condition scored, underwent a physical and reproductive examination and had semen collected for morphological assessment. These procedures were repeated on the 7 and 8 November 2001. Within each breed, bulls were allocated to treatments in blocks using a stratified randomisation of the average of the percent normal sperm of the 2 assessments with the proviso that the liveweights in November of the 2 groups were comparable. Bulls were vaccinated against the common clostridial diseases and bovine ephemeral fever prior to relocation.

On the 26 November 2001, bulls were mustered to the yards, the treatment groups were drafted and the NR bulls were returned to the paddock. The R bulls were then trucked 630 km to Swan's Lagoon departing Belmont at 1230 and arriving at Swan's Lagoon at 2330. On arrival, bulls were placed in a yard on grassy hay and water. At 0600 next morning, the bulls were blood sampled from the jugular vein, vaccinated for botulism and then paddocked.

After relocation, bulls were mustered 8 times at 5 –7 weekly intervals, at both properties, for weighing and physical and reproductive examination with the final examination in October 2002. The examinations on each property were conducted within 2 - 3 days of each other, with all of the physical and reproductive assessments being done by the same operator throughout the experimental period. At Belmont, bulls were mustered the previous 1-2 days and held in a small paddock adjacent to the cattle yards. Bulls were mustered into the yards about 0700, weighed, and examinations commenced about 0830 and were completed by about 1600. At Swan's Lagoon, bulls were mustered from their paddock and were into the yards on the same day by about 0730, weighed with examination times similar to Belmont.

A blood sample was taken from the coccygeal vein from each bull on 7 November 2001 (prior to relocation), within 8 hours of arrival at Swan's Lagoon on the 27 November 2001 (from the jugular vein), then again at the January and March examinations (coccygeal vein) for haematology and biochemistry assay. Blood was drawn into 10mL EDTA evacuated tube (Vacutainers, Becton Dickinson) and stored in a cool-box until all samples had been collected. Total erythrocytes, leucocytes and haemoglobin concentration were determined using a Sysmex electronic counter. The sample was then centrifuged (1000g, 20 min) and five 1mL aliquots of plasma were aspirated and placed into 1mL cryotubes. Plasma samples were stored at  $-20^{\circ}$ C awaiting biochemical analysis. For biochemical analysis, a 1mL plasma aliquot from each animal was thawed and centrifuged (1000g, 20 min) to remove fibrin precipitate. Using an Olympus Reply Chemistry Analyser, each sample aliquot was tested for the following : aspartate aminotransferase (AST), creatine phosphokinase (CPK), L-lactate and non-esterified fatty acids (NEFA). Plasma cortisol levels were determined using the EIA Cortisol ELISA kit (DSL Laboratories).

After relocation, at each property a faecal sample was collected at each examination, at the time of insertion of the electroejaculation probe, from about 50% of bulls, bulked, sub-sampled, oven-dried at 60<sup>o</sup>C for determination of dietary crude protein (CP), in vitro digestibility of dry matter (IVDMD) and

grass/non-grass content by near-infrared reflectance spectroscopy (NIRS) (Lyons and Stuth 1992; Coates 1998)

### Statistical analysis

All statistical analyses were performed using GenStat for Windows 6<sup>th</sup> Edition.

### Scrotal circumference

A random coefficients regression approach was used to analyse scrotal circumference data for measurements from November 2001 to October 2002. The average growth pattern appeared to be approximately quadratic over the time span considered. However, the individual bulls have their own pathway of growth so that there is potentially variation in intercepts, slopes and curvature across bulls.

The full model fitted was

- Fixed effects: breed + treatment + time + treatment.breed + treatment.time + breed.time + time<sup>2</sup> + treatment.time<sup>2</sup> + breed.time<sup>2</sup> + treatment.breed.time<sup>2</sup>
- Random effects: bull + bull.time
- Random intercepts and slopes within bulls are correlated.

### Where

- breed has 2 levels, Brahman (B) and Composite (C),
- treatment has 2 levels, non-relocated (NR) and relocated (R),
- time is a covariate, days since 12 July 2001 (date bulls first examined),
- time<sup>2</sup> is the quadratic of time,
- bull is the variance component for random intercepts for bulls, and
- bull.time is the variance component for random slopes for bulls.

Random terms with zero variance components and non-significant fixed terms (P>0.05) were progressively removed to arrive at the final model.

### Other physical and reproductive traits

A linear mixed model was used to analyse all other traits recorded over time. The first time of measurement was generally just prior to relocation while the remaining measurements were recorded after relocation. Analysis of data from these times allows assessment of the immediate effect of relocation and also any longer term effects. The full model fitted was:

- Fixed effects: breed+treatment+time+breed.treatment+breed.time+treatment.time+ breed.treatment.time+age+breed.age+treatment.age+breed.treatment.age
- Random effects: bull+bull.time

Where

• breed has 2 levels, Brahman (B) and Composite (C),

- treatment has 2 levels, non-relocated (NR) and relocated (R),
- time is a factor with varying levels depending on when data was recorded,
- age is a covariate age of bull (days) at time of relocation (26 November 2001), and
- bull.time represents the residual variance.

The full model was fitted initially. Zero variance components were removed from the model, and then non-significant (P>0.05) fixed effects were progressively removed, except that the main effects of treatment, breed and time were always retained. Approximate least significant differences were calculated for significant (P<0.05) fixed effects.

Repeatability (intraclass correlation) was estimated as the ratio of variance components:

Repeatability = bull/(bull + bull.time)

A log (x+1) transformation was applied to abnormal heads, abnormal midpieces, abnormal tails and cytoplasmic droplets prior to analysis to stabilise the variance. Also, a log (x) transformation was applied to CPK data and log (x+0.1) to NEFA data prior to analysis.

Sperm morphology variables were analysed 3 ways:

- Including all samples with 100 sperm counted in the spermiogram (all-bulls),
- Including samples from bulls with 50+% normal sperm in November 2001 and with 100 sperm counted (50+%), and
- Including samples from bulls with 70+% normal sperm in November 2001 and with 100 sperm counted (70+%).

### Biochemical and haematological traits

In addition to the analysis outlined under 'Other physical and reproductive traits' above, a second analysis was performed for biochemical and haematological traits. Blood samples were taken from relocated bulls only immediately after relocation. This additional analysis considered relocated bulls only and compared the 2 breeds and their biochemical and haematological profiles over the 4 times of measurement for the relocated bulls. The full model fitted was:

- Fixed effects: breed \*time+age+breed.age
- Random model: bull+bull.time

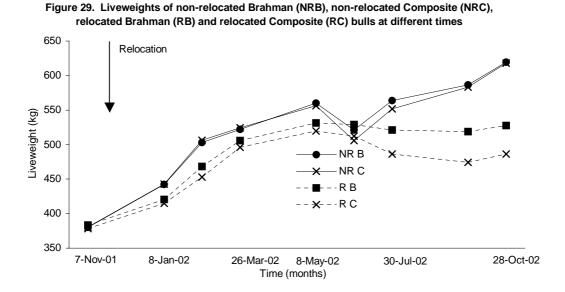
Where

- breed has 2 levels, Brahman (B) and Composite (C),
- time has 4 levels, 7 November 2001, 27 November 2001, January 2002 and March 2002,
- age is a covariate age of bull (days) at time of relocation (26 November 2001),
- bull represents the variance component for bulls, and
- bull.time represents the residual variance.

# 6.1.5.4 Results

### Liveweight

There was a significant (P<0.001) breed x relocation x time interaction. Overall, NR bulls had better liveweight gains than R bulls (Figure. 29). There was a general increase over time in both breeds, where the pattern of liveweight gain for B and C bulls was similar in non-relocated animals. In the R bulls, liveweights plateaued after 6 months and B bulls were heavier than C bulls in the latter part of the experiment. Twelve-month gains and final liveweights were respectively 239.5 kg and 619.6 kg for NRB; 237.5 kg and 618.1 kg for NRC; 144.1 kg and 527.6 kg for RB and 108.1 kg and 486.2 kg for RC.



### **Body condition score**

There were significant breed x relocation (P=0.009), breed x time (P<0.001) and relocation x time (P<0.001) interactions with body condition score. NRB bulls were in significantly better condition than other treatments with RC bulls being the poorest in condition (Table 19).

score		
Treatment	Brahman	Composite
NR	6.0c	5.3ab
R	5.4b	5.1a
lsd	0.20	

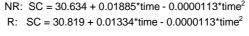
Table 19. Effect of breed and relocation on body condition score

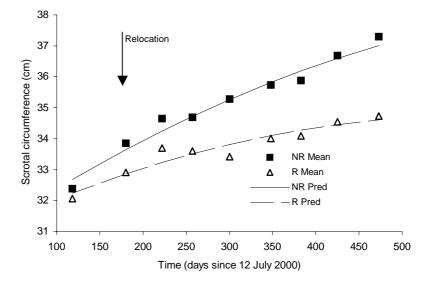
Means followed by different letters differ significantly (P<0.05)

# Scrotal circumference

There was no significant effect of breed. Scrotal circumference increased more rapidly in NR bulls than R bulls, with the response in both groups tending to plateau towards the end of the experiment (Figure 30).

# Figure 30. Scrotal circumference of non-relocated (NR) and relocated (R) bulls at different times





### **Testicular tone**

There was no breed or relocation effect on testicular tone, only a significant effect of time (P<0.001) with tone being highest (mean of 4) on 7 November 2001, prior to relocation and lowest on 8 May 2002 (mean of 3.5).

### Mass activity

There were significant breed x time (P=0.004) and relocation x time (P=0.003) interactions on mass activity (Table 20). There were no significant breed effects from November 2001 through to March 2002 but from then on, C bulls had significantly higher mass activity than B bulls. There were no differences between relocation treatments except in January 2002 when NR bulls had higher mass activity than R bulls (3.2 v 2.6 respectively).

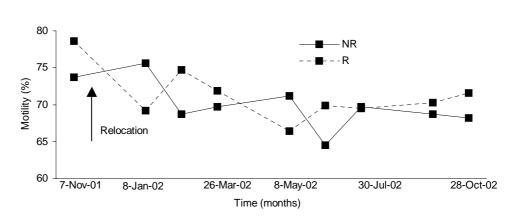
### Motility

There were significant breed x time (P=0.003) and relocation x time (P<0.001) interactions (Table 21) for motility. C bulls had significantly higher motility than B bulls prior to relocation. After relocation, there was no difference between breeds except in May 2002 and June 2002 when C bulls had significantly more motile semen than B. Motility gradually decreased over time, with the relocation x time interaction being from motility in R bulls being highest prior to relocation and lowest in May 2002 whilst in NR bulls, motility was highest in January 2002 and lowest in June 2002 (Figure 31).

Parameter	Treatment	7-Nov-01	8-Jan-02	19-Feb-02	26-Mar-02	8-May-02	25-Jun-02	30-Jul-02	10-Sep-02	28-Oct-02	lsd
Mass activity (0-5)	В	2.6a	2.9a	2.5a	2.8a	2.3a	2.3a	2.5a	2.4a	2.6a	
	С	3.0a	2.9a	2.9a	3.2a	3.3b	3.3b	3.1b	3.1b	3.1b	
	lsd within times	0.45									
	NR	2.7a	3.2b	2.6a	3.1a	2.8a	2.6a	2.8a	2.8a	2.7a	
	R	2.9a	2.6a	2.8a	2.9a	2.8a	3.0a	2.9a	2.7a	2.9a	
	Isd within times	0.45									
Motility (%)	В	72.4a	72.0a	69.7a	67.3a	63.8a	59.4a	66.8a	65.9a	66.2a	
	С	80.0b	72.8a	73.8a	74.3a	73.8b	75.0b	72.4a	73.1a	73.5a	
	lsd within times	7.4									
	NR	73.7a	75.6a	68.7a	69.7a	71.2a	64.5a	69.7a	68.7a	68.2a	
	R	78.6a	69.2a	74.7a	71.9a	66.4a	69.9a	69.5a	70.3a	71.6a	
	lsd within times	7.4									
Normal sperm (%)		61.4bc	60.7bc	56.6a	59.7b	63.0c	61.8bc	61.4bc	69.0d	61.2bc	3.1
Abnormal heads (%)	NR	3.19a(24.3)#	3.15a(23.3)	3.04a(20.9)	3.20a(24.5)	2.94a(18.9)	3.20a(24.4)	3.17a(23.7)	2.93a(18.6)	3.26a(25.9)	
	R	3.17a(23.7)	3.11a(22.4)	3.26b(26.0)	3.31a(27.4)	3.25b(25.8)	3.21a(24.8)	3.19a(24.3)	2.91a(18.4)	3.15a(23.4)	
	Isd within times	0.2									
Abnormal mid-pieces (%)		1.39ab(3.0)#	1.47abc(3.4)	1.51bc(3.5)	1.30a(2.7)	1.42ab(3.1)	1.56bc(3.8)	1.64bc(4.1)	1.48abc(3.4)	1.58bc(3.9)	0.21
Abnormal tails (%)		0.43ab(0.5)	0.46abc(0.6)	0.39ab(0.5)	0.43ab(0.5)	0.31a(0.4)	0.48abc(0.6)	0.57bc(0.8)	0.62c(0.9)	0.38a(0.5)	0.19
Cytoplasmic droplets (%)	NR	1.83a(5.2)#	1.68a(4.4)	2.17a(7.8)	1.76a(4.8)	1.77a(4.9)	1.10a(2.0)	1.24a(2.5)	1.13a(2.1)	1.27a(2.6)	
	R	1.66a(4.3)	1.93a(5.9)	1.85a(5.4)	1.55a(3.7)	1.49a(3.4)	1.28a(2.6)	1.56a(3.8)	1.43a(3.2)	1.78b(4.9)	
	lsd within times	0.38									

#### Table 20. Means of semen and sperm traits at different times

For traits, means within rows or columns not followed by a common letter differ significantly; Both log (x+1) abd back transformed values presented

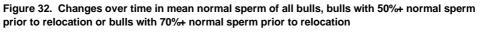


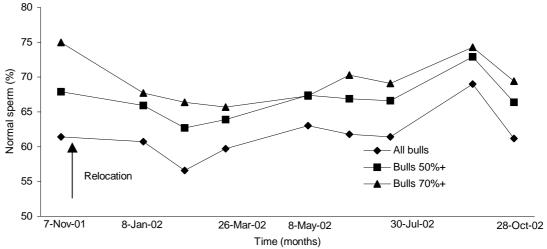
# Figure 31. Semen motility of non-relocated (NR) and relocated (R) bulls at different times

### Sperm morphology

### Normal sperm

With all-bulls (n = 74), there was no effect of breed or relocation on changes in normal sperm. There was a significant (P<0.001) effect of time (Table 20) with values being lowest in February 2002 and highest in September 2002. Similar trends occurred when the analyses included only bulls that had 50+% (60 bulls) or 70+% (30 bulls) normal sperm prior to relocation (Figure 32).





### Sperm abnormalities

The levels of significance and interaction of the different categories of abnormalities are presented in Table 21. The trends for the various categories of bulls (all-bulls, 50+% and 70+%) are similar except for some minor differences; eg there was no significant time effect for abnormal midpieces for 70+% bulls whilst for the other 2 categories time was significant. There was a breed x time interaction for droplets in 70+% bulls but not in the other 2 categories and there were significant relocation x time interactions for

droplets for all bulls and 50+% bulls but not in the 70+% bulls. Therefore for simplicity only data of the allbull category has been presented for changes in the various classes of abnormalities.

Abnormality	Category	Breed	Relocation	Time	Breed x Relocation	Breed x time	Relocation x time
Heads	All bulls	ns (P=0.828)	ns (P=0.452)	*** (P<0.001)			*** (P=0.001)
	50+% <sup>1</sup>	ns (P=0.686)	ns (P=0.955)	*** (P<0.001)			** (P=0.003)
	70+% <sup>2</sup>	ns (P=0.153)	ns (P=0.800)	** (P=0.002)	* (P=0.011)		
Midpieces	All bulls	ns (P=0.825)	ns (P=0.631)	* (P=0.043)			
	50+%	ns (P=0.350)	ns (P=0.661)	* (P=0.047)			
	70+%	ns (P=0.647)	ns (P=0.451)	ns (P=0.258)			
Tails	All bulls	ns (P=0.760)	ns (P=0.218)	* (P=0.036)			
	50+%	ns (P=0.318)	ns (P=0.319)	* (P=0.016)			
	70+%	ns (P=0.367)	ns (P=0.233)	** (P=0.008)			
Droplets	All bulls	ns (P=0.057)	ns (P=0.651)	*** (P<0.001)			*** (P<0.001)
	50+%	ns (P=0.096)	ns (P=0.660)	*** (P<0.001)			*** (P=0.001)
	70+%	ns (P=0.211)	ns (P=0.410)	*** (P<0.001)		* (P=0.016)	

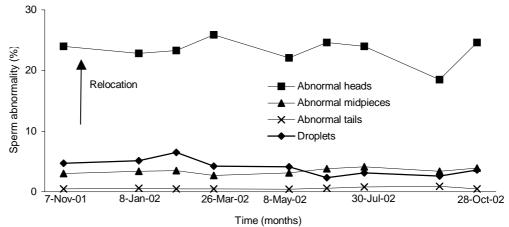
 Table 21. Levels of significance and interactions for sperm abnormalities

<sup>1</sup>Analyses included only bulls that had at least 50% or more normal sperm at time of relocation

<sup>2</sup>Analyses included only bulls that had at least 70% or more normal sperm at time of relocation

The mean levels of abnormal heads ranged from a maximum of 25.9% in March 2002 to 18.5% in September 2002 (Figure 33). There was no significant effect of breed but a significant relocation x time (P=0.001) interaction effect on abnormal heads (Table 21) where abnormal heads were significantly greater in R than NR bulls in February and May 2002. There was only a significant effect of time on both midpiece (P=0.043) and tail abnormalities (P=0.036) (Table 21) but overall mean values for midpieces and tails were low and averaged 3.4% and 0.6% respectively. Mean values for droplets ranged from 6.6% in February 2002 to 2.3% in June 2002. There was no significant difference between treatments at any time except October 2002 when R bulls had higher mean droplets than NR bulls.





### **Repeatability of reproductive traits**

With all bulls, repeatability of testicular tone and mass activity was low whilst motility was moderate and normal sperm was high (Table 22). The various categories of abnormalities tended to be in the moderate range of repeatability except for abnormal tails, which was low. Repeatability of morphology traits declined within the subsets of bulls having either 50+% or 70+% normal sperm prior to relocation, compared with the all-bulls category.

Attribute	All bulls	Bull with 50+% normal sperm	Bulls with 70+% normal sperm
Testicular tone	$0.34 \pm 0.05$		
Mass activity	$0.32 \pm 0.05$		
Motility	$0.61 \pm 0.05$		
Normal sperm	$0.69 \pm 0.04$	$0.26 \pm 0.05$	0.21 ± 0.07
Abnormal heads	$0.48 \pm 0.05$	$0.22 \pm 0.05$	$0.06 \pm 0.05$
Abnormal midpieces	$0.43 \pm 0.05$	$0.34 \pm 0.05$	0.31 ± 0.08
Abnormal tails	$0.27 \pm 0.05$	$0.20 \pm 0.05$	0.16 ± 0.06
Cytoplasmic droplets	$0.35 \pm 0.05$	$0.28 \pm 0.05$	$0.29 \pm 0.08$

Table 22. Repeatability of reproductive traits

### Correlations between times for normal sperm

All correlations were highly significant (P>0.001) with r values ranging from 0.538 to 0.773 (Table 23).

Table 23.	Correlation	ns between	times for	normal spe	erm				
	Sep-01	Nov-01	Jan-02	Feb-02	Mar-02	May-02	Jun-02	Jul-02	Sep-02
Nov-01	0.538***								
Jan-02	0.664***	0.691***							
Feb-02	0.588***	0.642***	0.687***						
Mar-02	0.609***	0.562***	0.629***	0.665***					
May-02	0.613***	0.544***	0.659***	0.586***	0.722***				
Jun-02	0.659***	0.679***	0.711***	0.606***	0.677***	0.692***			
Jul-02	0.609***	0.697***	0.697***	0.561***	0.668***	0.668***	0.761***		
Sep-02	0.696***	0.544***	0.657***	0.555***	0.702***	0.701***	0.652***	0.689***	
Oct-02	0.603***	0.669***	0.654***	0.743***	0.722***	0.665***	0.757***	0.773***	0.699***

able 23. Correlations between times for normal sperm

### Haematology

With white blood cell (WBC) counts, there were significant relocation x time (P<0.003) and breed x time (P<0.001) interactions (Table 24). WBC was higher in R than NR bulls after relocation and C bulls had higher WBC than B bulls in January 2002 but no difference at other times. With red blood cell (RBC) counts, there was a significant (P<0.001) effect of breed and a significant relocation x time (P=0.018) interaction. RBC counts ( $10^{12}$ /L) were significantly (P<0.001) higher in B than C bulls (8.7 ± 0.2 v 7.5 ± 0.2) and significantly higher in R than NR after relocation but not before relocation. With packed cell volume (PCV), there was a significant (P<0.001) effect of time with a significant (P=0.023) breed x

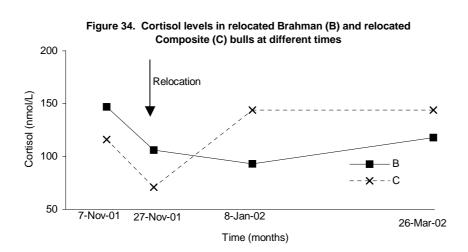
relocation interaction (Table 24). PCV was significantly lower in NRC bulls (33%) than other treatments (37 – 38%). With haemoglobin (Hb), there was a significant (P<0.001) effect of time with a significant (P=0.023) breed x relocation interaction. NRC bulls had significantly lower Hb values than RC bulls which in turn were significantly less than both NRB and RB bulls (Table 24).

Trait	Treatment	7-Nov-01	8-Jan-02	26-Mar-02
WBC (10 <sup>9</sup> /L)	NR	11.3a	12.8a	11.8a
	R	11.3a	14.1b	13.8b
	Isd within dates	1.03		
	В	11.5a	12.7a	13.1a
	С	11.2a	14.2b	12.5a
	lsd within dates	1.01		
RBC (10 <sup>12</sup> /L)	NR	7.0a	7.9a	8.1a
	R	7.1a	9.1b	9.4b
	Isd within dates	0.72		

Table 24.		white blood	cell (WBC	) and red	blood cell	counts at o	different
	times.						

### **Biochemistry**

There was a significant (P<0.001) relocation x time interaction with cortisol in all bulls. NR bulls had significantly higher cortisol levels than R bulls in January 2002 but not at the other times (Table 25). With the subset of relocated bulls there was a significant (P=0.045) breed x time interaction with no significant change in cortisol levels of B bulls but there was a significant reduction immediately after relocation in C bulls (Figure 34).



There was a significant (P<0.001) relocation x time interaction for CPK with NR bulls having significantly higher CPK than R bulls in March 2002 but not at the other 2 times (Table 25). With the subset of relocated bulls, CPK significantly (P<0.001) declined immediately following relocation then increased through to March 2002 (Figure 35).

Trait	Treatment	7-Nov-01	8-Jan-02	26-Mar-02
Cortisol	NR	4.83a (125) <sup>#</sup>	5.84b (343)	4.80a (121)
(nmol/L)	R	4.86a (129)	4.76a (117)	4.88a (131)
	lsd	0.38	0.38	0.38
СРК	NR	4.69a(109) <sup>#</sup>	5.22a(185)	6.13b(459)
(IU/L)	R	4.86a(128)	4.94a(140)	5.39a(219)
	lsd	0.29	0.29	0.29
AST	NR	66a	101b	115b
(IU/L)	R	66a	78a	84a
	lsd	7.9	7.9	7.9
L-lactate	NR	4.5a	8.2b	9.5a
(nmol/L)	R	4.4a	5.8a	4.9a
	lsd	0.95	0.95	0.95
NEFA	NR	-0.49a (0.51) <sup>#</sup>	-1.42b (0.14)	-0.32a (0.63)
(nmol/L)	R	-0.51a (0.50)	-0.88a (0.32)	-0.40a (0.57)
	lsd	0.12	0.12	0.12
	В	-0.64b (0.43) <sup>#</sup>	-1.01a (0.26)	-0.35a (0.61)
	С	-0.36a (0.60)	-1.29b (0.18)	-0.37a (0.59)
	lsd	0.13	0.13	0.13

Table 25. Means of biochemical parameters at different times

Within columns, means not followed by a common letter differ significantly (P<0.05); # Back-transformed means presented

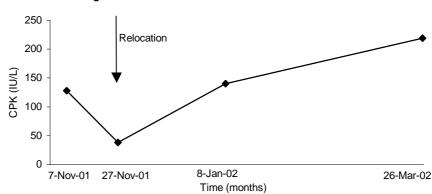
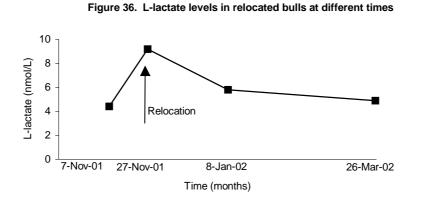


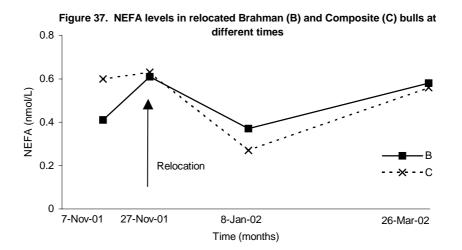
Figure 35. CPK levels in relocated bulls at different times

For AST there was a significant (P=0.024) breed x relocation interaction with means for the interaction being 103 and 79 for NRC and RC respectively and 85 and 73 for NRB and NRB respectively (Isd = 8.0). As well there was a significant (P<0.001) relocation x time interaction where NR bulls had significantly higher AST than R bulls in January and March 2002 than in November 2001, prior to relocation. There was no significant change in AST immediately post-relocation.

With all bulls there was a significant (P<0.001) relocation x time interaction with L-lactate where NR bulls had significantly higher L-lactate levels than R bulls in January and March 2002 but not in November 2001 (Table 25). With the subset of relocated bulls, L-lactate levels increased immediately after relocation and then declined to pre-relocation levels by March 2002. (Figure 36).



With NEFA, there was both a significant breed x time (P<0.001) and relocation x time (P<0.001) interaction in all bulls. NEFA was significantly higher in R than NR bulls in January 2002 but not at other times (Table 25). C bulls had significantly higher levels of NEFA than B bulls in November 2001, the reverse in January 2002 with no difference by March 2002 (Table 25). With the subset of relocated bulls only, there was a breed x time interaction with C having significantly higher levels than B pre-relocation and in January 2002, but there was no difference between breeds immediately post-relocation or in March 2002 (Figure 37).



### **Dietary intake**

Dietary measurements of CP and IVDMD by faecal NIRS were not statistically tested and thus the data gave an indication of pasture quality at each site. Overall CP of the diet was greater at Belmont than at

Swan's Lagoon. CP increased at both locations until February 2002 and then steadily declined until May 2002 (Figure 38). From there, the levels at Belmont fluctuated but the overall trend was to rise whilst the levels at Swan's Lagoon kept declining until September 2002 then increased. IVDMD values were higher throughout at Belmont than at Swan's Lagoon with values steadily declining until June–August then increasing at both sites (Figure 39).

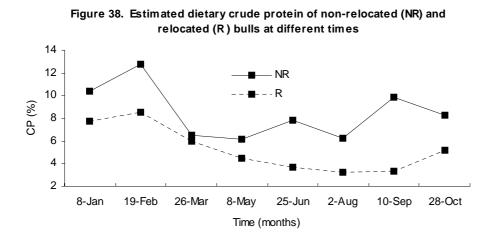
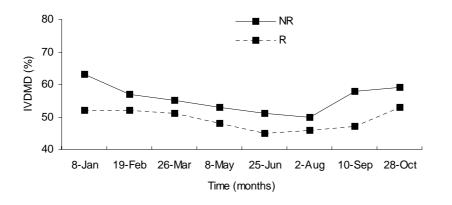


Figure 39. Estimated dietary IVDMD of non-relocated (NR) and relocated (R) bulls at different times



### 6.1.5.5 Discussion

Relocation had only a minor effect on semen traits. Mass activity was only less in R bulls at the first collection after relocation (3.2 v 2.6) but from then on there was no difference at either site. There was no effect of relocation on semen motility but there was a breed difference, independent of relocation where semen motility was better in C than B at 3 of the 9 examinations. As well there was an irregular depression in motility in both treatments until August 2002 and this is possibly related to seasonal conditions. These minor differences in mass activity and motility may be of little consequence in terms of herd reproductive performance as both of these crush-side tests are poor indicators of calf output of bulls in multiple sire herds (Holroyd et al 2002a).

There was no effect of relocation or breed on normal sperm or in any of the various classes of sperm abnormalities, either in the immediate period post-relocation or over the 12-month period. However there were changes over time with normal sperm being lowest in February 2002 (57%) and highest in August 2002 (69%). This increase in normal sperm through the winter months coincided with the decline in motility supporting previous work (Fitzpatrick et al 2002) that normal sperm is poorly correlated with motility.

Pasture conditions at Belmont appeared better than at Swan's Lagoon. In the 12-month period, the NR bulls at Belmont gained 238 kg compared to the R bulls at Swan's gaining 126 kg. The pattern of liveweight for the B and C bulls was similar for the non-relocated bulls, but for the relocated bulls, B bulls were significantly heavier than C bulls in the latter stages of the experiment. Changes in body condition mirrored liveweight whilst there were no treatment or breed differences in testicular tone. Whilst scrotal circumference increased over time, the increase was greatest in NR bulls. Despite the generally inferior performance of R compared with NR bulls in most physical traits, this inferiority was not reflected in differences in semen or sperm characteristics. Again this agrees with previous studies that physical traits such as liveweight, condition score, testicular tone and scrotal circumference are poorly correlated with semen traits and sperm morphology (Fitzpatrick et al. 2002).

Blood samples taken pre-relocation, on arrival (R bulls only) and twice in the first 3 month post-relocation for haematological and biochemical assessment were used as indicators of both acute and chronic stress associated with transport injury and adaptation to a new environment. Whilst there were some differences in these various parameters associated with both breed and relocation treatment, none could be attributed to relocation *per se* apart from an elevation in L-lactate on arrival at Swan's Lagoon.

In this study, bulls were relocated under conditions better than those experienced by many industry bulls. Prior to transportation, bulls had been kept as one group so that presumably dominance ranking would have been relatively stable. Bulls were not subjected to the sale yard or mixing with strange animals. The transport process was relatively short. Bulls were yarded at 0800, drafted and trucked at 1230, arriving at Swans Lagoon at 2300 where they were placed onto feed and water. The next morning after blood sampling, bulls were then paddocked and maintained as one group for the duration of the experiment.

# Conclusions

Our conclusion is that, providing bulls are relocated under favourable conditions, there appears to be neither a breed nor treatment (relocation) effect on sperm morphology. Any depressions in reproductive traits associated with relocation are therefore likely to be from factors other than semen quality or that occur in bulls that undergo a more stressful relocation process than that experienced by bulls in this study.

# 6.2 Increasing the working life of bulls – Identifying criteria whereby bulls can be selected for fertility at an early age and evaluation of yearling mating

# 6.2.1 Background

Selection and use of bulls as yearlings (12-15 months of age) appears to have a number of advantages. Firstly, it can accelerate annual genetic improvement by reducing the generation interval. Secondly, younger bulls are easier to handle than older bulls. Thirdly, relocating bulls as yearlings and thus allowing them to adapt to their new environment, may prevent some of the perceived subfertility issues associated with relocation. Also Mackinnon *et al.* (1990) suggested that selection for increased testis size early in life might improve reproductive rates of female progeny by reducing age of puberty.

There is a large variation in reported age of puberty in bulls of tropically adapted genotypes. In a histological study of testes obtained by castration, Christensen *et al.* (1980) studied the presence of lumen formation in the seminiferous cords in a number of genotypes at Belmont Research Station. The earliest age at which patency was seen was at 26 weeks for both Africander cross and Brahman cross bulls and the authors concluded that well grown bulls of these genotypes would be capable, at 43 weeks of age, of fertilising cows. Estimates of later puberty were recorded by Wildeus *et al.* (1984) in Brahman cross and Sahiwal cross bulls at Swan's Lagoon Research Station. Estimation of puberty in this latter study was based on the first collection, by electroejaculation, of an ejaculate containing 50 x  $10^6$  sperm with at least 10% motility. This occurred in 25% of the bulls at 15.3 months of age and from 84% of bulls by 2 years of age. Despite the apparent effect of poor nutrition on pubertal development (Wildeus *et al.* 1984), both of these studies indicate that a proportion of *Bos indicus* bulls are sexually mature as yearlings. However neither these studies nor any other studies indicate whether this early maturity is related to subsequent calf output when these bulls are mated.

There have been a number of studies that have related traits in yearling bulls with later life reproductive performance. In a study of Hereford bulls, Price and Wallach (1991) found that some animals in serving capacity tests that had low libido at 12 months showed satisfactory libido at 18-24 months of age. Farid *et al.* (1987) found that with bulls used in single–sire matings, there was no consistent relationship between fertility, measured as pregnancy rates and calving dates, of a yearling bull with that when it was a 2-year-old bull. A review by D'Occhio and Kinder (1993) indicated that behavioural indices of fertility in yearling bulls appeared to bear no relationship to aggressive or sexual behaviours in later life. The study by Wildeus *et al.* (1984) in yearling *Bos indicus* cross bulls showed that there was a biphasic pattern of development in sperm morphology. This suggested that sperm morphology in yearling bulls may have a low repeatability although the data of Wildeus *et al.* (1984) did not specifically demonstrate this. None of the above studies attempted to relate reproductive traits in yearling bulls with calf output.

Selection of bulls for the northern beef industry is usually done at 2 to 3 years of age. There is increasing interest in selecting bulls at a younger age for several reasons. Firstly, this will decrease the generation interval by mating bulls at a younger age and thus speed up genetic improvement of the herd. Secondly, younger bulls are easier to handle and pose fewer problems for mustering and handling compared to older bulls. Thirdly, potential sires can be identified and moved to their new environment so that they can adapt to these conditions prior to mating. We hypothesise one way to reduce any fertility problems associated with relocation is to encourage industry to buy yearling bulls so that they have time to adapt before maturity.

There has been little work done on yearling *Bos indicus* bulls to identify whether any physical or reproductive traits can predict their later life fertility such as calf output, particularly in multiple-sire herds. Such traits include structural soundness, scrotal circumference and semen quality and sperm morphology. Also, can these traits be identified in yearling bulls rather than identifying these traits prior to mating as a two-year old?

Our hypothesis was that there are measurable traits in yearling bulls that reflect their fertility both as yearlings and as 2-year-olds. Studies focused on measuring a number of traits including sperm

morphology.

# 6.2.2 Sexual development in Brahman and Composite bulls – Belmont Research Station

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# 6.2.2.1 Summary

The experiment was conducted at Belmont Research Station in central Queensland. Forty-one Brahman (B) and 29 Composite (C) bulls were subjected to a physical and reproductive examination and semen collected, apart from the first examination, on a 4-6 weekly basis from 25 November 1999 (392 days of age) to 29 August 2000 (670 days of age). Mean ages of bulls at the start of the experiment were 385 and 402 days for B and C bulls respectively.

There was an increase in liveweight with age for both breeds. Gains and final liveweights for B and C bulls were respectively 175 and 441 kg and 166 and 463 kg. C bulls had larger scrotal circumferences throughout although B bulls tended to catch up. The initial difference in scrotal circumference of 3.4 cm was reduced to 0.9 cm at 670 days of age (32.8 v 31.9 cm) for C and B bulls respectively.

Mass activity but not motility of semen increased with age. C bulls had significantly better mass activity and motility than B bulls whilst sperm concentrations were less in B than C bulls until 602 days of age, and then similar thereafter. Older bulls had higher mass activity, motility (B bulls only) and sperm concentrations. With normal sperm, B bulls had significantly lower levels at 433 days of age at the first collection than C bulls but there was no difference subsequently. Whilst there was some evidence of an increase in normal sperm with age in B bulls, values for C bulls remained relatively constant. There were no significant breed effects on the various categories of sperm abnormalities. Younger bulls had a higher incidence of distal cytoplasmic droplets.

The repeatability of sheath score was moderate whilst all other physical, semen and morphology traits were low. A number of physical and seminal traits were significantly correlated in both breeds although the relationship of normal sperm with other physical and seminal traits was either low or non-significant. More C than B bulls were producing 50+% normal sperm at 433 - 490 days (14.2 - 16.1 months) of age, but from then on there was no difference between breeds. There was generally little relationship between normal sperm from one observation to the next.

C bulls reached puberty significantly (P<0.001) earlier than B bulls (462 v 521 days of age i.e. 15.4 v 17.4 months) with a range of 180 days for C bulls and 315 days for B bulls. There was no relationship between puberty class (early, mid or late) and subsequent level of normal sperm at either 629 or 670 days of age.

# 6.2.2.2 Aim

The aims of the experiment were to measure physical and reproductive traits in Brahman and Composite yearling bulls to estimate puberty and to measure the repeatability and correlation of these traits with each other.

# 6.2.2.3 Materials and methods

# Experimental design

The experiment was an unbalanced longitudinal study of monthly changes in physical and reproductive

traits of 2 breeds. The animal was the experimental unit.

### Experimental sites and animals

The experiment was conducted at Belmont Research Station (23<sup>o</sup>S, 150<sup>o</sup>E) in sub-coastal central Queensland. The paddocks were flat open savannah woodland and consisted of a mix of improved (buffel, Rhodes, green panic) and native pastures (predominantly black spear grass and blue grasses) with stocking rates of 1 AE to 2.5 ha.

The bulls were born on Belmont between late August and mid December 1998, weaned in May 1999 and were run on pasture until the commencement of the experiment in November 1999. The bulls were either Brahman (B) or Composites (C), the latter mainly Belmont Red (½ Africander, ¼ Hereford ¼ Shorthorn) as well as some ¼ Africander ¼ Brahman ¼ Hereford ¼ Shorthorn composites.

### Procedures

On 25 November 1999, 41 B bulls (mean age and range of 385 and 330-451 days respectively) and 29 C bulls (mean age and range of 402 and 334-451 days respectively) were selected from the bull herd, these numbers representing the majority of bulls available of each breed for the study. The experimental bulls were weighed, scrotal contents palpated and circumference measured and returned to their paddock which was adjacent to the cattle yards.

On 5 January 2000, bulls were mustered to the yards by 0730, about 40 were retained and the balance returned to an adjacent paddock. Bulls were then weighed (full) then subjected to a physical and reproductive examination and semen collected for both a crush-side assessment of mass activity and motility and morphological assessment. Bulls were returned to their paddock about 1700 h. The following day, the remaining bulls were mustered to the yards and the procedures repeated. Bulls were then mustered at 4-6 weekly intervals until August 2000 and examined over a 2-day period, as previously described.

### Statistical analyses

All statistical analyses were performed using GenStat for Windows 6<sup>th</sup> Edition.

### Scrotal circumference

A random (quadratic) coefficient regression analysis was used to model the scrotal circumference data across ages. This allowed

- The mean scrotal circumference pattern to be generally quadratic but dependent on breed,
- The intercepts, slopes and quadratic coefficients for individual bulls to vary about the breed average, and
- The intercepts, slopes and quadratic coefficients to be correlated within bulls.

The full model fitted was

- Fixed effects:  $breed + age + breed.age + age^2 + breed.age^2 + birthdate$
- Random effects: *bull* + *bull.age* + *bull.age*<sup>2</sup> + *dev(age)* + *dev(breed.age)*

Where

• breed has 2 levels, B (Brahman) and C (Composite),

- age is the mean age at measurement,
- *birthdate* represents birth date of each of the bulls and is measured as days since 1 January 1900,
- *bull* is the variance component for random intercepts for bulls,
- *bull.age* is the variance component for random slopes for bulls,
- *bull.age*<sup>2</sup> is the variance component for random quadratic coefficients for bulls,
- *dev(age)* is a random lack of fit term for additional fluctuations about the linear trend, and
- *dev(breed.age)* is a random lack of fit term for additional fluctuations about the linear trend for each breed.

Random terms with zero variance components were removed from the model, then Wald tests were used to identify non-significant fixed terms (P>0.05), which were also removed to arrive at the final model.

### Liveweight

The analysis of liveweight was similar to that for scrotal circumference except that the response was linear rather than quadratic. Hence the full model fitted had all *age*<sup>2</sup> terms removed.

### Other traits

The remaining traits did not tend to follow a smooth pattern with time. Therefore, the following linear mixed model was used to analyse these traits. A log transformation was applied to semen concentration data and a log (x+1) transformation applied to abnormal heads, abnormal midpieces, proximal cytoplasmic droplets and distal cytoplasmic droplets prior to analysis to stabilise the variance. The full model fitted was:

- Fixed effects: *breed+age+breed.age+birthdate+breed.birthdate*
- Random effects: *bull+bull.age*

#### Where

- breed has 2 levels, B (Brahman) and C (Composite),
- *age* is a factor generally with 8 levels; 285, 327, 362, 390, 423, 466, 494, 528, 636, and 685 days of age,
- *birthdate* represents the birth date of each bulls and is measured as days since 1 January 1900,
- bull represents the variance component for individual bulls, and
- *bull.age* is the residual variance.

In all linear mixed models, random terms with zero variance components were removed from the model, then Wald tests were used to identify non-significant fixed terms (P>0.05) which were also removed to arrive at the final model. Approximate least significant differences were calculated for significant (P<0.05) fixed effects.

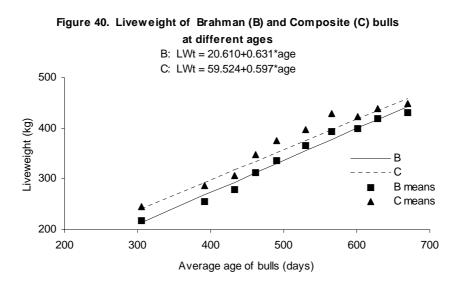
Repeatability (intraclass correlation) was estimated as the ratio of variance components:

Repeatability = bull/(bull + bull.age)

# 6.2.2.4 Results

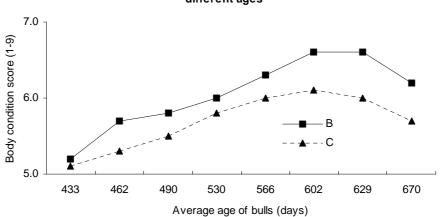
### Liveweight

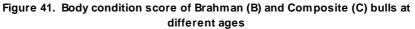
Liveweight responses were linear with C bulls being heavier than B bulls throughout. C bulls were 31 kg heavier than B bulls at the start of the experiment with gains and final liveweights for B and C bulls being respectively 175 and 430 kg and 162 and 449 kg. Fitted growth patterns for liveweight are presented in Figure 40.



### **Body condition score**

There was a significant (P=0.006) breed x date interaction. Both breeds had similar body condition at 433 days of age (14.2 months) but in subsequent months B bulls were in better body condition than C bulls (Figure 41). Condition improved in both breeds until 602 days of age with greater improvement in B bulls. Condition then decreased until 670 days of age in both breeds.





### Scrotal circumference

The average scrotal growth pattern was approximately quadratic with age for the majority of bulls (Figure 42). Composite bulls had larger scrotal circumferences throughout although B bulls tended to catch up. The initial difference in scrotal circumference of 3.8 cm at 433 days of age was reduced to 0.6 cm at 670 days of age (32.7 v 32.1 cm for C and B bulls respectively).

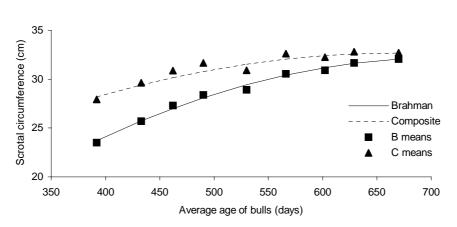


Figure 42. Scrotal circumference of Brahman (B) and Composite (C) bulls at different ages

### Testicular tone.

Age was the only significant fixed effect (P<0.001) with predicted means for B and C bulls being  $3.7 \pm 0.04$  and  $3.8 \pm 0.05$  respectively. Mean tone varied with age with values being lowest at 490 days of age (Figure 43).

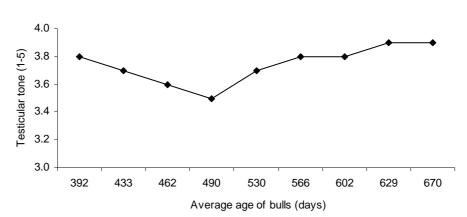


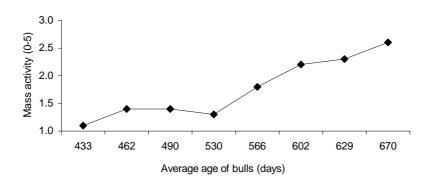
Figure 43. Testicular tone of all bulls at different ages

# Sheath score

The only significant effect was breed (P<0.001). Predicted means were 2.6  $\pm$  0.1 (n = 336) and 8.0  $\pm$  0.2 (n = 211) for B and C bulls respectively.

## Mass activity

There were significant effects of breed (P<0.001), date (P<0.001) and birthdate (P=0.023) (Table 26). C bulls had higher levels of mass activity than B throughout  $(1.3 \pm 0.1 \text{ and } 2.2 \pm 0.1 \text{ respectively})$ . Until 530 days of age, values were relatively constant but from then on they increased markedly (Figure 44). The coefficient for birthdate was  $-0.006 \pm 0.003$  indicating that older bulls had higher mass activity.



#### Figure 44. Semen mass activity of all bulls at different ages

# Motility

There was a significant (P<0.001) effect of date with a significant (P<0.001) breed x birthdate interaction (Table 26). C bulls had significantly (P<0.001) higher motility than B bulls (71.7  $\pm$  2.1% and 60.2  $\pm$  1.7% respectively). There was no clear trend with age for motility (Table 26). The coefficient for birthdate was  $-0.26 \pm 0.05$  (P=0.001) for B bulls (older bulls had higher motility) but with C bulls, the coefficient was not significant (0.05  $\pm$  0.09).

## Semen concentration

There were significant effects of birthdate (P=0.012) with a significant (P=0.002) breed x date interaction (Table 26). Concentrations were less in B than C bulls until 602 days of age and then similar thereafter. In C bulls there was little change in concentration with age whereas in B bulls there was a trend for concentrations to increase with age (Figure 45). Concentrations were higher in older bulls with the regression coefficient for birthdate being  $-0.010 \pm 0.004$ .

# Sperm morphology

## Normal sperm

There was a significant (P=0.008) breed x date interaction (Table 26). B bulls had significantly lower normal sperm at 433 days of age than C bulls but there was no difference subsequently. Whilst there was some evidence of an increase in normal sperm with age in B bulls, C bulls remained relatively constant (Figure 46).

Trait	Breed	n	5-Jan-00	3-Feb-00	3-Mar-00	11-Apr-00	17-May-00	23-Jun-00	20-Jul-00	29-Aug-00
			433 days	462 days	490 days	530 days	566 days	602 days	629 days	670 days
Mass activity (0-5)	All bulls	48-69	1.1±0.2a	1.4±0.2a	1.4±0.2ab	1.3±0.2a	1.8±0.2b	2.2±0.1c	2.3±0.2c	2.6±0.2c
Motility (%)	All bulls	48-69	60.7±2.5ab	74.7±2.7e	58.9±2.4a	56.6±2.3a	65.9±2.4bc	67.1±2.3cd	71.1±2.3cde	72.6±2.3de
Concentration (10 <sup>6</sup> /L)	в	12-41	3.49±0.41ab (32.7)#	2.91±0.29a (18.3)	3.79±0.25b (44.3)	3.99±0.25b(54.3)	4.190.25bc (66.3)	5.12±0.25de (168.0)	5.11±0.25de (164.8)	5.59±0.25de (267.2)
	С	20-28	4.83±0.31cd (125.1)	5.13±0.34de (168.7)	4.94±0.32cd (139.5)	5.01±0.29d (149.5)	5.11±0.32de (165.5)	5.43±0.29de (228.4)	5.50±0.29de (244.9)	5.79±0.30e (328.3)
Normal sperm (%)	В	10-38	41.8±4.8a	62.1±3.4bc	67.0±2.7bcd	63.0±2.5bc	60.2±2.9b	61.9±2.5b	68.9±2.5cd	71.6±2.6d
	С	19-27	62.6±3.2bc	66.1±3.5bcd	67.6±3.2bcd	61.0±3.0b	59.4±3.3b	59.8b±3.1	62.1bc±3.0	67.2±3.2bcd
Abnormal heads (%)	All bulls	34-64	1.98 ±0.16c(6.2)##	2.04±0.15c (6.7)	1.35±0.13a (2.9)	2.06±0.12c (6.8)	1.89±0.13bc (5.6)	1.59±0.12ab (3.9)	1.55±0.12a (3.7)	1.38±0.12a (3.0)
Abnormal midpieces (%	) All bulls	34-64	1.85±0.14a(5.4)##	2.13±0.13ab (7.4)	2.50±0.11cd (11.2)	2.13±0.10 (7.4)	2.37±0.11bcd (9.7)	2.26±0.10bc (8.6)	2.59±0.10d (12.3)	2.66±0.11d (13.3)
Proximal droplets (%)	All bulls	34-64	2.32±0.19d(9.2)##	2.08±0.18cd (7.0)	1.54±0.15ab (3.7)	1.79±0.14bc (5.0)	1.35±0.15 (2.9)	1.44±0.14ab (3.2)	1.43±0.14ab (3.2)	1.44±0.14 (3.2)
Distal droplets (%)	All bulls	34-64	1.61±0.18abc (4.0)##	1.75±0.17bc (4.8)	1.85±0.14c (5.4)	1.33±0.13a (2.8)	2.06±0.15c (6.8)	1.99±0.13c (6.3)	1.88±0.13c (5.6)	1.37±0.14ab (2.9)

#### Table 26. Semen and sperm traits (± se) of Brahman (B) and Composite (C) bulls at different ages

Means within traits not followed by a common letter differ significantly (P<0.05); #Log(x) transformed means (back-transformed means); ##Log (x+1) transformed means (back-transformed means)

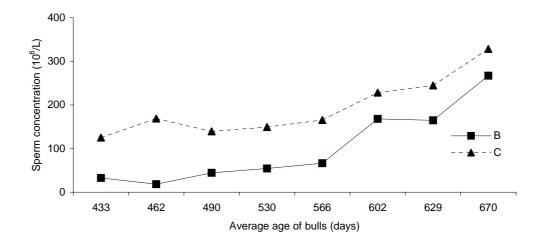
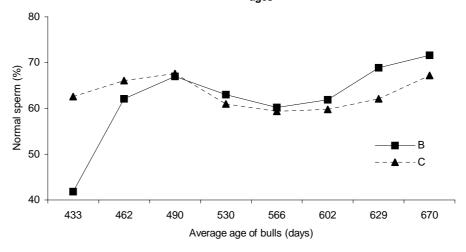


Figure 45. Sperm concentration of Brahman (B) and Composite (C) bulls at different ages

Figure 46. Normal sperm of Brahman (B) and Composite (C) bulls at different ages



#### Abnormal sperm categories

There were no significant breed effects on the various categories of abnormalities (Table 26). All of the categories had significant (P<0.001) fixed effects for date. There was no clear trend for abnormal heads with age whilst there was some evidence of an increase with age for abnormal midpieces. The incidence of proximal droplets was highest at 433 and 462 days of age but from then on it was reasonably constant. With distal droplets, there was no clear pattern with age. The coefficient for birthdate of distal droplets was  $-0.005 \pm 0.002$  indicating that younger bulls had a higher incidence of distal droplets.

# Repeatability of physical, seminal and sperm traits

Repeatability of all traits was low with the exception of sheath score, which was moderate (Table 27).

Trait	Repeatability	Comment
Condition score	$0.23 \pm 0.05$	Low
Testicular tone	$0.21 \pm 0.04$	Low
Sheath score	$0.50 \pm 0.05$	Moderate
Concentration	$0.30 \pm 0.05$	Low
Mass activity	$0.23 \pm 0.005$	Low
Motility	$0.21 \pm 0.05$	Low
Normal sperm	$0.09 \pm 0.04$	Low
Abnormal heads	$0.07 \pm 0.04$	Low
Abnormal midpieces	$0.02 \pm 0.03$	Low
Proximal droplets	$0.04 \pm 0.03$	Low
Distal droplets	$0.01 \pm 0.03$	Low

Table 27. Repeatability of various physical, seminal and sperm traits

# Correlations between physical and seminal traits

The following traits were significantly correlated (P<0.05) in both breeds; mass activity with motility, scrotal circumference, liveweight and age; motility with scrotal circumference; scrotal circumference with liveweight and age; and liveweight with age (Table 28).

In addition the following traits were significantly correlated (P<0.05) in B bulls only; mass activity with testicular tone; motility with liveweight and age; normal sperm with liveweight and age; and testicular tone and liveweight with age and scrotal circumference. The correlations of normal sperm with liveweight and age, although significant were low ( $r = 0.145^*$  and  $r = 0.258^{**}$  respectively) in B bulls.

# Correlations of normal sperm at different ages

Table 29 indicates there is generally little relationship between normal sperm from one age to the next. However Figure 47 indicates that although about 1/3 of the bulls had less than 50% normal sperm at 530 days of age, except for one bull, all were above the 50% normal threshold by 670 days of age.

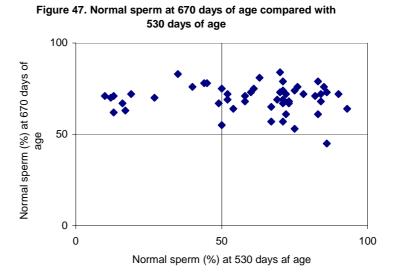
Age	Mass activity	/ Motility	Normal sperm	Scrotal circumf.	Testicular tone	Liveweigh
1.000						
0.441**	1.000					
0.263**	0.520**	1.000				
0.258**	0.046	0.049	1.000			
0.617**	0.485**	0.349**	0.024	1.000		
0.292**	0.176**	0.094	0.055	0.341**	1.000	
0.881**	0.465**	0.284**	0.145*	0.690**	0.247**	1.000
Age	Mass activity	/ Motility	Normal sperm	Scrotal circumf.	Testicular tone	Liveweight
1.000						
0.290**	1.000					
0.089	0.636**	1.000				
-0.050	0.002	0.010	1.000			
0.465**	0.327**	0.195**	-0.064	1.000		
0.121	0.079	-0.003	0.043	0.101	1.000	
0.821**	0.353**	0.139	-0.014	0.679**	0.066	1.000
	1.000 0.441** 0.263** 0.258** 0.617** 0.292** 0.881** Age 1.000 0.290** 0.089 -0.050 0.465**	1.000         0.441**       1.000         0.263**       0.520**         0.258**       0.046         0.617**       0.485**         0.292**       0.176**         0.881**       0.465**         Age       Mass activity         1.000       0.290**         0.089       0.636**         -0.050       0.002         0.465**       0.327**	1.000         0.441**       1.000         0.263**       0.520**       1.000         0.258**       0.046       0.049         0.617**       0.485**       0.349**         0.292**       0.176**       0.094         0.881**       0.465**       0.284**         Age Mass activity Motility         1.000       .290**       1.000         0.290**       1.000       .0089         0.636**       1.000       .010         0.465**       0.327**       0.195**	1.000         0.441**       1.000         0.263**       0.520**       1.000         0.258**       0.046       0.049       1.000         0.617**       0.485**       0.349**       0.024         0.292**       0.176**       0.094       0.055         0.881**       0.465**       0.284**       0.145*         Age Mass activity Motility Normal sperm         1.000       0.290**       1.000         0.299**       1.000       0.089       0.636**         0.089       0.636**       1.000       1.000         0.465**       0.327**       0.195**       -0.064	1.000         circumf.           0.441**         1.000           0.263**         0.520**         1.000           0.258**         0.046         0.049         1.000           0.617**         0.485**         0.349**         0.024         1.000           0.292**         0.176**         0.094         0.055         0.341**           0.881**         0.465**         0.284**         0.145*         0.690**             Age         Mass activity         Motility         Normal sperm         Scrotal circumf.           1.000         .         .         .         .         .           0.089         0.636**         1.000         .         .         .           -0.050         0.002         0.010         1.000         .         .           0.465**         0.327**         0.195**         -0.064         1.000	Line         circumf.         tone           1.000         0.441**         1.000

Table 28. Correlations across all ages (433-670 days of age) for Brahman and Composite bulls

Table 29. Correlations of normal sperm at different average ages of bulls

	462 days	490 days	530 days	566 days	602 days	629 days	670 days
433 days	0.255	0.470**	0.023	-0.139	-0.203	-0.076	0.073
462 days		0.589**	-0.019	-0.174	0.062	0.061	0.091
490 days			0.249	0.026	0.070	-0.002	-0.008
530 days				0.193	0.158	0.127	-0.073
566 days					0.338*	0.247	0.117
602 days						0.169	0.180
629 days							0.247

\*=P<0.05; \*\*=P<0.01



# Proportion of bulls producing at least 50% or 70% normal sperm

Of the bulls tested, more C than B bulls were producing 50+% normal sperm in the period from 433 to 490 days of age, but from then on there was no difference between breeds (Figure 48). At 433 days of age only 12% of B and 59% of C bulls were producing 50+% normal sperm. The proportions for each breed peaked initially in 490 days of age with 100% of C bulls and 75% of B bulls producing 50+% normal sperm, then declined and increased at 629 – 670 days of age. A similar pattern, but lesser proportion occurred with bulls producing 70+% normal sperm where 48% of C bulls and 55% of B bulls had produced 70+% normal sperm by 490-530 days of age.

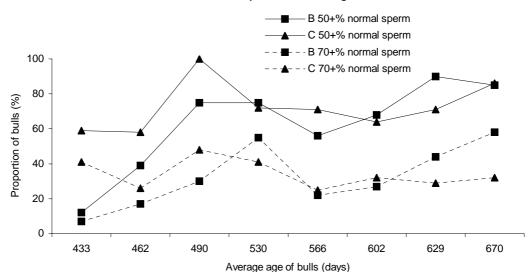


Figure 48. Proportion of Brahman (B) and Composite (C ) bulls producing at least 50% normal or at least 70% normal sperm at different ages

# Estimation of age at puberty

Age at puberty was estimated using 3 different criteria. Firstly if a bull had an ejaculate containing a concentration of at least 50 x  $10^6$  sperm/mL and motility of sperm was at least 10%; secondly bulls had at least a scrotal circumference of 26 cm; and thirdly the first and second criterion combined (Table 30). Using scrotal circumference alone biased the estimation of puberty as 35 bulls had already reached a scrotal size of 26 cm by the date of first recording. There was little difference in estimated age of puberty between methods one and three and this latter procedure combining concentration, motility and scrotal circumference was used for reporting. Regardless of which measurement was used, one-way analysis of variance indicated that C bulls reached puberty significantly (P<0.001) earlier than B bulls (462 v 521 days i.e. 15.4 v 17.4 months) with a range of 180 days for C bulls and 315 days for B bulls (Table 30). There was one B and one C bull that had not reached puberty by the end of the experiment. The correlation between scrotal circumference at puberty and age at puberty was not significant with values for B and C bulls being 0.019 and 0.073 respectively

Criteria used to estimate puberty	Breed	n	Mean (days)	Range (days)	se
Semen concentration and motility <sup>a</sup>	В	41	520	406-721	10.2
	С	28	462	375-555	7.6
Scrotal circumference <sup>b</sup>	В	41	458	379-622	7.9
	С	29	411	364-509	6.3
Semen concentration, motility and scrotal	_				
circumference	В	41	521	406-721	10.3
	С	28	462	375-555	7.6

#### Table 30. Means and range (days) for estimating puberty by 3 methods in Brahman (B) and Composite (C) bulls

<sup>a</sup>One B and one C bull did not reach puberty and not included in analyses

<sup>b</sup>One B bull did not achieve scrotal circumference of 26 cm

# Relationship between age of puberty and subsequent normal sperm

Bulls were divided into 3 puberty classes, early, mid or late. The divisions were < mean  $-\frac{1}{2}$  sd (early), mean  $\pm \frac{1}{2}$  sd (mid) and >mean  $+\frac{1}{2}$  sd (late), where sd was 65 and 40 days for B and C respectively. The effect of puberty class on normal sperm at either 629 or 670 days of age was assessed by one-way analysis of variance. There was no relationship between age at puberty and subsequent normal sperm at either 629 or 670 days of age (Table 31).

# 6.2.2.5 Discussion and conclusions

The data indicate that throughout the study C bulls were more sexually mature than B bulls in some reproductive traits, eg earlier puberty, higher concentration of semen, higher mass activity and motility levels and greater proportions of bulls producing 50+% and 70+% normal sperm at 14 months of age. Higher semen concentration, higher motility and larger scrotal circumferences reflected early puberty. However reaching puberty earlier does not necessarily translate into bulls being more fertile in terms of calf output in multiple sire herds as both scrotal circumference and

semen motility are poorly related to calf output in multiple-sire herds (Holroyd et al. 2002a). As well, from this experiment, early puberty was not related to normal sperm at 670 days (21 months) of age. Normal sperm has been demonstrated as the most important trait in predicting individual calf output of 2- to 4-year old bulls, in multiple-sire herds (Holroyd et al. 2002a).

Breed I	Puberty clas	s Age range of puberty class	629 days			670 days			
В		(days)	n	Normal sperm (%)	se	n	Normal sperm (%)	se	
	Early	<488	15	66.5	2.5	15	72.0	2.0	
	Mid	488-553	13	68.7	2.7	13	71.8	2.1	
	Late	>553	13	70.6	2.7	13	70.0	2.1	
				P=0.53			P=0.75		
С	Early	<442	10	67.6	4.4	10	70.1	2.0	
	Mid	442-482	11	59.7	5.2	11	67.1	2.0	
	Late	>482	7	59.0	4.2	7	65.1	2.4	
				P=0.32			P=0.23		

Table 31 Duberty class and subsequent normal snorm at either 500 or 570 days of each for Brokman (B)

The data indicate that the age of puberty is very variable phenotypically with a range of 6 months for C bulls and 10 months for B bulls. Whilst this data supports the argument that early puberty does not necessarily relate to higher calf output, early puberty in bulls may be heritable and has been demonstrated to be genetically correlated with early puberty in female by some authors (Brinks et al. 1978), but not by others (Perry et al. 1990).

Normal sperm was poorly correlated with other semen and physical traits. This poor relationship highlights the need, as part of a BBSE, for the inclusion of a laboratory morphological assessment of semen rather than relying solely upon a crush side examination of physical and semen traits such as mass activity and motility as indicators of semen quality.

The clear patterns of liveweight gain and increase in size of scrotal circumference, and the moderate repeatability of sheath scores indicate that preliminary selection can be applied to yearling bulls to identify potential herd sires. However the repeatability of normal sperm was poor as was the association between puberty and normal sperm some months later, indicating that a morphological assessment as a yearling will not necessarily reflect their morphology status by the time bulls are This supports the argument for a BBSE examination including a 21-22 months of age. morphological assessment of semen to be conducted close to the commencement of mating.

It was not possible to obtain detailed mating or calf output information on any of these bulls as many were sold after the completion of the experiment. However the level of normal sperm can be used as an indicator of a bull's fertility because of its positive relationship with calf output (Holroyd et al. 2002a). The poor repeatability of sperm morphology does not support our hypothesis that reproductive traits can be identified in yearling bulls that are important for calf output of 2-year-old bulls in multiple-sire herds.

# 6.2.3 Sexual development in Santa Gertrudis bulls - Gyranda

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# 6.2.3.1 Summary

The study was conducted at "Gyranda", Theodore in central Queensland from 23 August 2000 to 21 August 2001 (405 days) using 39 yearling Santa Gertrudis bulls representing 16 sire lines. The mean age of the bulls at the start of the experiment was 9 months (range 190 to 351 days). Bulls were subjected to 10 physical, behavioural and reproductive examinations at 1-3 monthly intervals. Bulls were run as one group on pasture until 27 September 2000 when a subset of 15 bulls (F) was selected by the owner as a show team. These 15 bulls grazed a different paddock and were supplemented with a commercial grain and concentrate pellet mix (CP 14%, ME 11.0 MJ/kg) in troughs twice daily at the rate of 0.5 - 2.0% body weight. The balance of the bulls (NF) grazed pasture only.

There was a linear increase in liveweight with F bulls having a greater increase in weight in the latter stages of the experiment. Weights for NF and F bulls at the start of the experimental period were 277 and 316 kg; at the start of feeding 352 and 369 kg, and final liveweights were 683 and 745 kg respectively. Mean scrotal circumference for all bulls at the start and end of the experiment was 25 and 38 cm respectively. There was a quadratic response with age for an increase in scrotal circumference, but there was no effect of feeding. The average age at which 34 cm was reached was 521 days (range 403 – 732 days).

In serving capacity tests, F bulls achieved higher libido scores than NF bulls in the latter part of the experiment. Of all bulls, 56.4% achieved one serve, with 3 being the maximum number of serves achieved by any bull. The average age, at first service, of the 22 bulls that successfully served was 464 days (range 327 - 647 days).

Sheath depth increased with age with NF bulls having greater depths than F bulls towards the end of the experimental period. Sire means of sheath depths ranged from 95 - 196 mm. Sheath scores varied from 1.4 to 3.7 with no clear pattern with age. Navel thickness scores fluctuated with age and sire means ranged from 1.4 - 3.7. Rosette scores were generally low (mean range 0.4 - 0.8) but increased with age.

Liveweight, scrotal circumference, sheath depth and rosette score were highly repeatable. Navel thickness scores, mounts, mount + serves and libido score were moderately repeatable, whilst all other physical and behavioural traits had low repeatabilities.

Both semen mass activity and motility increased with age and both traits had negative coefficients for date of birth indicating that that these traits increased in older animals. There was no effect of feeding on either mass activity or motility. There were significant effects of age and birth date, but no significant effect of feeding, on normal sperm. Mean values for normal sperm were initially 18.6% at 362 days of age and steadily increased to 62.3% at 636 days of age. The coefficient of birth date was – 0.0993  $\pm$  0.0487 indicating that normal sperm increased with age. There was no effect of feeding on any of the abnormal sperm categories. Age was the only significant effect with abnormal heads decreasing towards the end of the experimental period. Mean values ranged from 27.8 % at 494 days of age to 13.6 % at 636 days of age. Mean values for abnormal midpieces ranged from 8.4 to 13.3 % and were significantly influenced by birth date. Abnormal tails were generally less than 1%. Cytoplasmic droplets were significantly effected by age with values steadily dropping from 38.1% at 362 days of age to 9.0% at 685 days of age. Most semen and sperm traits had low repeatabilities.

There were some significant correlations for normal sperm measured at short intervals (1 to 2 months), but generally there was no relationship for longer periods. Some of the more important significant

correlations between various traits were; scrotal circumference with age and liveweight; mass activity with age, liveweight, testicular tone, scrotal circumference and motility; % normal sperm with age, liveweight, scrotal circumference and mass activity.

In conclusion, this study demonstrated that there was no significant effect of this level of supplementation on scrotal circumference, mating behaviour, semen or sperm traits even though F bulls being 62 kg heavier and 1 unit of body condition better than NF bulls at the end of the feeding period. There were generally some significant correlations for normal sperm measured at 1 to 2 month intervals. However there was no relationship when the time between measurements was longer suggesting that normal sperm values as a yearling does not reflect values at 2 years of age.

# 6.2.3.2 Aims

The aims of the experiment were to measure, from weaning to 2 years of age, physical, behavioural and reproductive traits in Santa Gertrudis bulls; to correlate and measure the repeatability of these traits and to assess the calf getting ability of these bulls when first mated as yearlings.

# 6.2.3.3 Materials and methods

# **Experimental design**

The experiment was an unbalanced longitudinal study from 12 July 2000 until 21 August 2001 using 2 nutritional treatments

(i) NF – non-supplemented

(ii) F – supplemented with commercial bull pellets from 27 September 2000 until the end of the experiment, 21 August 2001.

The primary variables were changes in physical, mating behaviour and reproductive traits and the animal was the experimental unit.

# Experimental site and animals

The experiment was conducted at Gyranda, a 9400 ha Santa Gertrudis stud and commercial beef breeding property located between Theodore and Cracow (25<sup>°</sup> S, 150<sup>°</sup> E) in the brigalow lands of central Queensland. Gyranda lies within the 600-700 mm isohyets and rainfall was below average during the experimental period. The land system is generally grey/black Brigalow soils with stony outcrops.

The study was conducted from 12 July 2000 to 21 August 2001 using 39 Santa Gertrudis bulls representing 16 sire lines. The dates of birth of bulls were from 27 July 1999 to 4 January 2000 (range of 161 days) with mean age of bulls at the commencement of the experiment being 9 months.

# Procedures

Bulls were mustered to the yards on the 12 July 2000, weighed full then subjected to a physical and reproductive examination and semen collected for morphological assessment. Bulls were retained in the yards overnight and the following day they were examined in a serving capacity mating behaviour test. These procedures were then repeated 9 times at 1-3 monthly intervals over the next 12 months.

The bulls were run as one group until 27 September 2000 on pasture and were supplemented with Anipro molasses/urea supplement. At this date a subset of 15 bulls was selected by the owner as part of a show team. These 15 bulls grazed a different paddock and were provided with 'Beef Expandat' a commercial grain and concentrate pellet mix (14% CP; 11.0 MJ ME/kg) in troughs twice daily. This was initially at 0.5 body weight then increased gradually over a monthly period to 2.0% body weight. Table 32 provides a

summary of examination dates, nutritional conditions, and a description of environmental conditions when bulls were examined, including ground surface on which the serving capacity test was conducted.

Bulls were vaccinated against the common clostridial disease prior to the experiment and also vaccinated against bovine ephemeral fever on 27 November 2000

Part of the study was to compare the calf-output of bulls mated as yearlings (11 - 14 months old) with older bulls during the summer of 2000/01. Also the yearling bulls were to be re-mated again as 2-year-olds. Neither of these matings took place due to very dry conditions and reduced breeder numbers.

Date	Nutritional conditions	Weather conditions at examination	Ground surface of cattle yard
12 July 2000	Buffel grass pasture; Anipro / urea supplement mix	2-18 <sup>0</sup> C, cool	Soft soil
23 August 2000	Buffel grass pasture; Anipro / urea supplement mix	16-22°C, overcast	Soft moist soil
27 September 2000	Dry buffel grass, ceased supplement	18-33 <sup>0</sup> C; overcast, light breeze	Soft moist soil
	Commenced 15 bulls on pasture + pellets		
25 October 2000	NF - Irrigated green pastures	18-32 <sup>0</sup> C; RH 75-85%, showery,	Soil moist
	F - 15 bulls on pasture + pellets	rain, cool breeze	
27 November 2000	NF - Dry pastures	31-36°C	Dry and soft
	F - 15 bulls on pasture + pellets		
9 January 2001	NF - Green pastures	22-36 °C; cloudy and overcast,	Soft moist soil
	F - 15 bulls on pasture + pellets	very humid	
6 February 2001	NF - Green pastures	26-32 °C; light breeze, overcast,	Dry and soft
	F - 15 bulls on pasture + pellets	humid	
12 March 2001	NF - Pastures haying off	32-36°C; very humid	Soft moist soil
	F - 15 bulls on pasture + pellets		
26 June 2001	NF - Grass pasture hayed off	18-25 <sup>°</sup> C, overcast, cool	Dry, soft
	F - 15 bulls on pasture + pellets		
21 Aug 2001	NF - Fresh green oats	8-25°C	Dry and soft soil
	F - 15 bulls on pasture + pellets		

 Table 32. Examination date, nutritional and environmental conditions

# **Statistical analysis**

#### Liveweight and scrotal circumference

A random coefficients regression approach was used to analyse liveweight and scrotal circumference from the 10 observation times. This allowed modelling of individual animal patterns over time. The average growth pattern appeared to be approximately linear over the time span considered for the majority of bulls for liveweight and quadratic for scrotal circumference. However, the individual bulls had their own pathway of growth so that there was potentially variation in intercepts and slopes across bulls. This was represented by random intercepts and slopes for individual bulls and correlations between the random intercepts and slopes.

The full model fitted for liveweight was

- Fixed effects: feed + age + feed.age
- Random effects: *bull* + *bull.age* + *dev(age)* + *dev(feed.age)*

### Where

- feed has 2 levels, supplemented and non-supplemented,
- age is mean age at time of measurement,
- *bull* represents the variance component for random intercepts for bulls,
- *bull.age* represents the variance component for random slopes for bulls,
- *dev(age)* is a random lack of fit term for additional fluctuations about the linear trend, and
- *dev(feed.age)* is a random lack of fit term for additional fluctuations about the linear trend for each level of supplementation.

For scrotal circumference the full fitted model was as for liveweight with the addition of the fixed effects,  $age^2 + feed.age^2$ , for the quadratic response.

Random terms with zero variance components were removed from the model, then Wald tests were used to identify non-significant fixed terms (P>0.05) which were also removed to arrive at the final model.

## All other traits

A linear mixed model was used to analyse all other traits recorded across time. The full model fitted was:

- Fixed effects: Feed+birthdate+feed.birthdate+sire+age+feed.age
- Random model: *Bull+bull.age*

## Where

- feed has 2 levels, supplemented and non-supplemented,
- *birthdate* is a covariate the birth date of each bull,
- sire has 16 levels representing the 16 sires of the bulls,
- age has between 6 and 10 levels, 285, 327, 362, 390, 423, 466, 494, 528, 636 and 685 days of age,
- *bull* represents the variance component for individual bulls, and
- *bull.age* represents the residual variance.

In all linear mixed models, random terms with zero variance components were removed from the model, then Wald tests were used to identify non-significant fixed terms (P>0.05) which were also removed to arrive at the final model, except that the main effect of age was always retained. Approximate least significant differences were calculated for significant (P<0.05) fixed effects.

For behavioural traits, the method of analysis across time had a square root (x+0.5) transformation applied to the data prior to analysis. For some of the sperm abnormalities (abnormal tails and cytoplasmic droplets), an angular transformation was applied to the data prior to analysis to stabilise the variance.

Repeatability (intraclass correlation) was estimated as the ratio of variance components:

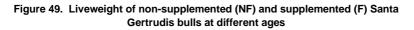
Repeatability = bull/(bull + bull.age).

# 6.2.3.4 Results

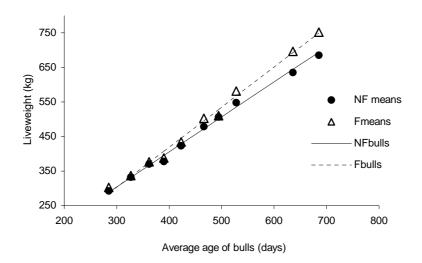
## **Physical traits**

## Liveweight

The pattern of liveweight was linear (Figure 49). F bulls had a greater increase in weight in the latter stages of the experiment. Weights for NF and F bulls at the start of the experimental period were 277 and 316 kg; at the start of feeding, 352 and 369 kg, and final liveweights were 683 and 745 kg respectively.



NF: LWt = 274.36 + 1.010\*age F: LWt = 279.80 + 1.162\*age



## **Condition score**

There was a significant (P=0.007) feed x age interaction (Table 33). F bulls were in better condition at 466, 494, 528 and 685 days of age compared with NF bulls.

Trait	Treatment	285 days	327 days	362 days	390 days	423 days	466 days	494 days	528 days	636 days	685 days	lsd
Condition score (1-9)	NF	3.4a	4.0cd	3.3a	3.5ab	4.5efg	4.7f	4.4ef	4.6ef	4.5efg	5.1h	0.39
	F	3.2a	4.3de	3.3a	3.8bc	4.7f	5.1h	4.8g	4.8g	4.6ef	6.1i	
Testicular tone (1-5)	NF	3.8abc	3.8abc	3.9ab	3.7bcd	3.5de	4.0a	3.8abc	3.8abc	3.5de	3.6cde	0.26
	F	3.6cde	3.8	3.9ab	3.8abc	3.9ab	3.7bcd	3.6cde	3.8abc	3.4e	3.6cde	
Sheath depth (mm)	NF	152a	146a	155a	163a	174a	178a	161a	175a	195a	209b	
	F	145a	143a	142a	155a	165a	166a	166a	175a	183a	188a	
	lsd within columns	17.6										
Sheath score (1-9)	All bulls	4.6def	2.3a	4.3cd	3.5b	3.6b	4.5cde	4.7ef	4.3cd	4.9f	4.1c	0.36
Navel thickness (1-5)	NF	2.9a	1.7a	2.4a	2.0a	1.8a	3.0a	2.9a	2.8a	3.3a	2.6a	
	F	3.0a	1.8a	2.6a	2.4a	2.5b	2.7a	2.8a	3.1a	3.4a	2.9a	
	lsd within columns	0.53										
Rosette score (0-3)	All bulls	0.7bc	0.4a	0.5ab	0.5ab	0.8c	0.8c	0.7c	0.8c	1.5e	1.1d	0.25
Interest (n)	All bulls	2.1a (3.8) <sup>#</sup>	2.4b (5.4)	2.5bc (5.6)	2.2ab (4.5)	2.0a (3.7)	2.3ab (4.6)	2.4b (5.3)	2.3ab (4.7)	2.2ab (4.3)	2.8c (7.1)	0.31
Mounts (n)	All bulls	2.5d (5.9) <sup>#</sup>	2.3cd (4.9)	2.5d (5.8)	2.0abc (3.4)	1.8a (2.6)	2.1abc (3.8)	2.1bc (4.1)	2.3cd (4.8)	2.2cd (4.4)	1.8ab (2.7)	0.36
Mounts+serves (n)	All bulls	6.5c	5.7bc	6.7c	4.0ab	2.9a	4.5ab	5.1bc	5.7bc	5.1bc	3.2a	1.75
Libido score (0-10)	NF	5.0a	4.6a	5.0a	4.5a	3.8a	4.5a	4.9a	5.0a	4.8a	4.5a	
	F	4.2a	4.3a	4.9a	4.5a	4.8a	5.4a	6.5a	5.1a	6.4b	6.2b	
	Isd within columns	1.56										

#### Table 33. Physical and behavioural traits of Santa Gertrudis at different ages

For each trait, means within rows or columns not followed by a common letter differ significantly (P<0.005) # Back-transformed means

### Testicular tone

There was a significant (P=0.031) feed x age interaction (Table 33), this interaction being due to inconsistent differences between NF and F bulls. There was limited variability with most values in the range of 3 to 4.

#### Scrotal circumference

There was a quadratic response with age, with no effect of feeding. Figure 50 shows the predicted smooth model for scrotal circumference. Mean scrotal circumference at the start and end of the experiment was 25 cm and 38 cm respectively. Of the 39 bulls, 37 reached a scrotal circumference of at least 34 cm, the minimum recommended by the AACV at 24 months (Entwistle and Fordyce 2003). The average age at which 34 cm was reached was 521 days (range of 403 – 732 days).

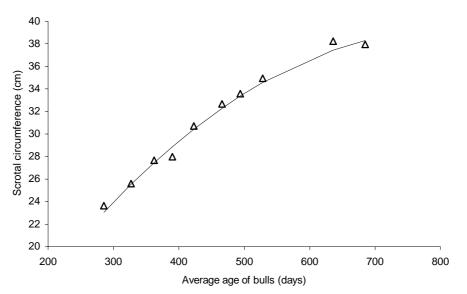


Figure 50. Scrotal circumference of Santa Gertrudis bulls at different ages  $SC = 23.001 + 0.06264^*age - 0.00006203^*age^2$ 

#### Sheath depth

There were significant (P<0.001) effects of sire and a significant (P<0.001) feed x age interaction (Table 33). There was a trend for an increase with age with NF bulls having greater sheath depths towards the end of the experimental period than F bulls (Figure 51). Sire means ranged from 95 - 196 mm (Table 34)

#### Sheath score

There were significant effects of sire (P=0.008) and age (P<0.001) with no clear pattern in sheath score with age. Sire means ranged from 3.3 to 5.6 (Table 34).

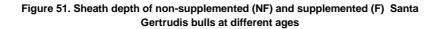
#### Navel thickness score

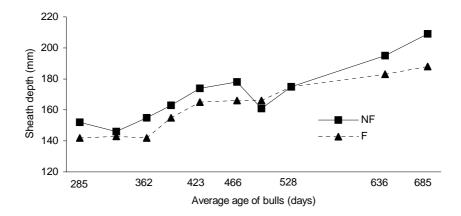
There were (P<0.001) significant effects of sire with a significant (P=0.014) feed x age interaction. Scores fluctuated with age with F bulls having significantly larger scores than NF bulls at 423 days of age. Sire means ranged from 1.4 - 3.7 (Table 34).

Sire ID	Sheath depth (mm) ± se	Sheath score (1-9) ± se	Navel thickness score (1-5) ± se
160633	171 bcde ± 10	4.2 abcd ± 0.3	2.6 cde ± 0.2
166495	188 de ± 14	3.7 ab ± 0.4	$2.3 \text{ abcd} \pm 0.3$
166904	175 cde ± 11	4.0 abc ± 0.3	2.8 de ± 0.3
168714	180 de ± 12	3.7 ab ± 0.3	2.9 de ± 0.3
174938	193 de ± 20	$4.0 \text{ abc} \pm 0.5$	$2.2 \text{ abcd } \pm 0.5$
174959	160 bcde ± 20	4.1 abcd ± 0.6	3.7 e ± 0.5
177964	173 bcde ± 14	3.9 abc ± 0.4	2.5 bcde ± 0.3
184650	143 abc ± 11	$4.2 \text{ abcd} \pm 0.3$	1.8 abc ± 0.3
184657	180 de ± 10	3.6 ab ± 0.3	3.2 de ± 0.2
189701	95 a ± 20	5.6 d ± 0.5	2.8 cde ± 0.5
192968	171 bcde ± 20	3.5 ab ± 0.6	3.3 de ± 0.5
197621	196 e ± 10	3.9 ab ± 0.3	1.7 ab ± 0.2
199854	134 ab ± 14	5.0 cd ± 0.4	1.4 a ± 0.3
199879	148 abcd ± 20	$5.0 \text{ bcd} \pm 0.5$	3.0 de ± 0.5
201600	175 cde ± 11	3.3 a ± 0.3	2.8 de ± 0.3
291682	188 de ± 10	3.5 a ± 0.3	3.0 de ± 0.2

Table 34. Sire effects on sheath depth, sheath score and navel thickness score

Means within columns not followed by a letter in common differ significantly (P<0.05)





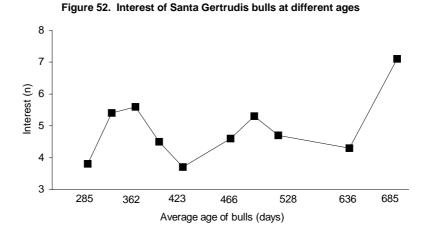
#### Rosette score

Age was the only significant (P<0.001) effect. Mean scores were generally low (range 0.4 - 0.8) but increased with age.

## Mating behaviour traits

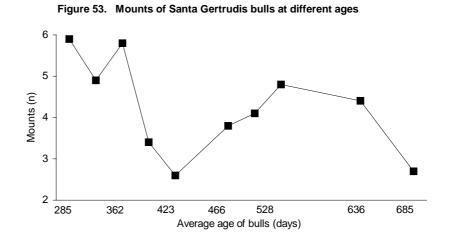
#### Interest

There was a significant (P<0.001) effect of age (Table 33) but no effect of feeding. There was substantial variation among bulls for interest responses with no clear pattern with age (Figure 52).



#### Mounts

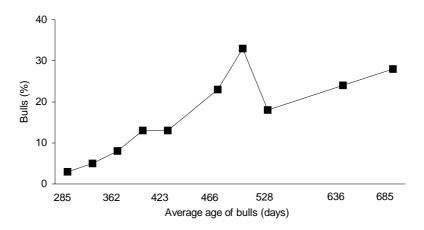
The only significant (P<0.001) effect was age (Table 33) with no clear pattern although there were less mounts at the end of the experimental period than at the beginning (Figure 53).



#### Serves

56.4% of bulls achieved a serve with 3 being the maximum number achieved by any bull. The average age (at the time of their first serve) of the 22 bulls that did successfully serve was 464 days with a range from 327 to 647 days. Conversely, the average age (at their last serving capacity test) of the 17 bulls that did not serve was 684 days with a range of 595 to 741 days. Percentage of bulls with one or more serves is presented in Figure 54.

Figure 54. Percentage of Santa Gertrudis bulls achieving 1 or more serves in a serving capacity test at different ages



### Mounts+serves

There was only a significant (P<0.001) effect of age with a general trend for mounts+serves to decrease with age (Table 33). Mounts+serves ranged from 0 to 22.

## Libido score

There was a significant (P=0.032) effect of age with a significant (P=0.021) feed x age interaction (Table 33). F bulls had higher libido scores than NF bulls at 636 and 685 days of age.

## Repeatability of physical and behavioural traits

Liveweight, scrotal circumference, sheath depth and rosette score were highly repeatable (Table 35). Navel thickness scores, mounts, mount+serves and libido score were moderately repeatable whilst all other traits were low.

Trait	Repeatability	Comment	
Liveweight	$0.73 \pm 0.05$	High	
Condition score	0.16 ± 0.05	Low	
Testicular tone	$0.10 \pm 0.04$	Low	
Scrotal circumference	$0.73 \pm 0.05$	High	
Sheath depth	$0.70 \pm 0.07$	High	
Sheath score	$0.28 \pm 0.08$	Low	
Navel thickness score	$0.39 \pm 0.09$	Moderate	
Rosette score	$0.66 \pm 0.06$	High	
Interest	$0.27 \pm 0.06$	Low	
Mounts	$0.38 \pm 0.07$	Moderate	
Mounts+serves	$0.36 \pm 0.07$	Moderate	
Libido score	$0.42 \pm 0.07$	Moderate	

 Table 35.
 Repeatability of physical and behavioural traits of Santa Gertrudis bulls

# Correlations between sheath traits at different ages

There were strong relationships between ages for sheath depth and to a lesser extent sheath score, navel thickness score and rosette score. However, as these latter 3 traits are scores with a limited range of values, only the data for sheath depth is presented in Table 36.

Age (days)	285	327	362	390	423	466	494	528	636	685
285	1.000									
327	0.878**	1.000								
362	0.852**	0.828**	1.000							
390	0.864**	0.884**	0.863**	1.000						
123	0.834**	0.827**	0.822**	0.913**	1.000					
466	0.784**	0.760**	0.795**	0.847**	0.915**	1.000				
194	0.719**	0.774**	0.700**	0.820**	0.848**	0.881**	1.000			
528	0.689**	0.742**	0.689**	0.808**	0.853**	0.881**	0.904**	1.000		
636	0.660**	0.692**	0.683**	0.787**	0.826**	0.837**	0.846**	0.883**	1.000	
685	0.739**	0.700**	0.812**	0.797**	0.836**	0.842**	0.747**	0.841**	0.847**	1.000
	** P<0.01	l								

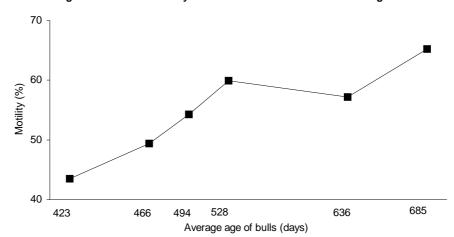
# Semen and sperm traits

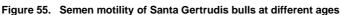
### Mass activity

There were significant effects of age (P<0.001) and date of birth (P = 0.004). Mass activity steadily increased with age with mean values at 327 days of age being 0.3 and at 685 days of age, 2.1 (Table 37). The coefficient for date of birth was  $-0.007 \pm 0.002$  indicating that mass activity increased with age.

## Motility

There were significant effects of age (P<0.001) and date of birth (P = 0.017). Mean motility increased from 43.5 % at 423 days of age to 65.2% at 685 days of age (Table 37, Figure 55). The coefficient for date of birth was -0.154  $\pm$  0.065 indicating that motility increased with age.





Trait	327 days	362 days	390 days	423 days	466 days	494 days	528 days	636 days	685 days
Mass activity (0-5)	0.3±0.2a	0.7±0.2ab	0.8±0.2b	1.0±0.2b	1.4±0.2c	1.5±0.2c	2.0±0.2d	2.0±0.2d	2.1±0.2d
Motility (%)	id^	id	id	43.5±4.4a	49.4±4.1ab	54.3±4.0bc	59.9±4.1cd	57.2±4.1bcd	65.2±4.4d
Normal sperm (%)	id	18.6±6.4a	25.1±4.2a	26.3±3.9a	29.0±3.3ab	36.3±3.2b	44.1±3.1c	62.3±3.1d	58.7±3.2d
Abnormal heads (%)	id	19.1±4.5ab	26.7±2.9bcd	21.5±2.6bc	25.9±2.2bcd	27.8±2.1d	26.5±2.1cd	13.6±2.1a	20.7±2.2b
Abnormal midpieces (%)	id	21.4±2.8 (13.3) <sup>#</sup>	20.7±12.5 (12.5)	20.6±12.4 (12.4)	17.4±1.5 (8.9)	17.5±1.4 (9.0)	17.0±1.4 (8.6)	18.6±1.4 (10.2)	16.8±1.4 (8.4)
Abnormal tails (%)	id	15.9±1.9c (7.5) <sup>#</sup>	1.5±1.2a (0.1)	2.7±1.1ab (0.2)	3.7±0.9ab (0.4)	2.8±0.9ab (0.2)	2.7±0.9ab (0.2)	1.9±0.9a (0.1)	4.9±0.9b (0.7)
Cytoplasmic droplets (%)	id	38.1±4.4e (38.1) <sup>#</sup>	33.7±3.0de (30.7)	36.9±2.7e (36.1)	34.2±2.3e (31.7)	27.2±2.2cd (20.9)	23.4±2.2bc (15.8)	18.9±2.2ab (10.5)	17.4±2.2a (9.0)

Table 37. Semen and sperm traits of Santa Gertrudis bulls at different ages

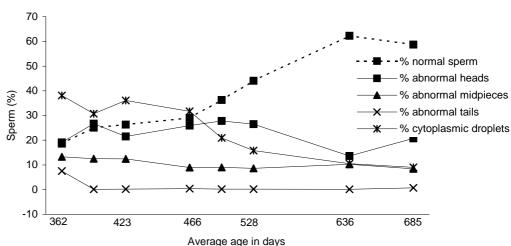
Means within rows not followed by a common letter differ significantly (P<0.005) ^Insufficient data; # Back-transformed means

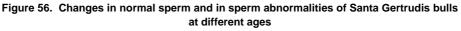
## Normal sperm

There were significant effects of age (P<0.001) and date of birth (P = 0.042) but no significant effect of feeding on normal sperm. Mean values were initially 18.6% at 362 days of age, with a steady increase with age. Then values plateaued at 636 and 685 days at 62.3% and 58.7% respectively (Table 37, Figure 56). The coefficient of birth date was  $-0.0993 \pm 0.0487$  indicating that normal sperm increased with age.

### Abnormal sperm

There was no effect of feeding on any of the abnormal sperm categories (Table 37, Figure 56). With abnormal heads, age was the only significant (P<0.001) factor, with abnormal heads low at the start, increasing and then decreasing towards the end of the experimental period. Mean values ranged from 13.6% at 636 days of age to 27.8% at 494 days of age. With abnormal midpieces, only date of birth was significant (P = 0.028) with the coefficient for date of birth being 0.0492  $\pm$  0.0.224. Overall mean values for abnormal midpieces ranged from 8.4 to 13.3%. There was a significant (P<0.001) effect of age on abnormal tails with means less than 1 except for 362 days of age when there were 7.5% abnormal tails. For cytoplasmic droplets, there was a significant (P<0.001) effect of age with values steadily dropping from 38.1% at 362 days of age to 9.0% at 685 days of age.





## Repeatability of semen and sperm traits

Mass activity was moderately repeatable with all other traits being lowly repeatable (Table 38).

## Correlations between ages for sperm morphology traits

These correlations indicate that there is generally some relationship for normal sperm when measured within 1 to 2 months but generally there was no relationship when the time between measurements was longer (Table 39). Correlations for various abnormality categories, not presented as tables, followed similar patterns to that of normal sperm, except for cytoplasmic droplets where there were significant correlations between consecutive monthly measurements from 390 to 494 days of age, (over 4 recordings) but from then on there was no significance in the correlations.

Trait	Repeatability	Comment
Mass activity	$0.34 \pm 0.07$	Moderate
Motility	$0.30 \pm 0.08$	Low
Normal sperm	$0.29 \pm 0.08$	Low
Abnormal heads	0.17 ± 0.07	Low
Abnormal midpieces	$0.33 \pm 0.08$	Low
Abnormal tails	$0.05 \pm 0.06$	Low
Cytoplasmic droplets	$0.29 \pm 0.08$	Low

Table 38. Repeatability of semen and sperm traits of Santa Gertrudis bulls

Table 39. Correlations between ages for normal sperm of Santa Gertrudis bulls

Age (days)	362	390	423	466	494	528	636	686
362	1.000							
390	0.711**	1.000						
423	0.537**	0.589**	1.000					
466	0.238	0.374*	0.503**	1.000				
494	0.211	0.334*	0.584**	0.437**	1.000			
528	0.176	0.433**	0.407*	0.216	0.695**	1.000		
636	0.115	0.182	0.022	0.335*	0.127	0.32	1.000	
686	0.067	0.192	0.386*	0.131	0.586**	0.455	0.009	1.000

\* P<0.05; \*\*P<0.01

# Correlations between physical, behavioural and seminal traits

The following traits were significantly (P<0.05) positively correlated at almost all ages:

- liveweight with age and scrotal circumference,
- mass activity with motility, and
- normal sperm with mass activity and motility.

The following traits were significantly (P<0.05) correlated at the early ages of measurement but not as the bulls aged:

- age with scrotal circumference,
- age with mass activity,
- age with normal sperm,
- age with motility,
- liveweight with mass activity,

- scrotal circumference with mass activity,
- scrotal circumference with normal sperm,
- scrotal circumference with motility, and
- interest with mounts.

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Correlations at 390 days of age are given in Table 40 as an example.

••

	Age	Liveweight	Scrotal circum.	Testicular tone	Interest	Mounts	Serves	Mass activity	Normal sperm	Motility
Age	1.000									
Liveweight	0.828**	1.000								
Scrotal circum.	0.578**	0.683**	1.000							
Testicular tone	-0.046	0.012	0.299	1.000						
Interest	0.249	0.093	0.052	-0.332*	1.000					
Mounts	0.062	0.002	0.067	-0.039	0.310	1.000				
Serves	0.165	-0.032	0.035	-0.134	-0.037	0.346*	1.000			
Mass activity	0.419**	0.415**	0.649**	0.259	-0.069	0.015	0.166			
Normal sperm	0.563**	0.567**	0.505**	-0.047	0.060	-0.043	0.078	0.617**		
Motility	0.426*	0.411*	0.511**	-0.188	0.040	-0.044	0.099	0.569**	0.676**	1.000

.. . ..

\*P<0.05; \*\* P<0.01

The correlations between traits bulked across all ages are presented in Table 41. There were many significant (P<0.05) correlations although many correlations were numerically small. Some of the more important and consistent significant correlations were:

- Scrotal circumference with age and liveweight .
- Mass activity with age, liveweight, testicular tone, scrotal circumference and motility
- Normal sperm with age, liveweight, scrotal circumference and mass activity.

# 6.2.3.5 Discussion

This study has demonstrated that, at a supplementary feed intake of 0.5-2.0% of body weight, which is equivalent to 1.5-14.0 kg of commercial pellets, there was no significant effect of feeding on scrotal circumference, semen or sperm traits despite F bulls being 62 kg heavier and 1 unit of body condition better than NF bulls. However F bulls did have higher libido scores than NF bulls towards the end of the feeding period.

The liveweight performance of the bulls was remarkable considering the drought conditions. NF bulls and F bulls had gains of 1.0 and 1.14 kg/day throughout the experimental period, which demonstrates both the genetic potential for growth in these bulls, plus the inefficiencies of feeding.

	Age	Liveweight	Scrotal circum.	Testicular tone	Interest	Mounts	Serves	Mass activity	Normal sperm	Motility
Age	1.000									
Liveweight	0.962**	1.000								
Scrotal circum.	0.858**	0.881**	1.000							
Testicular tone	-0.178**	-0.194**	-0.104*	1.000						
Interest	0.087	0.081	0.081	0.013	1.000					
Mounts	-0.122*	-0.103*	-0.096	0.076	0.221**	1.000				
Serves	0.226**	0.182**	0.177**	-0.060	0.003	0.134**	1.000			
Mass activity	0.545**	0.559**	0.636**	0.067	0.015	0.014	0.093			
Normal sperm	0.694**	0.670**	0.657**	-0.085	0.065	-0.024	0.168**	0.576**		
Motility	0.271**	0.284**	0.350**	0.058	0.026	0.162*	0.013	0.628**	0.461**	1.000

#### Table 41. Correlations between traits of Santa Gertrudis bulls across all ages

\*P<0.05; \*\* P<0.01

Serving values were low with only 22 of the 39 bulls having one or more serves in libido tests prior to potential mating as 2-year-old bulls. The sequential testing provided the opportunity for sexual experience for these young bulls, but did not appear to benefit some bulls. This poor performance by some bulls may be indicative of delayed mating activity when first placed in a breeder herd.

As expected there was a steady increase in the quality of semen assessed crush-side as measured by mass activity and motility. This increase in semen quality was mirrored in a corresponding increase in normal sperm which is an important fertility trait affecting calf-output.

The repeatability of behavioural, semen and sperm traits was either low or moderate indicating the difficulty of using these measurements, in yearling bulls, as a prediction of future values of these traits. Whilst there were some significant correlations between morphological traits at 1 to 2 month intervals, these were more pronounced early in life (between 11-14 months of age) rather than later in life (16-21 months of age). Generally there was no relationship over longer periods between normal sperm suggesting that a morphological assessment of a bull as a yearling is no indication of its morphological status as a 2-year-old.

Most semen and sperm traits were moderately correlated but there were few consistent relationships between semen and sperm traits with physical and mating behaviour traits. As expected there were strong relationships between age and liveweight and liveweight and scrotal circumference. Likewise older animals tended to have more serves and better semen quality, as measured by motility, mass activity and normal sperm.

Because of drought conditions we could not mate these yearling bulls and measure their calf output. Almost 95% of bulls reached the AACV minimum scrotal circumference standards by 2-year-old. There was a large range in age (13.4 - 24 months); mean 17 months) to meet the scrotal circumference of 34 cm, which is the minimum standard recommended by the AACV for bulls 24 months of age. The average age at first serve was 15.4 months and the average age that bulls' semen had 50% normal sperm was 17 months (April 2001). The values of these traits would suggest that some of these bulls would be suitable to use as herd sires as yearlings although their calf-output may not be as great as older bulls.

# 6.2.4 Sexual development in Droughtmaster bulls - Dilga

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# 6.2.4.1 Summary

The study was conducted using the same group of bulls at several locations in southern Queensland. Forty Droughtmaster bulls were subjected to a number of physical and behavioural reproductive examinations from 25 September 2000 (mean age 366 days) to 14 August 2001 (mean age 689 days).

Liveweight change was linear where age was the only significant (P<0.001) effect. Mean initial and final liveweights and liveweight gain were 383, 662 and 279 kg respectively. There was a linear increase in scrotal circumference with mean initial and final scrotal circumference being 29.7 and 38.2 cm respectively. The average age at which 34 cm was reached was 535 days (range of 320-689 days of age). There was no clear pattern of change with age for sheath depth, sheath score, navel thickness score and rosette score. There was a small increase with age in number of interest responses and little change in number of mounts with only 12 of the 40 bulls serving in a libido test. Their average age at their first serve of these 12 bulls was 521 days with a range from 346 to 707 days.

There was a general increase in mass activity, but not motility, of semen with age. Normal sperm was low initially (mean of 37.3% at 411 days of age), and then declined further prior to steadily increasing to 58% at 653 to 689 days of age. Abnormal heads ranged from 38.5% at 467 days to 14.9% at 653 days of age. Mean abnormal midpieces were 15.6%. Abnormal tails were low overall with values ranging from 0.3 to 4.5%. Cytoplasmic droplets decreased with age.

Liveweight and scrotal circumference were highly repeatable, navel thickness score and sheath score were moderately repeatable whilst condition score and testicular tone were lowly repeatable. The behavioural traits of interest and mounts were moderately repeatable with libido score being highly repeatable. Semen and sperm traits were lowly repeatable.

Sheath depth, navel thickness score and rosette score, in most cases, were strongly correlated between observations whilst testicular tone was poorly correlated between observations. There was little evidence of any significant relationships between observations for normal sperm except towards the end of the experimental period. There were many significant but numerically small value correlations between physical, behavioural and reproductive traits. Some of the more statistically significant correlations were mass activity with age, liveweight, motility and scrotal circumference; normal sperm with age, liveweight and scrotal circumference with age and liveweight; age with weight; interest with mounts and mounts with serves.

The data suggests that sheath depth, sheath score, navel thickness score and rosette scores recorded when bulls are yearlings (about 12-14 months) give a good indication of their relative size at 2 years of age. However, there was little evidence of any significant relationship between ages for normal sperm.

# 6.2.4.2 Aims

The aims of the experiment were to measure, from 12 months to 24 months of age, physical, behavioural and reproductive traits in Droughtmaster bulls and to correlate and measure the repeatability of these traits.

# 6.2.4.3 Materials and methods

# **Experimental design**

The experiment was an unbalanced longitudinal study of 40 Droughtmaster bulls that were sired by 4

groups of sires; 15 by Sire A (Bull BLB), 6 by Sire B (Bull 118/6) and 8 by Sire C (Bull GIX). A further 11 bulls were from multiple-sire matings and could not be identified to specific sires.

# Experimental sites and animals

The study was conducted from 25 September 2000 to 14 August 2001. Birth dates of bulls ranged from 7 September to 10 November 1999, a range of 64 days. Mean age of bulls at the commencement of the experiment was 12 months. Because of drought conditions, the bulls were moved to several locations after the commencement of the experiment at Dilga. Bulls had been vaccinated against the common clostridial diseases prior to the experiment and were vaccinated against tick fever in March 2001. Table 42 gives a brief description of location, environmental and nutritional conditions at and around examination dates.

Table 42. Examination date,	location and nutritional	conditions of bulls
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	La sa Cara	<b>T</b>	No. (20 construction of the second
Date	Location	Temperature at testing	Nutritional conditions
25 September 2000	Dilga, 28 <sup>o</sup> S, 149 <sup>o</sup> E. Glenmorgan	20-35 <sup>0</sup> C, light breeze	Supplemented with Anipro and cottonseed until relocation
9 November 2000	Aronui feedlot; Dalby	18-22 <sup>0</sup> C, overcast	Arrived 6 October 2000 until 23 February 2001 (140 days); fed a low gain silage ration (7.0%CP, 8MjME/Kg DM)
4 January 2001	Aronui feedlot; Dalby	32-38 <sup>0</sup> C	Low gain silage ration
25 April 2001	Dilga; Glenmorgan	22-28 <sup>0</sup> C	From 23 February 2001 to 15 June 2001, bulls grazed buffel and blue grass pastures.
9 July 2001	Talabanger (20 km south of Dilga)	12-18 <sup>0</sup> C	From 15 June 2001 to 31 July 2001, grazed a forage oat crop (app 10-12%CP, 8-10MJME/Kg DM)
14 August 2001	Dilga; Glenmorgan	18-20 <sup>0</sup> C	In early August 2001, bulls walked approximately 20km to Dilga and grazed buffel grass pastures

We intended following some of the bulls through two consecutive matings as multiple-sires and relating physical and reproductive traits to calf output. However drought prevented these matings.

# Procedures

Bulls were run as one group on each of the properties. At each examination, bulls were mustered to the yards, weighed full then subjected to a physical and reproductive examination and semen collected for morphological assessment. Bulls were then serving capacity tested on the following day. These procedures were performed 6 times from 25 September 2000 to 14 August 2001, the exception being that serving capacity tests could not be conducted at Aronui feedlot on 9 November 2000.

# Statistical analysis

## Liveweight and scrotal circumference

A random coefficients regression approach was used to analyse liveweight and scrotal circumference from the 6 times. This allowed modelling of individual animal patterns over time. The average growth pattern appeared to be approximately linear over the time span considered for the majority of bulls for both liveweight and scrotal circumference. However, the individual bulls had their own pathway of growth so that there was potentially variation in intercepts and slopes across bulls. This was represented by random intercepts and slopes for individual bulls and correlations between the random intercepts and slopes.

The full model fitted for liveweight and scrotal circumference was

- Fixed effects: age
- Random effects: *bull* + *bull.age* + *dev(age)*

Where

- age is the average age of bulls at measurement,
- bull represents the variance component for random intercepts for bulls,
- bull.age represents the variance component for random slopes for bulls, and
- *dev(age)* is a random lack of fit term for additional fluctuations about the linear trend.

Random terms with zero variance components were removed from the model, then Wald tests were used to identify non-significant fixed terms (P>0.05) which were also removed to arrive at the final model.

## All other traits

A linear mixed model was used to analyse all traits recorded at different ages except scrotal circumference and liveweight. The full model fitted was:

- Fixed effects: *birthdate+sire+age*
- Random model: *bull+bull.age*

### Where

- *birthdate* is a covariate the birth date of each bull,
- *sire* is the sire of the bull,
- age has between 4 and 6 levels; 366, 411, 467, 578, 653 and 689 days of age, and
- bull.age represents the residual variance.

In all linear mixed models, random terms with zero variance components were removed from the model, then Wald tests were used to identify non-significant fixed terms (P>0.05) which were also removed to arrive at the final model, except that the main effect of age was always retained. Approximate least significant differences were calculated for significant (P<0.05) fixed effects. For the majority of semen traits, due to the imbalance in number of bulls across measurement times, it was not possible to quote LSDs, thus SEs were included.

Repeatability (intraclass correlation) was estimated as the ratio of variance components:

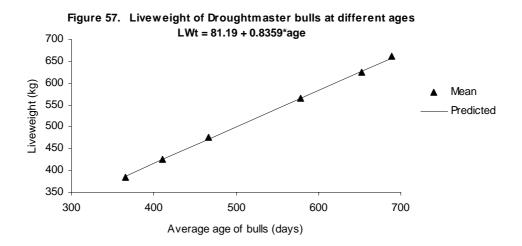
Repeatability = bull/(bull + bull.age).

# 6.2.4.4 Results

# Physical traits

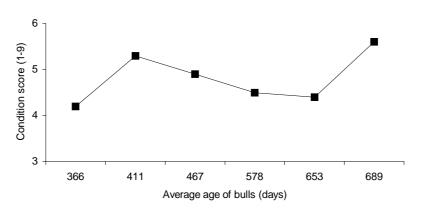
## Liveweight

The pattern of liveweight was linear (Figure 57) where age was the only significant (P<0.001) fixed effect. Mean initial and final liveweights and gain over the experimental period were 383, 662 and 279 kg respectively. This represented an average daily gain of 0.86 kg/day throughout the study period.



## **Condition score**

Age was the only significant (P<0.001) effect (Table 43). Condition scores initially increased to 5.3 at 411 days of age (while in the feedlot), then steadily declined to 4.4 in 653 days of age, then increased dramatically in 689 days of age when bulls had been grazing oats for the previous 80 days (Figure 58).





#### Testicular tone

There was little variation in testicular tone between bulls and ages with most values being 3 and 4.

#### Scrotal circumference

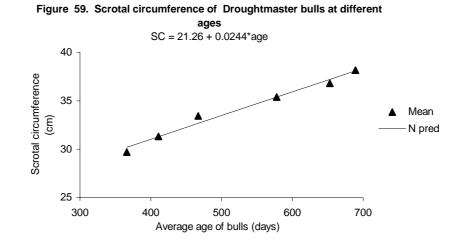
There was a significant (P<0.001) linear response with age. Figure 59 presents the predicted smooth model for scrotal circumference. Mean initial and final scrotal circumference were 29.7 and 38.2 cm respectively. Of the 40 bulls, 39 reached a scrotal circumference of at least 34cm, the minimum recommended by the AACV at 24 months (Entwistle and Fordyce 2003). The average age at which 34 cm was reached was 535 days (range of 320- 689 days).

Traits	366 days	411 days	467 days	578 days	653 days	689 days	lsd
Condition score (1-9)	4.2a	5.3d	4.9c	4.5b	4.4ab	5.6e	0.25
Sheath depth (mm)	177.5a	194.1b	200.1b	196.7b	185.2a	191.1b	8.21
Sheath score (1-9)	3.8b	2.5a	4.0bc	4.3c	4.3c	3.9bc	0.39
Navel thickness (1-5)	1.9bc	1.6a	2.1cd	1.7ab	2.3d	2.0bc	0.29
Rosette score (0-3)	1.0a	1.0a	0.9a	0.8a	1.3b	1.0ab	0.25
Interest (n)	1.6ab (2.0) <sup>#</sup>	nd <sup>##</sup>	1.5a (1.8)	1.7ab (2.3)	1.8bc (2.9)	2.0c (3.3)	0.28
Mounts (n)	1.5a (1.6) <sup>#</sup>	nd	1.2a (1.0)	1.3a (1.2)	1.2a (1.0)	1.2a (1.0)	
Mounts + serves (n)	2.9a	nd	2.0a	2.2a	1.8a	1.9a	
Libido score (0-10)	3.0a	nd	2.7a	2.7a	3.3a	3.1a	

Table 43. Physical and mating behaviour traits of Droughtmaster bulls at different ages

Means within rows not followed by a common letter differ significantly (P<0.05) ;# back-transformed mean;

##nd, no data



#### Sheath depth

The only significant (P<0.001) effect was age (Table 43). There was considerable variability between bulls with sheath depth initially increasing, peaking at 467 days of age, then decreasing until 653 days of age when grazing the oats, before increasing again at 689 days of age (Figure 60).

## Sheath score

The only significant (P<0.001) effect was age (Table 43) with scores declining in November 2000 before returning to original values (Figure 61).

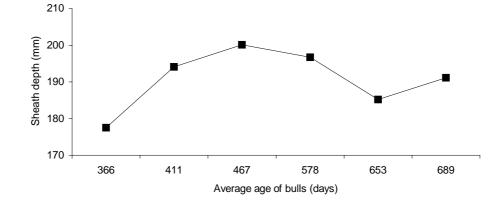
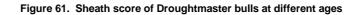
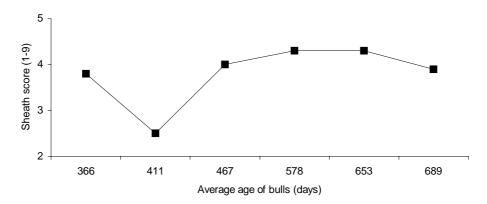


Figure 60. Sheath depth of Droughtmaster bulls at different ages





# Navel thickness score

The only significant (P<0.001) effect was age (Table 43) with no clear pattern of change (Figure 62).

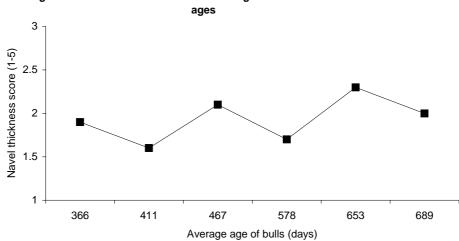


Figure 62. Navel thickness score of Droughmaster bulls at different

## Rosette score

The only significant (P=0.011) effect was age (Table 43) with no clear pattern of change.

## Mating behaviour traits

## Interest

The only significant (P=0.012) effect was age (Table 43) with a small increase in number of interest responses over the experimental period.

## Mounts

There were no significant effects with mean number of mounts ranging from 1.2 to 1.5.

## Serves

Only 12 bulls ever successfully served. The average age (at the time of their first serve) of these 12 bulls was 521 days with a range from 346 to 707 days. Of those bulls that did achieve at least one serve, the total number of serves over the 5 measurement times ranged from 1 to 6 serves with a mean of 2.8.

## Mounts and serves

There was no significant effect of age with mounts+serves with means of 1.9 to 2.9 and a range of 0 - 20.

## Libido score

There was no change with age with means of 2.7 to 3.1.

# Repeatability of physical and behavioural traits

Physical traits such as liveweight, scrotal circumference and rosette score had high repeatabilities (Table 44). Sheath depth, sheath score and navel thickness score were moderately repeatable whilst condition score and testicular tone were lowly repeatable. All mating behaviour traits (interest, mounts, mounts+serves and libido) were moderately repeatable.

Trait	Repeatability	Comment
Liveweight	$0.89\pm0.02$	High
Condition score	$0.17\pm0.07$	Low
Scrotal circumference	$0.67\pm0.06$	High
Testicular tone	$0.10\pm0.06$	Low
Sheath depth	$0.61\pm0.07$	Moderate
Sheath score	$0.41\pm0.08$	Moderate
Navel thickness score	$0.58\pm0.07$	Moderate
Rosette score	$0.73\pm0.05$	High
Interest	$0.59\pm0.07$	Moderate
Mounts	$0.54\pm0.08$	Moderate
Mounts+serves	$0.51 \pm 0.08$	Moderate
Libido score	0.62 ± 0.07	Moderate

### Correlations between times for physical traits

Testicular tone was poorly correlated between times (Table 45). With sheath depth there were consistently significant (P<0.001) relationships between times (r = 0.429 - 0.770). With sheath scores, the relationships were similar, however r values were less in all cases and not all were significant. In most cases there were strong significant relationships between times for both navel thickness score and rosette score.

Age in days	366	411	467	578	653	689
Testicular tone						
366	1.000					
411	0.155	1.000				
467	-0.141	-0.052	1.000			
578	-0.141	0.253	-0.239	1.000		
653	-0.122	0.153	-0.250	-0.128	1.000	
689	-0.370*	-0.217	-0.334*	0.006	0.034	1.000
Sheath depth						
366	1.000					
411	0.487**	1.000				
467	0.634**	0.665**	1.000			
578	0.596**	0.575**	0.758**	1.000		
653	0.541**	0.429**	0.614**	0.699**	1.000	
689	0.485**	0.591**	0.626**	0.770**	0.644**	1.000
Sheath score						
366	1.000					
411	0.505**	1.000				
467	0.276	0.532**	1.000			
578	0.417**	0.578**	0.564**	1.000		
653	0.536**	0.471**	0.308	0.303	1.000	
689	0.359*	0.453**	0.378*	0.349*	0.332	1.000
Navel thickness score						
366	1.000					
411	0.759**	1.000				
467	0.766**	0.718**	1.000			
578	0.756**	0.619**	0.588**	1.000		
653	0.362*	0.489**	0.370*	0.176	1.000	
689	0.782**	0.727**	0.722**	0.660**	0.475**	1.000
Rosette						
366	1.000					
411	0.712**	1.000				
467	0.798**	0.836**	1.000			
578	0.741**	0.564**	0.702**	1.000		
653	0.760**	0.677**	0.804**	0.662**	1.000	
689	0.705**	0.731**	0.780**	0.806**	0.724**	1.000

Table 45. Correlations within physical traits at various average ages of Droughtmaster bulls

\* P<0.05; \*\* P<0.01

# Semen and sperm traits

### Mass activity

There was a significant effect of age (P< 0.001) and sire (P=0.006) on semen mass activity. Predicted means, standard errors and levels of significant differences for sires A, B, C and multiple-sires were  $1.8 \pm 0.15b$ ,  $1.0 \pm 0.24a$ ,  $1.4 \pm 0.20ab$  and  $1.0 \pm 0.20a$  respectively. Mass activity was low initially (0.4 at 366 days of age) then generally increased with age (Table 46).

## Motility

There was no significant effect of sire or age on semen motility with mean values ranging from 49 to 61% (Table 46).

#### Normal sperm

The only significant effect was age (P<0.001). Normal sperm was initially low (mean of 37.3% at 411 days of age), declined further whilst in the feedlot, before subsequently increasing to 58% at 653 - 689 days of age (Table 46, Figure 63).

Traits	366 days	411 days	467 days	578 days	653 days	689 days	lsd
Mass activity (0-5)	0.4a	1.1b	1.1b	1.9c	1.3b	2.1c	0.4
Motility (%)	id	id	49.4a	52.8a	48.8a	60.7a	
Normal sperm (%)	id	37.3 ± 3.0b	27.5 ± 2.5a	36.6 ± 2.4b	58.0 ± 2.5c	58.6 ± 2.5c	
Abnormal heads (%)	id	25.9 ± 2.5b	38.5 ±2.0c	35.6 ±1.9c	14.9 ±2.0a	17.1 ± 2.0a	
Abnormal midpieces (%)	id	17.8 ±1.8a	14.8 ± 1.5a	13.6 ± 1.4a	14.4 ± 1.5a	17.5 ± 1.5a	
Abnormal tails (%)	id	9.5 (2.7) <sup>#</sup> ± 1.5bc	9.5 (2.7) ± 1.2bc	12.2 (4.5) ± 1.2c	6.8 (1.4) ± 1.2b	3.2 (0.3) ± 1.2a	
Cytoplasmic droplets (%)	id	18.3 (9.8) ± 1.9bc	21.4 (13.4) ± 1.5c	14.7 (6.4) ± 1.5ab	16.2 (7.8) ± 1.6b	10.9 (3.6) ± 1.5a	l

Table 46. Semen and sperm traits at various average ages of Droughtmaster bulls

Means within rows not followed by a common letter differ significantly (P<0.05); id Insufficient data; # Back-transformed mean

## Abnormal sperm categories

Trends for the various categories of sperm abnormalities are presented in Table 46 and Figure 63. There was no effect of sire on any of the abnormal sperm categories. With abnormal heads, there was only a significant (P<0.001) effect of age with abnormal heads being highest (38.5%) at 467 days of age when the bulls were in the feedlot, and then significantly decreasing to 14.9% at 653 days of age. With abnormal midpieces, there were no significant effects and mean values ranged from 13.6 to 17.6%. The only significant fixed effect with abnormal tails was age (P<0.001), but overall values ranged from 0.3 to 4.5%. For cytoplasmic droplets, significant main effects were age (P<0.001) and date of birth (P=0.023). The level of cytoplasmic droplets decreased with age and the coefficient for date of birth was 0.110  $\pm$  0.048 indicating that younger bulls had higher levels of droplets.

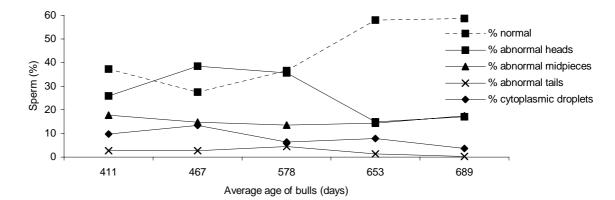


Figure 63. Normal sperm and abnormal sperm categories of Droughtmaster bulls at different ages

# Repeatability of semen and sperm traits

All semen and sperm traits were lowly repeatable (Table 47). For cytoplasmic droplets, repeatability was unable to be estimated since the bull variance component was zero.

Trait	Repeatability	Comment
Mass activity	$0.21\pm0.07$	Low
Motility	$\textbf{0.09}\pm\textbf{0.08}$	Low
Normal sperm	$\textbf{0.25}\pm\textbf{0.08}$	Low
Abnormal heads	$\textbf{0.12}\pm0.08$	Low
Abnormal midpieces	$\textbf{0.30}\pm\textbf{0.09}$	Low
Abnormal tails	$\textbf{0.16} \pm \textbf{0.08}$	Low

Table 47. Repeatability of semen and sperm traits of Droughtmaster bulls

# Correlations of normal sperm at different ages

There was little evidence of any significant relationships between ages for normal sperm except towards the end of the experimental period (Table 48). The best associations were between 578 days and 689 days of age (r = 0.494; P<0.01) and between 411 and 689 days of age (r = 0.465; P<0.05). With the various sperm abnormality categories, again there were few significant relationships between times of measurements.

	411 days	467 days	478 days	578 days	653 days	
	411 days	407 duy5	410 duy5	010 0030	000 days	
411 days	1.000					
467 days	0.285	1.000				
478 days	0.035	0.162	1.000			
578 days	0.328	-0.008	0.431*	1.000		
653 days	0.465*	0.052	0.494**	0.388*	1.000	

Table 48. Correlations of normal sperm of Droughtmaster bulls at different average ages

\* P<0.05; \*\* P<0.01

# Correlations between physical, behavioural and seminal traits

The following traits were significantly correlated (P<0.05) at almost every measurement time:

- Liveweight with scrotal circumference,
- Mass activity with scrotal circumference and motility,
- Interest with mounts, and
- Mounts with serves.

The correlations between physical, mating behaviour and seminal traits were bulked across all times of measurement and are presented in Table 49. The following identify some of the many significant (P<0.05) correlations. However, many were numerically low. Some of the numerically higher correlations were:

- Age with liveweight, scrotal circumference, mass activity and normal sperm,
- Liveweight with scrotal circumference, mass activity and normal sperm,
- Scrotal circumference with mass activity, motility and normal sperm,
- Interest with mounts and serves,
- Mounts with serves.
- Mass activity with motility and normal sperm, and
- Motility with normal sperm.

	Age	Liveweight	Testicular tone	Scrotal circum.	Interest	Mounts	Serves	Mass activity	Motility	Normal sperm
Age	1.000									
Liveweight	0.899**	1.000								
Testicular tone	0.020	-0.063	1.000							
Scrotal circum.	0.762**	0.806**	0.010	1.000						
Interest	0.151*	0.088	0.142	0.032	1.000					
Mounts	-0.103	-0.127	0.068	-0.174*	0.487**	1.000				
Serves	0.133	0.087	0.090	0.089	0.286**	0.428**	1.000			
Mass activity	0.423**	0.429**	-0.050	0.578**	-0.026	-0.123	0.043	1.000		
Motility	0.107	0.076	-0.008	0.238**	-0.215**	-0.142	-0.013	0.510**	1.000	
Normal sperm	0.560**	0.492**	0.014	0.461**	0.178*	-0.021	0.062	0.289**	0.237**	1.000

 Table 49. Correlations between traits across all times of measurement

\* P<0.05; \*\* P<0.01

# 6.2.4.5 Discussion

As a consequence of drought, bulls were relocated to two other locations before returning to Dilga prior to the last measurement in August 2001. The distances travelled were relatively short (<300 km) and had a similar temperature and humidity range to Dilga. As other studies in this project (see Sections 6.1.4 and 6.1.5) found relocation had little effect on semen quality or sperm morphology, it is unlikely that these shifts effected semen quality and sperm morphology of these bulls. However, a combination of relocation, mixing of bulls with unfamiliar cattle, high levels of nutrition in the feedlot and ambient heat loads should not be ruled out as a cause of depression in sperm morphology recorded between November 2000 (411 days of age) and January 2001 (467 days of age).

Sheath depth was relatively constant across at each observation from 12 months to 23 months suggesting that sheath development occurs relatively early in life. The decrease in sheath scores after the bulls had been in the feedlot may be a reflection of operator error in the different facilities and conditions that bulls were assessed in at the feedlot.

The data (Table 43) would suggest that measurement of traits such as sheath depth, sheath score, navel thickness score and rosette score as yearlings (12 months) gives a good indication of their size as 2-yearolds. This allows breeders greater confidence to select bulls 12 months earlier. However, this contrasts with sperm morphology where there was little evidence of any significant relationship between ages for normal sperm except towards the end of the experimental period when average age of bulls was 19 months. The decline in normal sperm with a corresponding rise in abnormal heads from 411 days to 467 days of age corresponds to the initial relocation and feedlot period. This suggests a potential insult to healthy spermatogenesis.

The repeatability of semen and sperm traits was low and correlations between ages for morphology traits were generally inconsistent and where there were significant relationships, r values were less than 0.5. This demonstrates that the measures for these traits in young bulls can vary within relatively short periods. In particular, younger bulls had higher levels of cytoplasmic droplets, which is consistent with the maturity of the bulls and their semen production. There were poor correlations between semen (mass activity, motility) and sperm traits although mass activity and motility were significantly correlated. Normal sperm was significantly correlated with age, liveweight and scrotal circumference. Hence, older heavier bulls will tend to have larger scrotal size and higher normal sperm values.

Interest was related to mounts as was mounts to serves indicating that bulls showing greater interest in the recipient females will also achieve more mounts and serves.

It was not possible to mate these bulls as yearlings to assess their calf output. The data demonstrates that there was a large range in many physical, mating behaviour and seminal traits suggesting that a few of the bulls would have been suitable for yearling mating. Mean scrotal circumference at 14–16 months was 31-33 cm and a few bulls had 2 serves in a serving capacity test by 14 months of age, demonstrating a capability to mate. However, whilst they would have passed the minimal standards for scrotal size at this age, mean normal sperm was only 38% and the maximum was 68%, which is still slightly lower than the AACV standard for normal sperm for purchased bulls.

# 6.2.5 Fertility of 5/8 Brahman yearling bulls – Swan's Lagoon Research Station

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# 6.2.5.1 Summary

Fifty 5/8 Brahman x 3/8 Shorthorn bulls weaned in May 2000 were supplemented till 30 October 2000 to achieve a growth rate of 0.4 kg/day over the dry season. Five of these bulls, and 3 (un-supplemented) bulls one year older, were selected, on the basis of a bull breeding soundness examination (BBSE) in January 2001 for 3 months single-sire mating, each to a group of 40 non-pregnant, non-lactating heifers and cows of the same genotype.

Yearling bulls were not sexually mature at mating, and 2 of the 5 mated did not reach the 50% threshold for normal sperm at the start of mating. Semen quality of one bull weighing only 264 kg pre-mating deteriorated over mating and failed to reach acceptable thresholds for both semen motility and normal sperm post-mating. Few of the heifers and cows mated with this bull became pregnant.

The rate at which females became pregnant was lower in these remaining yearling bulls than those that became pregnant to the 2-year-old bulls (4.3 v 5.2 estimated pregnancies per week; P<0.001). It is noted that caution should be exercised in extrapolating this result to mating of bulls in multiple-sire groups, where stimulation caused by competition may create different outcomes.

# 6.2.5.2 Aim

The aim of this study was to measure physical and reproductive traits of yearling bulls in a dry tropical region and to assess their reproductive performance by measuring conception patterns and pregnancy rates. The performance of the yearling bulls was compared with 2-year-old bulls.

# 6.2.5.3 Materials and methods

It is well established that bulls that pass a BBSE at 2 years of age have similar calf-getting ability to mature bulls. If younger bulls could be successfully mated, the annual genetic gain could be accelerated and lifetime calf-output of bulls could be increased.

#### Experimental site and animals

The study was conducted at Swan's Lagoon Research Station in the dry tropics of north Queensland. The cattle grazed native pastures, predominated by spear grass (*Heteropogon contortus*), on relatively flat savannahs. The duplex soils are of low fertility. The cattle used in this experiment were selected from a stabilised herd of approximately 5/8 Brahman, with 3/8 *Bos taurus,* which was mostly Shorthorn.

#### Procedures

The experiment was conducted during a period of below-average rainfall, which resulted in relatively low weights of experimental animals. At weaning of the commercial herd in May 2000, 50 bull calves (average age 6 months) were left entire. These calves were subsequently offered an *ad lib.* supplement of molasses with 10% cottonseed meal and 5% urea added for the duration of the dry season. Supplementation ceased when storm rains, on 30 October 2001 stimulated green grass growth sufficient to satisfy voluntary intake by the bulls.

At the same time, bulls weaned in May 1999 were grazed on similar pastures without supplements within their own cohort. None of these bulls had been mated prior to this experiment.

After a standard BBSE (Entwistle and Fordyce 2003) of all available bulls on 15 January 2001, 5 yearling (average age 14 months) and three 2-year-old bulls (average age 26 months) were selected for mating in this experiment. The yearling bulls selected were the most reproductively advanced within their cohort (Table 50). These bulls were yarded overnight with females in oestrus and then submitted to a standard 20-minute serving capacity test using restrained oestrus females on the morning of 25 January 2001.

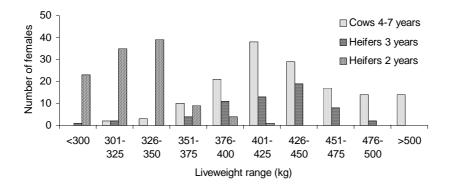
On 23 January 2001, non-lactating, non-pregnant females (148 cows aged 4-7 years, 60 heifers aged 3 years, 111 heifers aged 2 years; average weight of 440 kg) were allocated to 2 blocks of four 100 ha paddocks by stratified randomisation on weight within condition score (13% score 6, 65% score 7, 22% score 8; 9-point scale) and age (Figure 64).

Bulls were randomly allocated to each block of paddocks on age so that in one block there was three yearling bulls and one 2-year-old bull and in the other block two yearling and two 2-year-old bulls. Each bull was therefore single-sire mated to a group of 40 females, i.e., at 2.5% bulls:females.

		-	-							
Bull ID	Liveweight	Condition score	Scrotal circumference	Testicular tone	Sheath score	Semen density	Mass activity	Motility	Normal sperm	Primary sperm abnormality
	kg	1-9	cm	1-5	1-9	1-5	0-5	%	%	
Yearling	bulls: F	Pre-mati	ng							
000221	373	8	33.5	3	3	4	2	40	47	Loose heads
000233	319	8	27.0	3	6	1	0	70	53	Dag defect
000404	264	7	27.0	3	7	3	1	80	52	Proximal droplets
000768	336	7	26.0	3	7	2	0	60	65	Pyriform heads
000826	310	7	29.5	3	7	3	1	50	43	Pyriform heads
Two-yea	r-old bu	Ills: Pre-	mating							
990034	388	7	32.0	3	9	2	0	40	68	Dag defect
990043	435	7	30.5	3	5	3	1	90	75	Distal cytoplasmic droplets
991503	457	7	32.5	3	6	4	2	80	67	Loose heads
Yearling	bulls: F	Post-ma	ting							
000221	388	7	32.0	3	2	5	4	90	68	Coiled tails
000233	359	7	28.5	3	5	3	0	60	61	Distal mid-piece reflex
000404	288	7	27.0	3	6	3	0	10	34	Bent mid-piece
000768	368	7	27.5	3	7	4	2	70	44	Bent mid-piece
000826	352	7	31.5	3	6	3	2	70	43	Proximal droplets
Two-yea	r-old bu	Ills: Pos		-						
990034	451	7	33.5	3	9	4	3	70	73	Distal mid-piece reflex
990043	468	6	30.5	3	6	5	5	90	71	Bent mid-piece
991503	490	7	32.5	3	6	3	3	90	58	Bent mid-piece

#### Table 50. Pre- and post-mating physical and semen evaluation of bulls

At the end of mating on 23 April 2001, and 8 weeks later, a rectal examination of all cows was conducted to estimate foetal age, at the same time as weighing and scoring of body condition. BBSE of bulls was also conducted at the end of mating (Table 50.



# Figure 64 . Weight distribution of non-lactating females allocated to mating groups

# Statistical analyses

A survival analysis compared the conception patterns resulting from the matings of the 1 and 2 year-old bulls by including age as a factor in the analysis and setting 2 year-old level as the base-line hazard.

# 6.2.5.4 Results

The yearling-mated bulls grew at an average of 0.4 kg/day between weaning and the end of mating (almost 12 months; Figure 65). Their scrotal circumference increased by 7 cm in the 5 months prior to mating, but only increased by 0.7 cm during the 3-month mating.

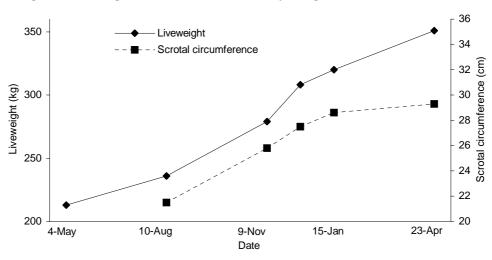


Figure 65. Liveweight and scrotal circumference of yearling 5/8 Brahman bulls

The 2-year-old bulls weighed an average of 427 and 470 kg, with an average scrotal circumference of 31.7 and 32.2 cm at the start and end of mating, respectively. All bulls remained close to condition score 7 (9-point scale), with an average testicular tone of 3 (5-point scale).

Pre- and post-mating, all bulls passed a BBSE for physical, scrotal and semen traits, except for one yearling that failed semen assessment post-mating (Table 50). All 2-year-old bulls passed a sperm morphology assessment and the serving assessment (Tables 50 and 51). Pre-mating, 2 yearling bulls failed the sperm morphology assessment, but all were close to the 50% normal threshold. No yearling bulls passed the serving assessment, though 3 mounted. At the end of mating, 3 yearling bulls failed sperm morphology assessments.

Most pregnancies occurred within 6 weeks, or the equivalent of 2 oestrus cycles, of the start of mating. The analysis of conception patterns (Figure 66) excluded the poorest-performing, 1-year-old bull (Bull ID 000404 in paddock DMG); he was small (264 kg) at mating. Two-year-old bulls impregnated cows at a higher rate than did yearling bulls (P<0.001); hazard ratios were 1.00 and 0.59 (95% confidence interval of 0.45-0.77), respectively.

Pregnancy rate was unrelated to age, weight, or condition of females, except that pregnancy rate in the first 6 weeks increased directly with start of mating weight in 2-year-old heifers (Figure 67).

Bull ID	Interest	Mounts	Serves
	n	n	n
Yearling bulls			
000221	2	0	0
000233	0	3	0
000404	0	0	0
000768	0	3	0
000826	7	5	0
Two-year-old bulls			
990034	0	4	1
990043	3	3	1
991503	4	11	2

# 6.2.5.5 Discussion

Because of the poor nutritional conditions at the experimental site, growth of yearling bulls was poor. All yearling bulls used had reached puberty, but clearly were not sexually mature. This was emphasised in the serving assessment, where despite all bulls being heterosexually inexperienced, all 2-year-olds served, whilst no yearlings served.

The sexual immaturity of the yearling bulls may explain why they were unable to achieve the same pregnancy rates as 2 year-old bulls. However, they did achieve an estimated average of 4.3 pregnancies per week, compared with 5.2 for the 2 year-old bulls.

This study was conducted using single-sire mating. The outcome may not reflect potential fertility in multiple-sire matings where competition between bulls may stimulate several aspects of bull behaviour and function that contribute to calf-getting ability. It is certainly suspected that fertility in multiple-sire matings would not be lower.

The very low fertility of bull 000404 appeared related to his semen quality, which deteriorated substantially over mating; both motility and percent normal were below standard thresholds of 30% and 50% (Entwistle and Fordyce 2003), respectively. The small size of this bull and his failure to show even interest during a serving assessment were further expressions of his immaturity.

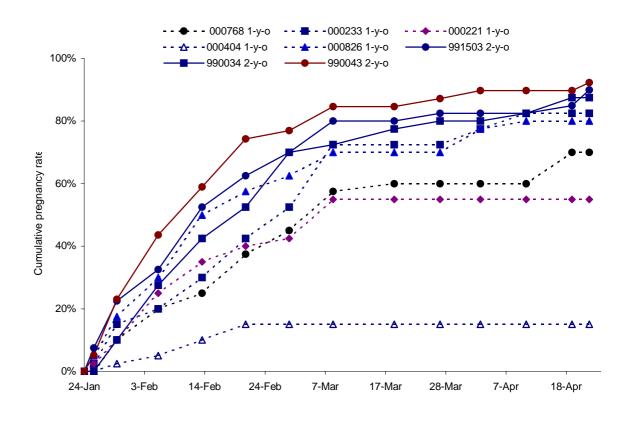
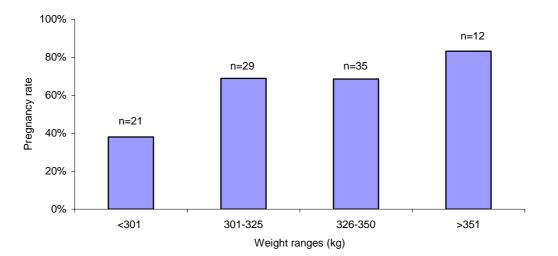


Figure 66. Cumulative pregnancy rates of heifers and cows mated to individual 5/8 Brahman bulls

Figure 67. Pregnancy rates by weight classes in the first 6 weeks of mating (Data for heifers mated to bull 000404 excluded)



# 6.2.6 Sexual development in Belmont Red bulls – Narayen Research Station

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# 6.2.6.1 Summary

Belmont Red bulls were examined at approximately six-week intervals between 9 and 24 months of age. Liveweight, scrotal circumference and normal sperm all increased whilst their mating behaviours changed to fewer mounts and increased number of serves in a serving capacity test. Mean normal sperm increased from 48% at 12 months of age to 85% at 16 months of age as the bulls matured. Data were collated from an earlier study on 4 Belmont Red yearling bulls that were mated as multiple-sires with individual calf-output identified by DNA typing for paternity. A calving rate of 97.5% was achieved during a 13 week mating with 81 mature age breeders and the percentage of calves by each sire were 0, 7, 37 and 53%. Yearling bulls should be selected on the basis of normal testicular development, percent normal sperm and ability to serve.

# 6.2.6.2 Aim

The aims of the experiment were to measure, from 9 months through to 2 years of age, physical, behavioural and reproductive traits of Belmont Red bulls; to correlate and measure the repeatability of these traits and to measure calf output of yearling bulls when mated at 13 to 15 months of age.

# 6.2.6.3 Materials and methods

#### Experimental design

The experiment was a longitudinal study of physical, behavioural and reproductive traits of 24 Belmont Red bulls and the 4 bulls mated to 81 cows in November 1995.

#### Experimental site and animals

The longitudinal study was conducted on Narayen Research Station (26<sup>°</sup> S, 150<sup>°</sup> E) near Mundubbera on undulating brigalow lands in central Queensland. The study was conducted from 3 January 1998 to 23 August 1999 using 24 Belmont Red bulls representing 12 sires. The birth dates of bulls were from 2 August 1997 to 12 October 97 with mean age of 9 months (range of 234 to 305 days) at the commencement of the experiment.

The calf output study of yearling bulls was also conducted at Narayen, but prior to the commencement of the Bullpower project. These results were collated for inclusion in the studies of assessing the feasibility of mating bulls as yearlings.

#### Procedures

Bulls were run as one group on pasture, but not all bulls were mustered on each occasion. Bulls were mustered to the yards on the 3 June 1998, weighed full then subjected to a physical and reproductive examination and semen collected for morphological assessment. Bulls remained in the yards overnight and the following day they were examined in a serving capacity test. These procedures were then repeated 5 times on 1 September 1998, 26 October 1998, 14 January 1999, 12 April 1999 and 23 August 1999.

Four Belmont Red bulls (aged 13-15 months) were multiple-sire mated to a group to 81 cows with calves at foot for 13 weeks (1 November 1995 to 2 February 1996). These 4 bulls and all calves resulting from

this mating were bled at branding for DNA testing and samples submitted to the Cattle DNA Typing Laboratory at the University of Queensland. Methods for paternity testing have been reported by Holroyd *et al.* (2002a).

### Statistical analysis

All statistical analyses were performed using GenStat for Windows 6<sup>th</sup> Edition.

#### All traits except scrotal circumference

The average age of the 24 Belmont Red bulls at each time point were 273, 363, 418, 498, 586 and 719 days. A linear mixed model was used to analyse all traits recorded across time except scrotal circumference. The full model fitted was:

- Fixed effects: *bull starting age + age*
- Random model: *bull* + *bull.age*

#### Where

- bull starting age is a covariate the age of each bull in days on 3 June 1998 (start of recording),
- age has between 4 and 6 levels 273, 363, 418, 498, 586 and 719 days, and
- bull represents the variance component for bulls,
- *bull.age* represents the residual variance

Random terms with zero variance components were removed from the model, then Wald tests were used to identify non-significant fixed terms (P>0.05) which were also removed to arrive at the final model. Approximate least significant differences were calculated for significant (P<0.05) fixed effects.

Repeatability (intraclass correlation) was estimated as the ratio of variance components:-

Repeatability=bull/(bull + bull.age)

#### Scrotal circumference

A random coefficients regression approach was used to analyse scrotal circumference from the 6 times. This allowed modelling of individual animal patterns over time. The average growth pattern for scrotal circumference was approximately quadratic over the time span considered for the majority of bulls. However, individual bulls have their own growth pathway so that there is potentially variation in intercepts, slopes and curvature across bulls. This is represented by random intercepts and slopes for individual bulls and correlations between the random intercepts and slopes.

The full model fitted for scrotal circumference was

- Fixed effects:  $age + age^2$
- Random effects: *bull* + *bull.* age + *dev(age)*

#### Where

- age is the average age (days) of the bulls at each time of measurement,
- *bull* represents the random intercepts,

- bull. age represents random linear trend, and
- *dev(age)* is a random lack of fit term for additional fluctuations about the linear trend.

Random terms with zero variance components were removed from the model, then Wald tests were used to identify non-significant fixed terms (P>0.05) which were also removed to arrive at the final model.

A chi-square test assessed equality of calf-output (as measured by percentage of calves per bull) within the mating group of the 4 Belmont Red bulls.

#### 6.2.6.4 Results

#### **Physical traits**

#### Liveweight

The response was linear until 586 days of age then levelled. There were significant (P<0.001) differences between ages. Bulls increased in mean liveweight from 242 kg to 481 kg between 273 and 719 days of age as shown in Figure 68 and Table 52.

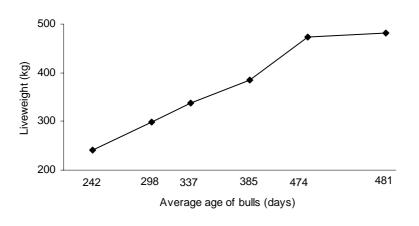


Figure 68. Liveweight of Belmont Red bulls at different ages

#### Testicular tone

Testicular tone was recorded 4 times. There was little variation in testicular tone across bulls or times with most values being in the score range of 3.5 to 4.

#### Scrotal circumference

There was a significant (P<0.001) quadratic response to time where the predicted smooth model for scrotal circumference is presented in Figure 69. Of the 24 bulls, 22 had a scrotal circumference of at least 34 cm (the minimum recommended by the AACV at 24 months, Entwistle and Fordyce 2003).

#### Mating behaviour traits

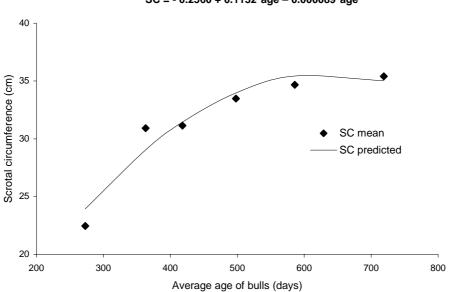
#### Interest

The only significant (P=0.002) effect was age (Table 52). Predicted means are presented in Table 52. There was a significant decrease in interest at the last observation at 719 days of age.

Trait	273 days	363 days	418 days	498 days	586 days	719 days	lsd
Liveweight (kg)	242a	298b	337c	385d	474e	481e	12.2
Interest (n)	2.5b	2.1b	2.9b	2.7b	3.0b	0.6a	1.3
Mounts (n)	4.5a	7.2bc	8.3c	5.3ab	6.2abc	5.3ab	2.3
Serves (n)	0.0a	0.5a	0.3a	1.5b	1.9b	1.9b	0.7
Mounts+serves (n)	4.4a	7.6b	8.6b	6.8b	8.1b	7.2b	2.2
Libido score (0-10)	4.6a	6.5b	6.3b	7.5c	7.8c	8.5c	1.1
Mass activity (0-5)	id	id	2.5a	3.2b	3.7b	3.6b	0.7
Normal sperm (%)	id	48.3a	73.6b	85.3c	84.8c	83.0c	8.6

Table 52. Physical, behavioural and reproductive traits of Belmont Red bulls at different ages

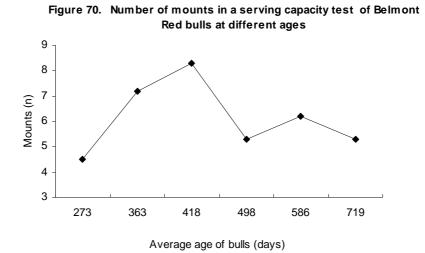
Means within rows not followed by a letter in common differ significantly (P=0.05); id, insufficient data



# Figure 69. Scrotal circumference (SC) of Belmont Red bulls at different ages $SC = -0.2560 + 0.1132^{*}age - 0.000089^{*}age^{2}$

#### Mounts

There was a significant (P=0.018) effect of age (Table 52). The mean number of mounts increased from 4.5 at 273 days of age to 8.3 at 418 days of age then declined to 5.4 at 719 days of age (Figure 70).



#### Serves

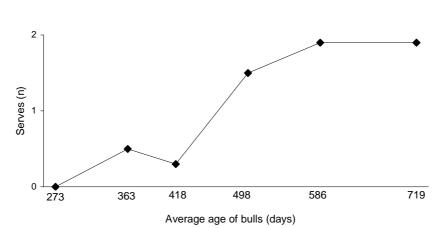
There was a significant (P<0.001) effect of age (Table 52). Mean number of serves steadily increased from 0 at 273 days of age through to 1.9 at 586 days (Figure 71). Of the 24 bulls, 20 successfully served at least once. The average age of first service was 476 days with a range from 260 to 723 days. Of those bulls that did achieve at least one serve, the total number of serves over the 6 measurement times ranged from 1 to 18 serves with a mean of 6.5.

#### Mounts and serves

There was a significant (P=0.007) effect of age (Table 52) with mounts+serves being significantly lower at 273days of age than at older ages.

#### Libido score

There was a significant change with age (P<0.001) with scores increasing from 4.6 at 273 days of age through to 8.5 at 719 days of age (Table 52).



# Figure 71. Number of serves in a serving capacity test of Belmont Red bulls at different ages

112

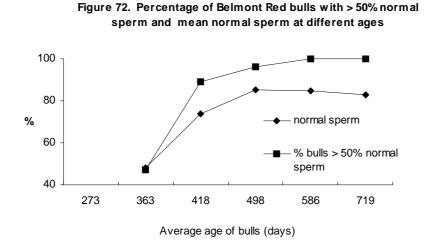
#### Semen and sperm traits

#### Mass activity

At 273 and 363 days of age there was insufficient semen collected to record mass activity. From then on, there was a significant (P=0.004) effect of age with mass activity at 418 days of age (score 2.5) being significantly less than at subsequent dates (scores 3.2 - 3.7, Table 52).

#### Normal sperm

There were so few bulls producing semen at 273 days of age that results for this observation were not included in the analyses. There was a significant (P<0.001) effect of age with mean values for normal sperm at 363 days of age being 48.3%, rapidly increasing to 73.6% by 418 days of age then plateauing at 83.0% by 719 days of age (Table 52, Figure 72). Of all the bulls that produced semen, 67% had >70% normal sperm by 418 days of age and 100% had >70% normal sperm by 719 days of age.



#### Repeatability of physical, behavioural and semen traits

Liveweight and scrotal circumference were moderately repeatable whilst behavioural and semen traits were lowly repeatable (Table 53).

Trait	Repeatability	Comment						
Liveweight	$0.61\pm0.09$	Moderate						
Scrotal circumference	$\textbf{0.49} \pm \textbf{0.10}$	Moderate						
Interest	$\textbf{0.19}\pm\textbf{0.09}$	Low						
Mounts	$\textbf{0.18} \pm \textbf{0.09}$	Low						
Mounts+serves	$0.31\pm0.10$	Low						
Libido score	$\textbf{0.29}\pm\textbf{0.10}$	Low						
Mass activity	$\textbf{0.13}\pm\textbf{0.12}$	Low						
Normal sperm	$0.33\pm0.11.$	Low						

Table 53. Repeatability of physical and behavioural traits ofBelmont Red bulls

### Correlations of normal sperm at different ages

Correlations between adjacent ages were reasonably high but these correlations tended to reduce for ages further apart (Table 54).

#### Table 54. Correlations of normal sperm at different averages ages of Belmont Red bulls

	363 days	418 days	498 days	586 days	719 days
363 days	1.000				
418 days	0.702**	1.000			
498 days	0.035	0.760***	1.000		
586 days	0.328	0.752***	0.677***	1.000	
719 days	0.130	0.467	0.367	0.439	1.000
	** D <0 01· **	**D <0 001			

\*\* P<0.01; \*\*\*P<0.001

#### Correlations between physical, behavioural and seminal traits

The correlations between physical, behavioural and seminal traits were bulked across all times of measurement and are presented in Table 55. There were many significant (P<0.001) correlations although some of the values were numerically small. Some of the important biological relationships were:

- Age with weight
- Scrotal circumference with age and liveweight
- Serves with age, liveweight and scrotal circumference
- Mass activity with age, liveweight and scrotal circumference
- Normal sperm with age, liveweight, scrotal circumference and mass activity

	Age	Liveweight	Scrotal circum.	Interest	Mounts	Serves	Mass activity	Normal sperm
Age	1.000							
Liveweight	0.922***	1.000						
Scrotal circumference	0.790***	0.830***	1.000					
Interest	-0.205*	-0.131	-0.060	1.000				
Mounts	-0.040	-0.056	0.107	0.169	1.000			
Serves	0.469***	0.441***	0.409***	-0.066	-0.018	1.000		
Mass activity	0.285**	0.337**	0.286**	0.000	-0.169	0.171	1.000	
Normal sperm	0.407***	0.448***	0.438***	-0.036	-0.096	0.142	0.423***	1.000

Table 55.	Correlations between	traits across all	times for Be	Imont Red bulls
10010 001	een elaliene setheen	thanto aono oo an		

\*\* P<0.01; \*\*\* P<0.001

# Calf output of 4 bulls mated in November 1995

At commencement of mating, the scrotal circumference of bulls ranged from 20.5 to 26.5 cm. The females that the bulls were mated to had a calving rate of 97.5%. The percentage of calves sired by each bull were 0, 7.4, 37.0 and 53.1 from a 13 week mating with 2.5% of calves unable to be assigned to a particular sire. A chi-square test indicates the bulls did not have equal calf-output ( $\chi^2$ =77; P<0.001).

# 6.2.6.5 Discussion

In this study, scrotal circumference was less than those reported for a range of other beef breeds. As liveweight increased so did scrotal circumference and percentage of normal sperm. This is consistent with the suggestion that scrotal circumference is positively related to measures of semen morphology. By 14 months of age, 67% of bulls had at least 70% normal sperm and by 16 months, most bulls had served at least once in a serving capacity test. The percentage of Belmont Red bulls that achieved at least one serve was greater than that reported for other tropically adapted breeds with about 25% of bulls achieving a serve by 12 months and 83% by 24 months.

There was a large difference in the number of calves sired by the 4 yearling bulls within the 13-week mating, November 1995 to February 1996. The bull that sired no calves also had the smallest scrotal circumference (20.5 cm) at the time of commencement of mating compared to the scrotal circumference (26.5 cm) of the bull that produced the most calves. The variation in calf-output is consistent with our previous work where one bull sired no calves yet another sired 53% of the calves when mated with mature age cows.

# 6.2.6 6 Conclusions

There was considerable variation in age at which bulls reach sexual maturity. Some bulls had high (>70%) normal sperm at an early age (12 months old), but others were up to 8 to 12 months older before they reached this threshold. This would indicate the potential for selecting Belmont Red bulls for sexual maturity at an early age. In this study, the 2-month interval between examinations was too large to precisely determine the age at which each bull achieved one serve and had an ejaculate with at least 70% normal sperm. Although a direct comparison with 2-year-old bulls was not made, some yearling bulls appear to have similar capabilities of siring calves, in a 3-month mating period, as older bulls.

# 6.3 Increasing the working life of bulls – Identifying traits that predispose bulls to preputial prolapse

# 6.3.1 Background

Problems associated with the sheath and prepuce can cause a economic loss in *Bos indicus* and *Bos indicus* derived bulls in northern Australia. These studies examined the extent of the problem, examined the sheath and prepuce (and related structures) in normal bulls, examined the organs in bulls with preputial prolapse to compare with normal bulls and studied the function of the penis and prepuce in live normal bulls to understand some of the mechanisms involved in eversion.

# 6.3.2 Survey of reasons for culling bulls

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A survey was conducted to determine the extent of preputial and related problems in bulls in northern Australia. Little information is available in the literature on the relative importance of specific reasons for culling bulls in the extensive areas of northern Australia. This survey covered eight cattle properties in northern Australia under the control of one pastoral company where 900 bulls were culled in the 1998 season.

Most (66%) of the bulls were culled for age or because they were surplus to requirements. The remaining culled bulls were culled for specific problems but totalled only 5% of the total number of bulls on the properties at that time. Of this 5%, the most important conditions identified were visual reproductive problems (28%), conformation (17%) and temperament problems (13%). The visual reproductive problems were mainly due to damaged penises (12%) and prepuces (9%).

# 6.3.3 A quantitative anatomical study of the sheath and prepuce of Santa Gertrudis bulls in relation to the occurrence of preputial eversion

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# 6.3.3.1 Summary

Eight polled and 32 horned bulls in a single herd were scored for sheath development, prepuce eversion and sheath skin thickness. After slaughter, various anatomical features of the sheath, prepuce and carcase weight were recorded. Relationships between the anatomical measurements, carcase weight, and scores were analysed statistically.

Polledness had no significant relationship to preputial eversion score, sheath score or the retractor

preputial muscle measurements. Sheath score was not significantly related to carcase weight, but the owner selecting bulls within the herd simultaneously for less pendulous sheaths and better growth rates may have influenced this. Preputial eversion score was not significantly related to the diameter of the preputial orifice, the retractor preputial muscle measurements or the length or weight of the penis. Preputial eversion was significantly (P < 0.05) negatively correlated with the preputial orifice to preputial fornix measurements but was not significantly correlated with the other five preputial measurements. Bulls with larger sheaths (Breedplan score < 3) displayed significantly (P < 0.05) more preputial eversion than those with smaller sheaths (Breedplan score  $\geq 3$ ).

The predisposing factors for preputial eversion may differ between *Bos indicus* derived breeds and *Bos taurus* breeds. There was no evidence of poorer preputial muscle development or more preputial eversion in polled than in horned bulls. The relationship between preputial eversion and sheath score may justify selection against *Bos indicus* derived bulls with pendulous sheaths if preputial eversion leads to an increase in preputial injury or infection.

# 6.3.3.2 Introduction

Baseline anatomical measurements are required to establish the range of normal variation found in the sheath and prepuce in bulls of *Bos indicus* derived breeds. Such information is necessary for an examination of the links between anatomical characteristics and problems developing in the sheath and prepuce. No comprehensive studies of the anatomy and function of muscles associated with the sheath and prepuce in *Bos indicus* derived bulls have been reported. Differences in these muscles between *Bos indicus* derived bulls that are polled or horned or between those that do or do not exhibit preputial eversion have not been adequately examined. The present study examines the sheath, prepuce and associated structures in bulls of one *Bos indicus* derived breed (Santa Gertrudis) to establish baseline data. Also examined were the relationships between the results of scoring systems for these organs in the live animal and measurements taken post-mortem at an abattoir.

# 6.3.3.3 Aim

The aim of the experiment was to establish baseline anatomical measurements of the sheath and prepuce in Santa Gertrudis bulls, to relate these to currently used scoring systems in current use and to establish whether sheath and prepuce measurements were associated with preputial eversion.

# 6.3.3.4 Materials and methods

#### Experimental animals

One Santa Gertrudis bull aged two years and 39 aged three years, from a central Queensland property were examined on-farm and scored by the owner for various traits related to their sheath and prepuce. The bulls were slaughtered at a local abattoir where their external reproductive organs were collected and dissected. All animals were considered sound by the owner and had been culled as excess to stud sale requirements. Eight were polled and 32 were horned.

# Definitions

*Sheath* - The hair-covered skin appendage that supports and protects the penis along the ventral abdomen. It extends from the scrotum to the preputial orifice.

*Prepuce* - The hairless epithelium within the sheath that extends proximally from the preputial orifice and is reflected distally at the fornix before attaching to the penis.

*Preputial eversion* – Temporary protrusion of a variable length of prepuce from the preputial orifice.

#### Farm scores

Sheaths were scored 4 times on-farm by the owner, an experienced cattle judge, before the bulls were slaughtered. Bulls were classed as polled only if totally polled. Sheath size was scored using the 9 scores of the Breedplan validation project scoring system (Agricultural Business Research Institute, Armidale). From these categories, sheath size was classed for analysis as either "small" (Breedplan score  $\geq$  3) or "large" (Breedplan score < 3). Preputial eversion was scored as: 1 = none, 2 = slight (< 4 cm), 3 = moderate (4 to 8 cm) and 4 = excessive (> 8 cm). Sheath skin was subjectively assessed as: 1 = thin, 2 = moderate and 3 = thick.

#### Abattoir procedures

The organs were dissected at a local abattoir within 12 h of slaughter and were measured and photographed. Hot carcase dressed weight (kg) was obtained from the abattoir carcase data.

#### Organ measurements

#### Prepuce

1. *Preputial attachment to end of penis* (mm) - The penis was fully extruded and the distance from the point of attachment of the prepuce to the tip of the penis was measured.

2. Fornix to attachment (mm) - The penis was fully extruded and the distance from the fornix (the point at which the surface of the mucosa changes from smooth to irregular) to the attachment of the prepuce to the penis was measured.

3. *Preputial orifice to fornix* (mm) - The penis was fully extruded and the connecting prepuce allowed to relax. The distance from the orifice (defined as the hair junction) to the fornix was measured.

4. *Preputial orifice to fornix* (detached) (mm) - The prepuce was dissected from the other structures and was suspended and measured from the hairline at the orifice to the fornix.

5. Total length of prepuce (mm) - The detached prepuce was suspended and measured.

6. Prepuce weight (g) - The detached prepuce was weighed.

#### Muscles of the penis and prepuce

1. Vertical thickness of penile retractor muscle (mm) – Dorso-ventral measurements of the muscle were taken with a ruler just posterior to the insertion of the muscle onto the penis.

2. *Width of penile retractor muscle* (mm) – The lateral transverse measurement was taken immediately posterior to the insertion of the muscle onto the penis.

3. Vertical thickness of preputial retractor muscle (mm) - This dorso-ventral measurement of the muscle was taken midway along the body of the muscle.

4. *Width of the preputial retractor muscle* (mm) - From a set height above the organs, photographs were taken. From these photographs, the lateral transverse diameter of the preputial retractor muscle was measured midway between the orifice and the base of the scrotum using the ruler included in the photographs.

5. *Preputial retractor muscle network* - The preputial retractor network was scored from photographs. Scores ranged from 1 to 5. (1 = network absent or only 1 to 2 muscle strands from each muscle present, 2 = network of 3 muscle strands, 3 = network of 4 muscle strands, 4 = dense network of more than 4 muscle strands, 5 = broad network of more than 4 muscle strands).

For the examination a 'small network' is defined as a network of 3 or less preputial retractor muscle

strands visible on the dorsal surface surrounding the prepuce. A 'large network' has 4 or more strands.

#### Penis

1. Dorso-ventral diameter of penis (mm) and the lateral transverse diameter of the penis (mm) - These were measured immediately proximal to the insertion of the retractor penis muscles.

2. *Penis length* (mm) - The suspended penis was measured from the tip of the penis to the distal boundary of the ischiocavernosus muscle.

3. Weight of penis (g) – For weighing, the penis was transected at the distal end of the ischiocavernosus muscle.

#### Sheath skin measurements

1. *Thickness at the preputial orifice* (mm) - Callipers were inserted into the orifice and the thickness of the skin a distance of 2 cm from the orifice was measured.

2. *Umbilicus measurement* (mm) – The width of the umbilical cord remnant was measured and included the double layer of skin of the sheath that covers the cord remnant.

3. Sheath skin thickness (mm) - The skin thickness was measured with callipers near the midpoint between the orifice and the base of the scrotum.

4. Sheath skin area to weight ratio (mm<sup>2</sup>/g) - A rectangular area of hide midway between the preputial orifice and the base of the scrotum was removed. The area was determined and the sample weighed.

#### Preputial orifice

*Preputial orifice diameter* (mm) - A conical object was inserted firmly into the orifice and released to obtain a standard pressure on the orifice. The measured distance along the cone at the orifice was converted into the calculated diameter.

#### Statistical analysis

Due to the low frequencies of many of the abattoir and on-farm score values, each of the 5 scores were collapsed to two-valued scores and selected  $2 \times 2$  cross-tabulations were calculated from these. Where all 4 frequencies in the margins of a  $2 \times 2$  table were greater than 10, the chi-square test of association was used, but otherwise the Fisher-Irwin exact test tail probability was calculated.

For each of the on-farm and abattoir measurements and scores, a one-way analysis of variance (ANOVA) was done in which the score (or the collapsed score) values were taken to be treatment levels in a one-way analysis of variance. Treatment significance was tested (F-test) at the 5% level. Possible relationships among abattoir measurements were examined by calculating a matrix of all possible pairwise simple correlation coefficients.

The *Statistix* package (Analytical Software, Tallahassee FI) was used to compute tests of association, ANOVAs and correlations.

#### 6.3.3.5 Results and Discussion

Polledness in *Bos indicus* derived bulls is considered by many cattlemen to be related to larger sheaths and penises. In the 8 poll bulls in the present study, polledness (homozygosity was not known) was not significantly correlated with preputial eversion score, sheath score, or penis length or weight (Tables 56 and 57). The polled characteristic had no significant relationship with the width, the vertical thickness, or the network of preputial retractor muscle bands. Although the difference was not significant, the average preputial retractor muscle network was more extensive in polled than in horned bulls. It has been suggested by some researchers (Rice 1987; Bruner and Van Camp 1992) that polled *Bos taurus* bulls are

susceptible to preputial prolapse because of a heritable weakness of the retractor and protractor muscles of the prepuce. No such reduction in the retractor muscle mass was observed in the polled *Bos indicus* derived bulls in this study. Protractor preputial muscles were present in all 40 bulls examined in this study, but were not measured.

For the purpose of this discussion it is assumed that sheath size, sheath score, sheath area and the degree to which the sheath is pendulous are similar and can be generally compared. In the present study, sheath score was not significantly related to carcase weight. This finding contradicts much of the literature where more pendulous sheaths have been linked to heavier cattle or greater body weight gains (Franke and Burnes 1985; Hoogenboezem and Swanepoel 1995; Bertram *et al.* 1997). In the present study it may reflect selection over time by the herd owner against more pendulous sheaths while continuing to increase growth rates in this herd. Thus we should be cautious in assuming that the data collected so far in this study reflects the true normal distribution of measured anatomical dimensions about the mean for this breed. Some results from the literature were in agreement with the present study and found no significant correlation between body weight and sheath depth (Lagos and Fitzhugh 1970; Bertram *et al.* 1997).

	Sheath		Prepu	itial eversion	Preputial retractor muscle network		
Classification	Small	Large	None	Eversion	Small	Large	
Horned	15 <sup>a</sup>	17	18	14	17	15	
Polled	4	4	3	5	3	5	
P = 1.00			F	P = 0.44	P = 0.69		

# Table 56. Relationship between polledness and sheath, preputial eversion and the preputial retractor muscle network.

<sup>a</sup> Results are given as the number of bulls in each category

Table 57. Relationship between polledness and the size of the preputial retractor muscle, the prepuce	
and the penis	

		Preputial retractor muscle		Pre	puce	Penis	
		Width (mm)	Vertical thickness (mm)	Weight (g)	Total length (mm)	Length (mm)	Weight (g)
Classification	n	Mean ± SD	$\text{Mean}\pm\text{SD}$	$\text{Mean} \pm \text{SD}$	$\text{Mean}\pm\text{SD}$	$\text{Mean}\pm\text{SD}$	$\text{Mean}\pm\text{SD}$
Horned	32	$40\pm17$	$4.3\pm2.0$	$174\pm50$	$369\pm52$	$1100\pm 66$	$883\pm92$
Polled	8	$50\pm8$	$\textbf{3.6} \pm \textbf{1.7}$	$201\pm50$	$390\pm71$	$1120\pm41$	$866\pm104$
		P = 0.12	P = 0.3	P = 0.7	P = 0.3	P = 0.5	P = 0.7

The extent of preputial eversion was not significantly (P > 0.05) associated with the diameter of the preputial orifice, length or weight of the penis, any of the preputial retractor muscle measurements, or with carcase weight or skin measurements. However, preputial eversion was significantly (P < 0.05) related to the distance from the preputial orifice to the fornix (measured while the prepuce was attached to the penis) but not to any of the other preputial length measurements; bulls that exhibited preputial eversion had shorter preputial orifice-fornix measurements than those that did not.

Eversion of the prepuce is seen by many authors to be a major predisposing factor in preputial injury and

infection (Johnson and Williams 1968; Supple-Kane 1969; Walker 1980). Others found that most cases of preputial prolapse requiring surgery have been in breeds in which preputial eversion is common (Monke 1976). This link was not universally observed and in another study there was no greater incidence of preputial disease in bulls that displayed preputial eversion than in those which did not (Long 1969). This was supported by a study of 487 bulls that reported the presence of preputial ulcers that may predispose to prolapse was statistically unrelated to eversion (Long and Rodriguez 1972). Our observations indicated that preputial eversion was significantly related to low sheath score and preputial eversion was most common in bulls with more pendulous sheaths (67% v 26%) (Table 58). This was in agreement with a study that found that sheath depth was significantly correlated with eversion score (Lagos and Fitzhugh 1970). If eversion of the prepuce is found to be an important factor in the pathogenesis of preputial prolapse, then evidence from the present study supports the culling of bulls with large sheaths because of the relationship between large sheaths and a higher preputial eversion score.

occurrence of preputial eversion							
	No eversion	Eversion					
	(n)	(n)					
Small sheath	14	5					
Large sheath	7	14					
	P < 0.05						

Table 58.	Relationship between sheath size and						
occurrence of preputial eversion							

P < 0.05

There have not been any observations published on the preputial retractor muscles in Bos indicus bulls (Hofmeyr 1987). Specifically, it was noted that there was nothing in the literature to confirm whether the preputial retractor muscles in Bos indicus bulls that are prone to, or are not prone to, preputial eversion or prolapse are different. Other authors had hypothesised that in Bos indicus bulls that are prone to preputial eversion or prolapse, there is an absence or incomplete development of the retractor muscles of the prepuce (Walker 1980) or that these muscles are underdeveloped with poor tone (Memon et al. 1988).

Eversion score in the present study was not significantly related to the size of the retractor prepuce muscle or to the size of the retractor prepuce muscle network (Table 59). This is different to the situation in Bos taurus bulls where eversion is seen more commonly in bulls that have a preputial retractor muscle deficiency (Long and Hignett 1970). Other authors also related this preputial retractor muscle deficiency to preputial prolapse and suggested that the predisposition to preputial prolapse involves breeds that have incomplete development or absence of the retractor prepuce muscle (Walker and Vaughan 1980: Wolfe 1986).

industrie network and the occurrence of preputation eversion					
	No eversion	Eversion			
	(n)	(n)			
Small network	11	9			
Large network	10	10			
	P = 0.75				

Table 59. Relationship between the size of the preputial retractor muscle network and the occurrence of preputial eversion

In a study on yearling bulls, no correlation between body weight and the preputial eversion score was found after an adjustment for age (Lagos and Fitzhugh 1970). Similar results were found in our study as preputial eversion was not significantly related to carcase weight. This would suggest that selection against eversion may not impact on the growth rate of Santa Gertrudis bulls.

Some authors have suggested that size of the prepuce may influence the probability of preputial prolapse (Arthur 1964). Some authors quantified size by measuring preputial length and they suspected that an increased preputial length is the major factor in prolapses (Hofmeyr 1968; Klug et al. 1979). Other authors stated that preputial length alone was not the cause of prolapse (Memon et al. 1988). Eversion score in this study was not significantly related to the detached prepuce weight or any of the detached preputial length measurements. This supports other information comparing preputial length measurements after slaughter that did not show a significant difference between bulls that displayed preputial eversion and those that did not (Long and Hignett 1970).

Eversion score was not significantly related to orifice diameter in the present study. In another study, no significant difference in orifice measurement between everting and non-everting bulls was found (Long and Hignett 1970). However, some authors (Van Den Berg 1984; St Jean 1995) indicated that orifice diameter was a contributing factor in prolapse, which appears to contradict this previous study and our results. A larger preputial orifice may be related to a higher risk of injury of the prepuce due to increased exposure of the prepuce at the orifice and not necessarily be related to prolapse due to eversion.

The size and weight of the penis was recorded to establish a range in normal young Santa Gertrudis bulls (Table 60). This range may be used as a reference by later studies to determine if penis size is a factor in preputial prolapse. Sheath skin measurements and scores were performed to confirm the relationship that has been suggested between skin composition and an increase in preputial eversion or preputial prolapse (Smit 1994). This was not supported by the present study, which found that eversion was not significantly related to any of the sheath skin measurements.

For the penis, its lateral diameter was significantly correlated (P < 0.05) with its dorso-ventral diameter, its weight and to each of the two preputial orifice-to-fornix measurements. The lateral diameter of the penis bore no significant relationship to the width or vertical thickness of the penile retractor muscle (r = 0.31, P = 0.055 and r = 0.31, P = 0.051 respectively). The weight of the penis bore a significant (r = 0.68, P < 0.001) relationship to its length. The diameter of the penis and the width and vertical thickness of the penile retractor muscles were not significantly related (P > 0.05) to the horned or polled state, the network score or to the width of the preputial retractor muscle. Although the size of the penis increased with increasing carcase weight (r = 0.33, P < 0.05), neither the penile length, nor either of its diameters were individually significantly correlated with carcase weight.

# 6.3.3.6 Conclusions

This study has recorded a range of measurements of the sheath and prepuce in young Santa Gertrudis bulls although care should be taken when interpreting results of this limited sample of bulls. Further studies of the pathology of the sheath and prepuce would complement this data. We concluded that there was no evidence that the polled bulls in this herd had poorer preputial muscle development than others of this herd or that they everted their prepuces further than horned bulls. This is different to the situation in polled *Bos taurus* bulls but may reflect the limited sample of bulls in this one herd. In this study preputial eversion was related to low sheath score, which may justify selection against *Bos indicus* bulls with pendulous sheaths if eversion leads to an increase in preputial injury or infection. Eversion of the prepuce was not related to the apparent degree of preputial retractor muscle development or to the muscle network score. Further work is needed to determine the reason for preputial eversion in these *Bos indicus* derived bulls.

	Mean	SD	Range <sup>a</sup>	Published measurements
Prepuce				
Preputial attachment to end of glans (mm)	121	10	100-142	٦
			62-137	Tip to orifice 548mm, Bos indicus (Bellenge
			97-263	J1971)
Fornix to attachment (mm)	99	19	156-360	
Preputial orifice to fornix (mm)	180	42	261-485	
Preputial orifice to fornix (mm) (detached)	268	46		
Total length (mm) (detached)	373	56		Total length of prepuce plus glans penis 548mm, <i>B indicus</i> (Bellenger 1971)
Weight (g)	179	51	77-281	
Muscles of the penis and prepuce				
Vertical thickness of penile retractor (mm)	9.8	1.9	6.1-13.5	
Width of penile retractor (mm)	16.2	2.9	10.4-22.0	
Width of preputial retractor (mm)	42	16	10-74	
Vertical thickness of preputial retractor (mm)	4.2	2.0	0.4-8.0	
Penis				
Dorso-vental diameter (mm)	30.8	1.7	27.4-34.2	
Lateral transverse diameter (mm)	35.2	1.8	31.5-38.9	40-50mm (Roberts 1971); 25-40mm (Trotter and Lumb 1958)
Length (mm)	1104	61	981-1229	1100mm, <i>B indicus</i> and crosses (Hofmeyr 1987); 900-1000mm (Bellenger 1971); 900mm, <i>B taurus</i> (Hofmeyr 1987); 900mm (Roberts 1971); 1109mm, <i>B indicus</i> (Bellenger 1971)
Weight (g)	880	90	700-1060	
Sheath skin				
Thickness at preputial orifice (mm)	9.7	1.7	6.3-13.1	
Umbilicus measurement (mm)	32	5	23-42	Up to 50mm (Bertram <i>et al.</i> 1997)
Thickness (mm)	6.7	1.6	3.6-9.9	
Area to weight ratio (sq mm/g)	97	15	68-127	
Other measurements				
Preputial orifice diameter (mm)	51	4	43-59	20mm (Sisson 1975); 20-40mm (Roberts 1971); 3 fingers, Santa Gertrudis (Hofmeyr 1987)
Hot carcase dressed weight (kg)	373	39	295-451	

#### Table 60. Mean, SD and range for various reproductive organs in 40 Santa Gertrudis bulls

<sup>a</sup> Range calculated as 2 standard deviations each side of the mean, normal distribution assumed

# 6.3.4 An anatomical study of chronic preputial prolapse in Santa Gertrudis bulls.

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# 6.3.4.1 Summary

The external reproductive organs of 32 Santa Gertrudis bulls with chronic preputial prolapse were examined after slaughter at a south-east Queensland abattoir. Hot standard carcase weights were collected from the abattoir and a history of the bull and farm of origin was obtained.

Polledness was significantly (P < 0.05) related to a deficiency in the size and development of the preputial retractor muscle. Data on the horned status of 23 of the bulls were available. There were eight polled bulls (homozygosity not known) and 15 horned bulls identified. This proportion of polled bulls (35%) is greater than the normal proportion of polled bulls seen in the Santa Gertrudis population. Most of the polled bulls (86%) were six years old or younger but more than half (53%) of the horned bulls were more than six years old. Polledness was not related to the size of either the penile retractor muscle or the penis. The weight, length and diameter of the penis and the width of the penile retractor muscle were significantly related to the carcase weight of the bulls.

Preputial prolapse in polled Santa Gertrudis bulls could be due to the significantly reduced size and development of the preputial retractor muscle complex in these bulls. No obvious cause of prolapse in the horned bulls was determined from the dissections.

# 6.3.4.2 Introduction

Preputial prolapse is perceived within the beef industry to be a cause of financial loss (Amaya Posada 1979). It has been suggested that *Bos indicus* and *Bos indicus* derived bulls should be culled for many reasons that are thought to be associated with preputial prolapse. This includes culling bulls with large sheaths or bulls in which preputial eversion regularly occurs (Donaldson and Aubrey 1960).

Previous studies with *Bos taurus* bulls showed that polled *Bos taurus* bulls have an absence in the preputial retractor muscle when compared to horned bulls. Of the prepuces from 30 *Bos taurus* bulls that were examined, all those from horned bulls had well developed preputial retractor muscles but these muscles were absent from all of the polled bulls (Long and Hignett 1970). In this present study, the anatomical features of the prepuces of 32 Santa Gertrudis bulls in which preputial prolapse had occurred were examined.

# 6.3.4.3 Aim

The aim of the experiment was to describe the anatomy of the sheath and prepuce of Santa Gertrudis bulls affected with chronic prolapse of the prepuce.

# 6.3.4.4 Materials and methods

#### Experimental animals and environment

The external reproductive organs, of 32 Santa Gertrudis bulls with chronic preputial prolapse that were slaughtered at a south east Queensland abattoir, were collected and hot standard carcase weights recorded. The bulls were from north-eastern Australia and were from 2 to 10 years old. In this study, 'old' bulls were classified as over 6 years of age. Eight of the bulls were polled and 15 were horned, but information was not available for the remaining 9 bulls. The organs were examined within 12 hours of

slaughter and were measured and photographed.

A history of the farm environment and the development of preputial prolapse on the farm was obtained from the owners and bull age was obtained from farm records.

#### Definitions

For this study, the prepuce is defined as the hairless epithelium extending from the preputial orifice to its attachment on the penis.

#### Measurements

#### Muscles of the penis and prepuce

1. Vertical thickness of penile retractor muscle (mm) – Dorso-ventral measurements of the muscle were taken with a ruler just posterior to the insertion of the muscle onto the penis.

2. *Width of penile retractor muscle* (mm) – The lateral transverse measurement was taken immediately posterior to the insertion of the muscle onto the penis.

3. *Vertical thickness of preputial retractor muscle* (mm) – This dorso-ventral measurement was taken midway along the body of the muscle.

4. *Width of the preputial retractor muscle* (mm) - The lateral transverse diameter of the preputial retractor muscle was measured midway between the orifice and the base of the scrotum.

5. *Preputial retractor muscle network* - The network was scored from one to 5; (1 = network absent or only 1 to 2 muscle strands from each muscle present, 2 = network of 3 muscle strands, 3 = network of 4 muscle strands, 4 = dense network of more than 4 muscle strands, 5 = broad network of more than 4 muscle strands). For the analysis, a 'small network' is defined as a network of 3 or less preputial retractor muscle strands visible on the dorsal surface surrounding the prepuce. A 'large network' has 4 or more strands.

#### Penis

1. Dorso-ventral diameter of penis (mm) and the horizontal transverse diameter of the penis (mm) – These were measured immediately proximal to the insertion of the retractor penis muscles.

2. *Penis length* (mm) - The suspended penis was measured from the tip of the penis to the distal boundary of the ischiocavernosus muscle.

3. Weight of penis (g) – For weighing, the penis was transected at the distal end of the ischiocavernosus muscle.

#### Sheath skin measurements

*Skin thickness* (mm) - The skin thickness was measured with callipers near the midpoint between the orifice and the base of the scrotum.

#### Statistical analysis

Due to low frequencies of the score values, each of the scores was collapsed to two-valued scores and then selected  $2 \times 2$  cross-tabulations were calculated from among the scores. Where all four frequencies in the margins of a  $2 \times 2$  table were greater than ten, the chi-square test of association was used but otherwise the Fisher-Irwin exact test tail probability was calculated.

For each of the measurements and for each of the scores, a one-way analysis of variance (ANOVA) was done in which the score (or collapsed score) values were taken to be treatment levels in a one-way

analysis of variance. Treatment significance was tested (F-test) at the 5% level. Relationships among abattoir measurements were examined by calculating a matrix of all possible pairwise simple correlation coefficients. The *Statistix* package (Analytical Software, Tallahassee FI) was used to compute tests of association, ANOVAs and correlations.

# 6.3.4.5 Results and Discussions

Complete data were not available for all of bulls so numbers between relationships in the analysis varied. Bulls ranged in age from 2 to 10 years old (mean 6.1, SD 2.1). Polled ( $819 \pm 88$  kg) and horned bulls ( $841 \pm 133$  kg) were of similar liveweights.

The prolapsed bulls could be separated into two main categories. One group consisted of horned bulls that were not deficient in preputial retractor muscles and the other group consisted of generally young polled bulls with a deficiency in the preputial retractor muscles (Table 61). This may be important as polledness is documented as being linked to preputial eversion in Bos taurus bulls (Long and Hignett 1970; Rice 1987; Bruner and Van Camp 1992). These polled bulls have an associated deficiency in the preputial retractor muscles. If there is a link between increased eversion and a higher incidence of preputial prolapse, this could explain the increased proportion of preputial prolapses in polled Bos taurus bulls. These polled Bos taurus breeds are predisposed to preputial injuries and preputial prolapse (Desrochers et al. 1995). The present study of Bos indicus derived bulls with preputial prolapses showed that there was a higher incidence of preputial prolapse in polled than in horned bulls as represented by the disproportionate percentage of prolapsed polled to prolapsed horned bulls. Eight bulls (35%) with prolapses were identified as polled and 15 (65%) were horned. This is compared to the industry figures of 5% of Santa Gertrudis bulls being polled when figures are taken from a semen price list (Beef Breeding Services 1999) and 15% in the 2002 progeny recording data from the Santa Gertrudis Breeders' (Australia) Association. The higher percentage in the more recent industry figures may be due to an increase in the popularity of polled Santa Gertrudis bulls.

			0	0
	Preputial retractor muscle network		А	ge
	Small	Large	$\leq$ 6 years old	> 6 years old
Horned bulls	1	11	7	8
Polled bulls	6	1	6	1
	P <	0.05	P = 0	.16

 Table 61. Relationship between horn status and preputial retractor muscle network and between horn status and bull age at slaughter

The increase in proportion of polled bulls prolapsing may be related to a deficiency in the preputial retractor muscle complex in these polled bulls. Polledness was significantly related to the preputial retractor muscle network and to the vertical thickness and width of the preputial retractor muscle (Tables 61 and 62). Six of the 7 prolapsed polled bulls were deficient in the preputial retractor network while only one of the 11 prolapsed horned bulls was deficient. Homozygosity of these polled bulls was not known. Deficient was defined as a preputial retractor muscle network of 3 or less obvious muscle strands.

The link between polled bulls and a deficiency of the preputial retractor muscle may not be universal in the *Bos indicus* derived breeds. From the evidence of an earlier study into the anatomy of the preputial retractor muscles in non-prolapsed bulls, it was concluded that there are lines of polled Santa Gertrudis bulls that do not have any preputial retractor muscle deficiencies (LB Turner, Section 6.3.3 this report).

Polledness was not related to the size of the penile retractor muscles or to the length, size or weight of the penis. There is no indication that the increased prevalence of preputial prolapse seen in polled bulls is due to larger penises or to a deficiency in the size of the penile retractor muscles. The reproductive organs of polled bulls without preputial prolapses were dissected in a previous study and a similar result

	Skin thickness (mm)		Depth of the preputial retractor muscle (mm)		Width of the preputial retract muscle (mm)	
	n	$\text{Mean}\pm\text{SD}$	n	$\text{Mean} \pm \text{SD}$	n	$\text{Mean}\pm\text{SD}$
Horned bulls	15	8.0 ± 1.5	15	$5.0\pm1.5$	11	$\textbf{46.4} \pm \textbf{21.3}$
Polled bulls	7	$\textbf{6.6} \pm \textbf{0.7}$	5	$\textbf{2.5}\pm\textbf{2.9}$	5	$\textbf{4.0} \pm \textbf{8.9}$
		P < 0.05		P < 0.05		P < 0.05
	Penil	e retractor muscle width (mm)	Height of penile retractor muscle (mm)		•	
	n	$\text{Mean}\pm\text{SD}$	n	$\text{Mean} \pm \text{SD}$	n	$\text{Mean}\pm\text{SD}$
Horned bulls	15	$20.0\pm5.4$	15	$10.8\pm3.6$	15	$\textbf{32.6} \pm \textbf{3.0}$
Polled bulls	8	$20.5 \pm 2.2$	8	$11.0\pm1.3$	8	$\textbf{32.8} \pm \textbf{2.1}$
		P = 0.8		P = 0.9		P = 0.9
	Hoi	rizontal diameter of the penis (mm)	F	Penis length (mm)	l.	Weight of penis (g)
	n	$\text{Mean}\pm\text{SD}$	n	$\text{Mean} \pm \text{SD}$	n	$\text{Mean}\pm\text{SD}$
Horned bulls	15	$\textbf{36.0} \pm \textbf{3.1}$	15	1152 ± 87	15	$1157\pm260$
Polled bulls	8	$34.8 \pm 2.8$	8	$1188 \pm 63$	8	$1158 \pm 169$
		P = 0.4		P = 0.3		P = 0.9

was found (LB Turner, Section 6.3.3 this report). Polledness in that previous study was also not related to the size of the penile retractor muscles or to the length, size or weight of the penis.

Table 62. Relationship between horn status and measurements of the preputial retractor muscle, the

Preputial prolapse was seen at a younger age in polled bulls compared with the horned bulls. Only one polled bull was classified as 'old' compared to more than half of the horned bulls. For the analysis, young was defined as  $\leq$  6 years. Six years old was the average age of the bulls in this study. Although this was not a statistically significant result, the data suggests that factors causing preputial prolapse in polled bulls generally affected younger bulls. This may be related to a congenital deficiency in the size and development of the preputial retractor muscle deficiency in these polled bulls. The age of the prolapsed bulls at the time of slaughter is indicative of the age of onset of the preputial prolapse as most bulls were sent to slaughter soon after developing preputial prolapse.

The size or weight of the penis has been considered by some in the cattle industry to be a possible factor in preputial prolapse with larger penises thought to predispose to preputial prolapses. Many of the organ measurements recorded in this study (Table 63) were significantly related to the weight of the bull. The hot standard carcase weight was significantly related to the weight of the penis (r = 0.65, P < 0.001), the length of the penis (r = 0.42, P < 0.05), the lateral transverse diameter (r = 0.57, P < 0.01) and the dorso-ventral diameter of the penis (r = 0.47, P < 0.05) and the width of the penile retractor muscle (r = 0.47, P < 0.05) with increasing weight of the penis and the increasing diameter of the penis.

Comparing the results from this study with the results from a previous study of normal bulls the penises of the bulls with preputial prolapses were thicker than the penises of the normal bulls (LB Turner, Section 6.3.3 this report). The bulls in the present study were 24% heavier than the normal bulls in the previous study and their penises were 31% heavier. However, this difference was not recorded in the dimensions of the penises, as the diameter and length of the penises in the present study were less than 5% greater than in the normal bulls of the previous study. This may be a reflection of a normal age effect rather than a possible factor in causing a preputial prolapse, as the bulls in this study averaged six years of age and the bulls in the previous study averaged three years old.

Trait	Mean	SD
Muscles of the penis and prepuce		
Height of penile retractor (mm)	10.9	± 2.6
Width of penile retractor (mm)	20.3	± 4.0
Width of preputial retractor (mm)	40.0	± 33.2
Depth of preputial retractor (mm)	4.0	± 2.2
Penis		
Dorso-ventral diameter (mm)	32.3	± 3.0
Lateral transverse diameter (mm)	36.3	± 3.2
Length (mm)	1160	± 80
Weight (g)	1150	± 220
Other traits		
Sheath skin thickness (mm)	7.3	± 1.5
Liveweight (kg)	825	± 118
Hot standard carcase weight (kg)	462	± 68

Table 63. Mean and SD of reproductive organ measurements in 32 Sa	anta
Gertrudis bulls with preputial prolapse	

The penile retractor muscle measurements were positively correlated with the penile measurements. No penile retractor muscle deficiency was found as the muscles increased in size in bulls with larger penises. These muscles were measured as they have been previously reported to be underdeveloped in Brahmans (Van Den Berg 1984).

Sheath skin measurements confirmed suggested relationships between skin composition and an increase in preputial prolapse (Baxter *et al.* 1989). Polled bulls in this study had significantly thinner skins than horned bulls but this may reflect the younger age of the polled bulls presented in this study.

A previous northern Australian abattoir survey examined preputial prolapse and found that most (76%) affected bulls were between 3.5 and 7 years old and 24% were more than seven years old (Mosaheb *et al.* 1973). These findings are very similar to the present study where 65% of the affected bulls were between 3.5 and 7 years old and 26% were more than seven years old.

The bulls with preputial prolapse in this study were either horned bulls with no preputial retractor muscle deficiency or young polled bulls with a significant preputial retractor muscle deficiency. The proportion of polled bulls (35%) is much greater than the normal proportion of polled bulls seen in the Santa Gertrudis population. This does not mean that polled bulls should not be used by the cattle industry. Evidence showing that populations of polled Santa Gertrudis bulls are available that do not have any preputial retractor muscle deficiency was provided by an earlier Queensland study (LB Turner, Section 6.3.3 this report). Polled Santa Gertrudis bulls in that study had an average preputial retractor muscle network that was more extensive than the network in the horned Santa Gertrudis bulls in this study.

# 6.3.4.6 Conclusions

Preputial prolapse in polled Santa Gertrudis bulls could be due to the significantly reduced size and development of the preputial retractor muscle complex in these bulls. No obvious cause of prolapse in the horned bulls was determined from the dissections. Further work is needed to investigate the cause of preputial prolapse in horned *Bos indicus* and *Bos indicus* derived bulls.

# 6.3.5 Relationship between the position of the penis and preputial eversion in *Bos indicus* derived bulls

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# 6.3.5.1 Summary

Twelve Droughtmaster bulls were examined and measurements of the position of the glans penis relative to the preputial orifice were recorded. Simultaneous measurements of the length of preputial eversion from the preputial orifice were also recorded. The relationship between the position of the glans penis and the length of preputial eversion was determined.

There was a significant constant linear relationship between length of preputial eversion and position of the penis in the sheath in all bulls studied. The length of preputial eversion, when the glans penis was at the orifice, varied significantly between bulls and this variation could not be fully explained by differences in factors such as farm, age, sire or whether the bull was horned, polled or scurred. The distance of the glans penis from the preputial orifice when preputial eversion was first observed was found to vary between the bulls. This distance was not significantly related to farm, age of bull, sire of bull or whether the bull was horned, polled or scurred. The length of preputial eversion when the glans penis was at the preputial orifice and the position of the glans penis when preputial eversion commences were not related to the distance from the scrotum to the preputial orifice or the vertical distance from the navel to the preputial orifice but were significantly related to the vertical distance from the ventral abdominal wall to the preputial orifice.

From this study we concluded that preputial eversion in these *Bos indicus* derived bulls was related to the position of the penis. The glans penis was found to constantly move within the sheath and was not found resting in any one position. Although this study measured only a limited number of bulls from two farms, these bulls displayed similar preputial eversion responses relative to the position of the glans penis. The position of the glans penis is a major factor in preputial eversion and may be more important than the presence or absence of the preputial retractor muscles.

# 6.3.5.2 Introduction

When the prepuce of a bull is everted, it is potentially exposed to many environmental factors, which could predispose to preputial prolapse. Although many authors suggest that eversion of the prepuce is a major predisposing factor in preputial injury and infection (Monke 1976; Larson 1986), there is a paucity of published information confirming this association.

Most reported cases of preputial prolapse requiring surgery have been in breeds known to display eversion of the prepuce (Monke 1976). This association was not universally observed and in a study of 244 bulls of 13 British breeds, health records revealed no greater incidence of preputial disease in bulls that displayed eversion of the prepuce than in those which did not (Long 1969).

Preputial eversion is considered to be related to a deficiency of the preputial retractor muscles in *Bos taurus* bulls (Long and Hignett 1970). Earlier anatomical studies in *Bos indicus* bulls (LB Turner Section 6.3.3 this report) indicated that preputial eversion might be related to position of the penis The present live animal study confirmed the effect of position of the glans penis on preputial eversion.

# 6.3.5.3 Aim

The aim of the experiment was to determine the relationship between the position of the penis and eversion of the prepuce in Droughtmaster bulls and examine factors that may affect this relationship.

# 6.3.5.4 Materials and methods

# Definitions

Sheath - The hair-covered skin appendage that supports and protects the penis along the ventral abdomen. It extends from the scrotum to the preputial orifice.

Prepuce - The hairless epithelium within the sheath that extends proximally from the preputial orifice and is reflected distally at the fornix before attaching to the penis.

Preputial eversion – Temporary protrusion of a variable length of prepuce from the preputial orifice.

### **Experimental animals**

Twelve young (11 to 24 months old) Droughtmaster bulls from 2 south-east Queensland properties were examined. Four of the bulls were polled, 3 were scurred (with horn-like growths in the same location as horns) and 5 were horned.

### Procedures

The bulls were examined while restrained in a cattle crush. The distance from the glans penis to the preputial orifice was measured externally with a flexible measuring tape and the glans penis was located visually where possible or by palpation. Simultaneous measurements of the length of the preputial eversion from the preputial orifice were recorded. As the position of the glans penis constantly changed, repeated measurements over a one hour period were recorded on each bull. Measurements were also recorded for some bulls during per rectal massage of the internal genitalia.

Measurements of the sheath were included to determine any relationship with preputial eversion. The measurements included the horizontal distance from the base of the scrotum to the preputial orifice, the vertical distance from the ventral abdominal wall to the preputial orifice and the vertical distance from the navel (umbilical scar) to the preputial orifice.

# Statistical analysis

All statistical analyses were performed using *Genstat* 5<sup>th</sup> Edition. Simple linear regressions were fitted for each bull to assess the relationship between the distance of the glans penis from the preputial orifice and the length of preputial eversion. These regressions were then compared using grouped regression analysis. There were significant differences between intercepts but not slopes. To assess whether the difference in intercepts could be attributed to the property (2 levels), sire (7 levels), horn status (3 levels) or age (2 levels,  $\leq$  16 months and > 16 months) the intercept term in the grouped regression was replaced by each of these factors in turn. Due to considerable confounding of these factors they could not be assessed simultaneously in the model. Correlations between the length of preputial eversion and other sheath measurements were calculated.

# 6.3.5.5 Results

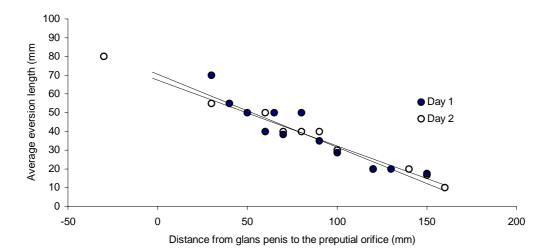
One bull did not evert his prepuce during the examination period and its data was excluded from the analysis. Regression analyses of the distance of the glans penis from the preputial orifice to the length of preputial eversion for data collected from bull A (Table 64, Figure 73) on 2 days, 3 weeks apart, were not significantly different (P < 0.05). Data for the 2 days for this bull were combined for further analysis. Repeated measurements for each of the other bulls were recorded on one day.

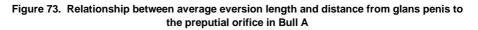
There was a linear relationship (P<0.001) between distance from the glans penis to the preputial orifice and the length of preputial eversion across all bulls with the bulls having significantly (P<0.001) different intercepts (Table 64, Figure 74). The difference between intercepts could not be fully explained by differences in factors such as farm, age, sire or whether the bulls were horned. There was evidence that some of the difference could be attributed to sires or age of bulls. The best model describing the results is one with a common slope and different intercepts.

	-	-	-						
Bull	Values#	Constant	Slope	Adj r <sup>2</sup>	Prob	Age (Months)	Property ##	Sire^	Horns <sup>^</sup>
А	50	68.7	-0.366	84.1	<0.001	16	1	1	Н
В	30	64.3	-0.382	85.8	<0.001	19	1	2	н
С	36	100.2	-0.382	86.7	<0.001	16	1	1	н
D	8	65.8	-0.341	64.3	<0.001	17	1	3	S
Е	14	67.6	-0.336	65.0	<0.001	16	1	3	Р
F	33	77.2	-0.398	93.2	<0.001	17	1	4	Р
G	21	72.7	-0.315	80.7	<0.001	13	2	5	Р
Н	17	68.4	-0.287	72.8	<0.001	11	2	6	S
Ι	17	74.2	-0.334	86.7	<0.001	11	2	6	н
J	21	70.9	-0.312	80.3	<0.001	24	2	5	S
к	23	81.8	-0.346	72.2	<0.001	22	2	7	Р

 Table 64. Number of preputial eversion measurements, relationship between preputial eversion and position of the glans penis, and physical characteristics of the bulls

#Values are defined as the number of measurements taken from that bull; ##property is identified by number; ^ Different sires were given different numbers; ^H horned, S scurred and P polled.





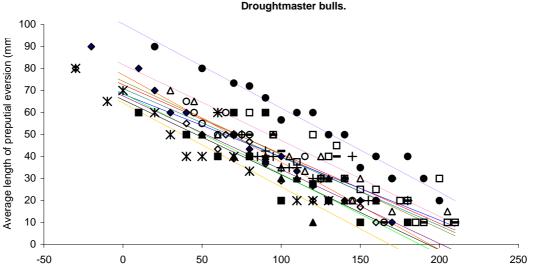


Figure 74. Regression of preputial eversion against position of the glans penis of 11 Droughtmaster bulls.

Distance between the glans penis and the preputial orifice (mm)

The distance of the glans penis from the preputial orifice when preputial eversion commenced was not related to the distance from the scrotum to the preputial orifice or the vertical distance from the navel to the preputial orifice. There was a significant positive relationship (P=0.04;  $r^2$ =0.38) between the distance of the glans penis from the preputial orifice when preputial eversion commenced and the vertical distance from the preputial orifice to the ventral abdominal wall (Figure 75).

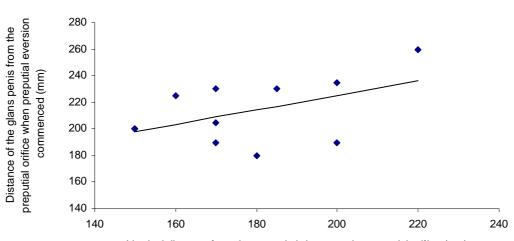
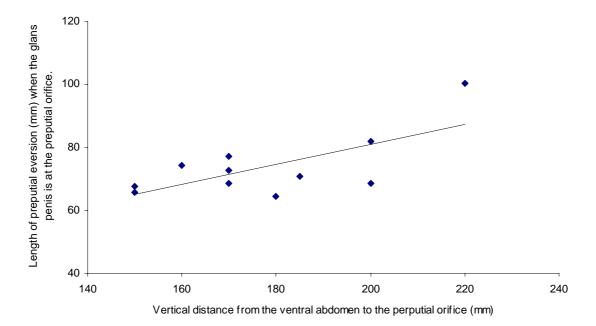


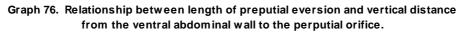
Figure 75. Relationship between position of the glans penis when eversion commenced and vertical distance from the ventral abdominal wall to the preputial orifice.

Vertical distance from the ventral abdomen to the preputial orifice (mm)

No relationship was found between the length of preputial eversion (when the glans penis was level with the preputial orifice) and the distance from the scrotum to the preputial orifice or the vertical distance from the navel to the preputial orifice. There was a significant positive relationship (P=0.04;  $r^2$ =0.38) between the vertical distance from the preputial orifice to the ventral abdominal wall and the length of preputial eversion when the glans penis was at the preputial orifice (Figure 76).

The distance of the head of the penis from the preputial orifice when eversion was first observed was found to vary significantly between bulls. This distance was not related to the age of the bull, the farm, the sire of the bull or whether the bull was polled, horned or scurred.





# 6.3.5.6 Discussion

In this study, the length of preputial eversion was significantly related to the distance of the glans penis from the preputial orifice. This finding was similar to other authors who examined 3 polled *Bos taurus* bulls and also found a correlation between movement of the penis and preputial eversion as preputial eversion accompanied forward movement of the glans penis (Long *et al.* 1970)

Age of the bulls was examined to determine if preputial eversion in relation to position of the glans penis changes with age. Any information related to age could be utilised by bull owners when considering culling of bulls. Although the effect of age of the bull was shown not to be significant, the range of ages of the bulls examined (11 to 24 months) may have been too small to detect a difference. Sires of the bulls were examined to determine if an effect of sire on the relationship between preputial eversion and position of the glans penis was detectable. No difference was noted between the sires. Seven different sires were represented in the 11 bulls analysed and too few progeny of each sire may have been examined to detect any difference. To examine the sire effect or age effect, further studies involving more representatives from each sire and a greater age range need to be completed. Whether the bulls were horned, polled or scurred was recorded (Table 65) in this study as the literature suggested that polled *Bos taurus* cattle have a deficiency of the preputial retractor muscle complex (Long and Hignett

1970). This has been linked to an increase in preputial eversion seen in these polled bulls. In a study of 244 bulls of 13 British breeds, 28 (85%) of 33 polled bulls exhibited preputial eversion whereas only 3 (1.4%) of 211 horned bulls did so (Long 1969). There seemed to be a clear relationship, in British breeds, between polled bulls and eversion of the prepuce. In this study, no difference was seen in the relationship between preputial eversion and the distance of the glans penis from the preputial orifice between polled, scurred or horned bulls. A similar result was obtained in the study of Santa Gertrudis bulls (LB Turner, see Section 6.3.4 this report) who showed that preputial eversion was not related to preputial retractor muscle development.

Horn status	Bulls (n)	Scrotum to preputial orifice (mm)	Ventral abdomen to preputial orifice (mm)	Umbilical scar to preputial orifice(mm)
Horned	4	$428\pm21$	$190\pm26$	$94\pm14$
Scurred	3	417 ± 55	$168 \pm 18$	$78\pm22$
Polled	4	$435\pm 66$	$173\pm21$	$70\pm17$

Table 65	Sheath measurements	(Means + SD)	) of bulls of different horn status
	oneath measurements	means ± 0D	

There was a significant relationship between the distance of the glans penis from the preputial orifice when preputial eversion commenced and the vertical distance from the preputial orifice to the abdomen (Figure 75). There was also a significant relationship between the length of preputial eversion when the glans penis is at the preputial orifice and the vertical distance from the preputial orifice to the abdomen (Figure 76). These relationships may be important as they confirm that a long or pendulous sheath may be a factor in preputial problems as stated by many authors (Monke 1976; Larson 1986). The relationship may not be significant in *Bos taurus* bulls as it was observed that pendulous sheaths may not necessarily lead to increased preputial eversion as Friesian bulls, which rarely exhibit eversion of the prepuce, generally have a more pendulous sheath than those of Angus bulls that exhibit preputial eversion more commonly (Long 1969).

One of the horned bulls examined did not exhibit eversion of the prepuce. The bull was able to constantly contract the preputial orifice towards the abdomen making the position of the penis in relation to the preputial orifice difficult to accurately assess. When the bull did relax his sheath during rectal examination, the glans penis was not observed to be close to the preputial orifice. This bull was excluded from the analysis as no preputial eversion was measured. Sheath measurements from this bull were within the range measured from the other bulls.

The relationship between the position of the glans penis in the sheath and the length of preputial eversion was the same for bull A for measurements taken on two separate days suggesting that this relationship is repeatable. Some measurements of the position of the glans penis in the sheath and length of preputial eversion were recorded during per rectal massage of the internal genitalia. Studies in Scotland suggested that the correlation between position of the glans penis and preputial eversion was similar for passive movement of the penis and for movement of the penis due to rectal massage (Long et al. 1970). Although some authors suggested that palpation might affect the position of the penis, the position of the glans penis in the present study was determined by palpation as simultaneous measurements of the length of preputial eversion were recorded so correlations could be accurately documented (Long et al. 1970). It was reported from preliminary studies that polled bulls do not retract the penis as far into the sheath as do horned bulls (Long and Hignett 1970). This was not confirmed in this study (Table 66). No significant difference was found in this study between polled bulls and horned bulls in the maximum distance the penis was withdrawn into the sheath. In the present study, the penis was found to be constantly moving and was not found resting at one position. This contrasts with reports of a constant position of the glans penis at rest (Long et al. 1970). The bulls in the present study were examined in a cattle crush and this may not reflect the paddock situation.

The position of the glans penis has been demonstrated in this study to be a major factor in preputial

eversion in *Bos indicus* derived bulls and may be more important than control of the prepuce by the preputial retractor muscles. This differs from *Bos taurus* bulls where the absence of the preputial retractor muscles is the major factor in preputial eversion. Polled *Bos taurus* bulls have an absence of the preputial retractor muscles and evert their prepuce while horned *Bos taurus* bulls do not have a preputial retractor muscle deficiency and generally do not evert the prepuce (Long and Hignett 1970).

Horn status	Bulls (n)	Average (mm) $\pm$ SD
Horned	4	$253\pm40$
Scurred	3	$313\pm6$
Polled	4	$280\pm50$

# Table 66. Maximum glans penis to preputial orificedistance recorded in the sheath

# 6.3.5.7 Conclusions

From this study we concluded that preputial eversion in these *Bos indicus* derived bulls was related to the position of the penis. The glans penis was found to constantly move within the sheath and was not found resting at any one position. Although this study measured only a limited number of bulls from two farms, these bulls displayed similar preputial eversion responses relative to the position of the glans penis. The position of the glans penis is a major factor in preputial eversion and may be more important than the presence or absence of the preputial retractor muscles.

# 6.4 Quantifying the role of sperm morphology as a criterion for bull selection and calf output

# 6.4.1 Background

Sperm morphology of bulls has been shown to be an important premating predictor of calf output of bulls in multiple-sire situations in *Bos indicus* and *Bos indicus* cross herds in northern Australia (Fitzpatrick *et al.* 2002; Holroyd *et al.* 2002a). These studies showed that in general, bulls with <50% normal sperm sired few calves whilst bulls with high calf outputs had >70% normal sperm (Fitzpatrick *et al.* 2002).

Whilst there have been many studies on bulls to determine the range of normal sperm in an ejaculate, there have been few studies to see if these values are repeatable in clinically normal bulls. If bulls are passed at a premating examination as being reproductively sound do they change in subsequent years? Initial studies in mature *Bos indicus* and *Bos indicus* cross bulls found that the repeatability of sperm motility and normal sperm was moderate and high respectively (Fitzpatrick *et al.* 2002). However, this needs to be validated under a range of seasonal conditions, genotypes and ages, particularly if greater emphasis is to be placed on morphology as a selection criterion for bulls.

Sub-fertility in bulls is expressed as both a reduction in the number of conceptions and a later date of conceptions in the mating period. This can have significant financial impacts by reducing weaning weights and lowering subsequent pregnancy rates. Sperm morphology appears to be one indicator of sub-fertility. Normal sperm appears to be poorly or inconsistently correlated with physical traits such as testicular tone and scrotal circumference and with other seminal traits such as mass activity and motility (Fitzpatrick *et al.* 2002). Bulls with low normal sperm can only be detected through laboratory assessment and not through palpation or a crush-side assessment of semen quality. Many producers are sceptical of laboratory assessments of semen quality such as sperm morphology. Thus there is a need to demonstrate that bulls with low normal sperm are sub-fertile i.e. reduced rates of conceptions and lowered pregnancy rates.

Research using single-sire mating at Swan's Lagoon in the 1980's showed that bulls with low levels of normal sperm usually had lower fertility (lower pregnancy rates) than bulls with at least 50% normal sperm (Holroyd *et al.* 1993). However, mating loads were generally low in this study as there were only about 20 cycling females per mating over the 3-month mating period. These studies need to be repeated to obtain sufficient data for reliable prediction of mating outcomes for bulls with low levels of normal sperm.

# 6.4.2 Longitudinal study on semen and sperm traits of bulls

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# 6.4.2.1 Summary

The experiment was conducted at Belmont Research Station in central Queensland. From 19 October 1999 to 05 November 2001, 5 Brahman (B), 32 Composite (C) and 5 Hereford-Shorthorn (HS) bulls were subjected to 9 physical and reproductive examinations and semen collected.

There was no effect of breed or time on semen mass activity. There was no effect of breed on semen motility, but there was a significant effect of time with no clear pattern of change. Although numbers of B bulls were small, they had significantly higher levels of normal sperm than C bulls with HS intermediate. There seemed to be no pattern in changes of normal sperm over time with maximum levels being recorded in November 1999 and minimum levels in April 2000 and August 2001. For abnormal heads, breed was not significant, but time was with no clear trend. C bulls had higher levels of abnormal midpieces than the other 2 breeds. Between bull variation was high in C bulls for cytoplasmic droplets, but breed means were reasonably constant over time. Repeatability of normal sperm was moderate (r =  $0.41 \pm 0.08$ ) with repeatabilities for semen and other abnormality categories being low.

The data set was not ideal for the estimation of changes in sperm morphology values and their repeatability because the bull population on Belmont was not stable. It was no possible to examine the same bulls repeatedly as a number of bulls were sold, or introduced during the experimental period.

# 6.4.2.2 Introduction

In clinically normal bulls, normal sperm ranged from about 20% to 90% with most bulls having greater than 50%. Normal sperm is one of the best pre-mating predictors of fertility as determined by calf output of bulls used in multiple-sire herds (Holroyd et al 2002a). Whilst there have been many studies done in bulls to determine the range of normal sperm in an ejaculate, there is little information on the repeatability on measures of semen and sperm traits

# 6.4.2.3 Aim

The aim of this experiment was to measure the repeatability of semen and sperm traits in clinically normal bulls of different ages and breeds.

# 6.4.2.4 Materials and methods

#### Experimental design

The experiment was an unbalanced longitudinal study of monthly changes in physical and reproductive traits of 3 breeds. The animal was the experimental unit.

#### Experimental sites and animals

The experiment was conducted at Belmont Research Station (23<sup>o</sup>S, 150<sup>o</sup>E) in sub-coastal central Queensland. Paddocks were flat open savannah woodland with improved (buffel, Rhodes, green panic) and native pastures (mainly black spear grass and blue grasses) with stocking rates of 1 AE to 2.5 ha.

The bulls were either born on Belmont or were introduced. The bulls were Brahman (B), Composites (C) or Hereford x Shorthorn (HS). The Belmont-bred Composites were mainly Belmont Red ( $\frac{1}{2}$  Africander,  $\frac{1}{4}$ 

Hereford <sup>1</sup>/<sub>4</sub> Shorthorn) or <sup>1</sup>/<sub>4</sub> Africander <sup>1</sup>/<sub>4</sub> Brahman <sup>1</sup>/<sub>4</sub> Hereford <sup>1</sup>/<sub>4</sub> Shorthorn. The introduced Composites were 37.5% Brahman, 25% Shorthorn, 25% Belmont Red and 12.5% Charolais.

#### Procedures

Forty-one bulls were first examined on 12 October 1999 with a further 8 bulls examined for the first time on 26 November 1999. Two bulls aged 10 years were excluded from the analysis, as they were substantially older than the rest of the bulls. The numbers in each age class of the 47 remaining bulls are presented in Table 67.

		, ,		,
Breed		Age class		
	<2 years	2 - 4.5 years	>4.5 years	Total
В	0	4 (2.9-4.1)	1 (5.0)	5
С	8 (1.9)	23 (2.0-4.0)	6 (4.8-5.1)	37
HS	0	5 (2.1-4.0)	0	5
Total	8	32	7	47

Table 67	. Number	of bulls by	age class	and breed	(range in bracke	ts)
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The number of times that bulls were examined varied. Table 68 summarises the number of observations by breed and date of examination for these 47 bulls, with the total number of observations being 194.

Table 68. Number of observations by breed and date				
Date	Breed			
	В	С	HS	
12 October 1999	5	30	5	
26 November 1999	5	37	4	
19 January 2000	3	16	2	
17 April 2000	5	10	4	
15 September 2000	5	8	2	
15 November 2000	3	7	3	
14 May 2001	4	4	4	
07 August 2001	3	6	4	
05 November 2001	4	7	4	

Table 68.	Number of	observations b	y breed and date
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Bulls were mustered to the yards by 0730, weighed (full) and then subjected to a physical and reproductive examination and semen collected for both crush-side and morphological assessment.

#### **Statistical analyses**

All statistical analyses were performed using GenStat for Windows 6<sup>th</sup> Edition. A linear mixed model was used to analyse sperm morphology traits recorded across time. An angular transformation was applied to abnormal midpieces, abnormal tails and cytoplasmic droplets prior to analysis to stabilise the variance.

The full model fitted was:

- Fixed effects: breed + age + time + breed.age + breed.time
- Random model: *bull+bull.time*

#### Where

- breed has 3 levels, B, C and HS,
- age is a covariate the age (years) of each bull on 12 October 1999,
- time has 9 levels,
- bull represents the variance component for bulls, and
- *bull.time* represents the residual variance

In all linear mixed models, random terms with zero variance components were removed from the model, then Wald tests were used to identify non-significant fixed terms (P>0.05) which were also removed to arrive at the final model, except that the main effects fo breed and time were always retained. Approximate least significant differences were calculated for significant (P<0.05) fixed effects.

Repeatability (intraclass correlation) was estimated as the ratio of variance components:-

Repeatability = bull/(bull + bull.time)

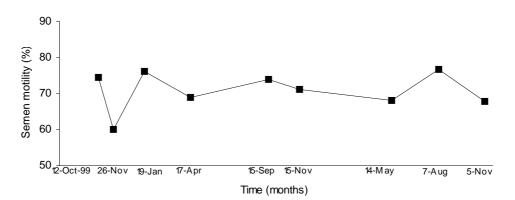
## 6.4.2.5 Results

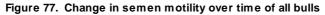
#### Mass activity

There was no significant effect of breed or time for mass activity. Predicted breed means were  $3.2 \pm 0.3$ ,  $2.8 \pm 0.2$  and  $2.3 \pm 0.3$  for B, C and HS respectively.

#### Motility

There was no significant effect of breed with predicted breed means being  $80.7 \pm 6.7$ ,  $69.5 \pm 3.3$  and  $62.0 \pm 6.9$  for B, C and HS respectively. There was a significant effect of time but there was no clear pattern of change (Figure 77). Semen motility was lowest in November 1999 and highest in January 2000 and August 2001.





#### Sperm morphology

#### Normal sperm

There were significant effects of breed (P=0.036) and time (P<0.001). B bulls had significantly higher levels of normal sperm than C bulls with HS intermediate (Table 69). There seemed to be no pattern in changes of normal sperm over time with highest levels being recorded in November 1999 and lowest levels in April 2000 and August 2001 (Table 69, Figure 78).

Breed	Bull observations	Normal sperm	Abnormal heads	Abnormal midpieces	Abnormal tails	Cytoplasmic droplets
	n	%	%	%	%	%
В	37	$67.9\pm4.7~\text{b}$	$20.2\pm3.1\text{a}$	12.7 (4.9) <sup>#</sup> ± 1.8 a	2.7 (0.2) <sup>#</sup> ± 0.7a	11.5 (4.0) <sup>#</sup> ± 2.3
С	125	$54.8\pm2.2~\text{a}$	26.5 ± 1.5a	17.4 (8.9) $\pm$ 0.9 b	3.1 (0.3) ± 0.4a	14.3 (6.1) ± 1.2
HS	32	$58.8\pm4.8~\text{ab}$	$\textbf{29.9} \pm \textbf{3.2a}$	12.7 (4.8) ± 1.8 a	3.0 (0.3) ± 0.7a	10.1 (3.0) ± 2.3

#### Table 69. Sperm categories by breed (mean ± se)

Means not followed by a letter in common are significantly different (P<0.05); # Back-transformed means presented

#### Sperm abnormalities

For abnormal heads, breed was not significant (P=0.076) but time was (P=0.003) with no clear pattern with highest levels in August 2001 and lowest levels in October 1999 (Tables 69 & 70, Figure 78). With abnormal mid-pieces, there was a significant effect of both breed (P=0.009) and time (P=0.015). C bulls had higher levels of abnormal midpieces than the other 2 breeds and values for abnormal midpieces of all bulls fluctuated between 4.0 and 9.8. Abnormal tails were very low (less than 1%) and as a consequence there was no effect of breed and time. Bull to bull variation was high in C bulls for cytoplasmic droplets but breed means were reasonably constant over time. In the final model, breed was not significant but time was (P=0.014), with no clear pattern.

Table 70.	Sperm	categories	by time
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Date	Bull observations	Normal sperm	Abnormal heads	Abnormal midpieces	Abnormal tails	Cytoplasmic droplets
	n	%	%	%	%	%
12-Oct-99	40	68.0 ± 3.9 d	20.0 ± 2.1 a	13.5 (5.5) <sup>#</sup> ± 1.2 ab	$2.4 (0.2)^{\#} \pm 0.7$	10.3 (3.2) <sup>#</sup> ± 1.6 ab
26-Nov-99	46	58.0 ± 2.9 ab	26.7 ± 2.1 bcd	14.8 (6.5) ± 1.2 ab	1.7 (0.1) ± 0.7	12.6 (4.7) ± 1.6 bc
19-Jan-00	21	66.4 ± 3.5 cd	21.8 ± 2.7 ab	15.5 (7.1) ± 1.5 abc	1.6 (0.1) ± 0.9	10.8 (3.5) ± 2.1 abc
17-Apr-00	19	53.0 ± 3.4 a	26.4 ± 2.6 bcd	18.3 (9.8) ± 1.5 c	4.2 (0.5) ± 0.9	15.4 (7.0) ± 2.1 c
15-Sep-00	15	59.8 ± 3.8 abc	22.7 ± 2.9 abc	12.5 (4.7)± 1.7 ab	3.3 (0.3) ± 1.1	15.6 (7.2) ± 2.3 c
15-Nov-00	13	66.3 ± 3.9 bcd	23.6 ± 3.1 abc	11.5 (4.0) ± 1.8 a	3.9 (0.5) ± 1.1	11.7 (4.1) ± 2.4 bc
14-May-01	12	57.5 ± 4.0 ab	26.8 ± 3.2 bcd	16.5 (8.1) ± 1.8 bc	2.6 (0.2) ± 1.2	12.3 (4.5) ± 2.5 bc
07-Aug-01	13	53.2 ± 3.9 a	31.9 ± 3.1 d	12.7 (4.8) ± 1.8 ab	2.7 (0.2) ± 1.1	13.3 (5.3) ± 2.4 bc
05-Nov-01	15	62.4 ± 3.7 bcd	29.4 ± 2.9 cd	13.1 (5.1) ± 1.7 ab	4.0 (0.5) ± 1.0	5.6 (0.9) ± 2.3 a

Means within columns not followed by a letter in common are significantly different (P<0.05); "Back transformed means in brackets

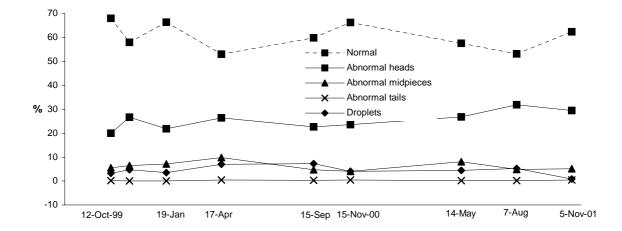


Figure 78. Changes in normal sperm (%) and in abnormal sperm categories (%) with time

#### Repeatability of sperm traits

Repeatability of normal sperm was moderate whilst semen mass activity and motility and sperm abnormality categories were low (Table 71).

Trait     Repeatability     Common       Mass activity     0.22 ± 0.08     Lo	
	w
Motility $0.30 \pm 0.08$ Lo	W
Normal sperm 0.41± 0.08 Mode	erate
Abnormal heads 0.27 ± 0.08 Lo	w
Abnormal midpieces 0.26 ± 0.08 Lo	W
Cytoplasmic droplets 0.22 ± 0.08 Lo	w

Table 71. Repeatability of semen and sperm traits

#### 6.4.2.6 Discussion

The data set was not ideal for the estimation of changes in sperm morphology values and their repeatability because the bull population on Belmont was not stable. It was not possible to examine the same bulls repeatedly over the 2 year experimental period as a number of bulls had to be sold after the second observation. As well, there were several introductions of Composite bulls during the experimental period. Nevertheless the data set has provided some useful information.

Overall B bulls had better quality semen, as indicated by higher levels of normal sperm, than C bulls. However the number of B bulls observed was only 5 so this result should be treated with caution. There appeared to be no seasonal effects on mass activity, motility or sperm morphology with changes in normal sperm and the various abnormality categories being very variable. Repeatability was moderate for normal sperm but low for semen traits and sperm abnormality categories.

# 6.4.3 Pregnancy rates achieved by mating bulls with different percentages of morphologically normal sperm

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## 6.4.3.1 Summary

Studies were conducted where bulls with varying percentage of normal sperm were mated for 3 months and pregnancy rate then recorded. These studies were carried out in a dry tropical environment at Swan's Lagoon Research Station using *Bos indicus* x *Bos taurus* cross cattle. Bulls were aged between 2 and 7 years. Bulls were mated as single-sires, apart from one group, with 15 to 50 females aged from 2 to 10 years, with most being non-lactating 2-year-old heifers.

In studies conducted in 1999 and 2000 where bulls were each mated to 46-52 two-year-old heifers, bulls with consistently more than 50% normal sperm pre-mating (fertile bulls) achieved higher pregnancy rates (P<0.001) than bulls with low percentages of normal sperm (sub-fertile bulls). In these studies, fertile bulls achieved an estimated 7-9 pregnancies per week during the first 3 weeks of mating, while sub-fertile bulls achieved between <1 and 5 pregnancies per week in the same period.

The 2002 study was carried out during very poor seasonal conditions, where despite 49-50 females being mated to each bull, fewer than 3 pregnancies per week were achieved by fertile bulls, reflecting low weights, and presumably low cyclicity rates, of the heifers. In this study, mating outcome for one bull with < 50% normal sperm both pre- and post-mating achieved the same pregnancy rate as his cohorts, as did another bull whose normal sperm increased from 14% to 82% over mating. However, within 2-year-old bulls, pre-mating normal sperm was positively related to pregnancy rate.

Across all studies, normal sperm post-mating was not consistently related to mating outcome.

Overall, the data support the conclusion derived from multiple-sire matings, that bulls with fewer than 50% normal sperm pre-mating are sub-fertile. The relationship appears clear when bulls are mated to high mating loads under average to good seasonal conditions, but can be variable when seasonal conditions are poor and mating loads are low.

## 6.4.3.2 Aim

The aim of the studies was to demonstrate that variations in conception patterns and pregnancy rates occurred in females when mated to bulls with varying levels of normal sperm.

## 6.4.3.3 Materials and methods

#### Location, cattle, and allocation

All studies were conducted at Swan's Lagoon Research Station in the dry tropics of north Queensland. The property is mostly flat open woodland savannahs with low-fertility duplex soils. A predominate species in the native pastures was *Heteropogon contortus*. All cattle used were 5/8 Brahman x *Bos taurus*, which was predominately Shorthorn.

On 29 January 1999 following a period of above-average rainfall, 2-year-old heifers were allocated by stratified randomisation on weight within condition score to 4 mating groups (Table 72) that were mated in adjacent paddocks (150-300 ha) for 88 days. Three bulls were mated as single sires to 46-48 heifers, and 2 bulls were mated together to 67 heifers.

In 2000 in an average-rainfall year, a similar study to the previous year was conducted (Table 72). However, heifers were allocated to 5 adjacent paddocks. Two bulls aged 3 years were allocated to 300

ha paddocks and three bulls aged 2 years were allocated to 150 ha paddocks. Within cohort, bulls either had high or low percentage normal sperm. Mating extended from 24 January 2000 for 93 days.

Bull ID	Bull age		oer of fers		Pre-m	ating			Post-r	nating		Paddock
				Wei	ght		dition ore	We	ight		dition ore	
	У	2 y	3 у	2 у	3 у	2 y	3 у	2 y	3 у	2 y	3 у	
1999 observation												
921306	7	46		437		7.0						ACWK
951134	4	48		440		7.2						DFWK
951084	4	47		426		7.2						GKWK
961203& 970019#	3&2	68		448		7.2						LG
2000 experiment												
960331	4	51		292		6.2		354		7.3		GKWK
960349	4	51		296		6.4		368		7.6		DFWK
961126	4	50		295		6.3		371		7.5		ACWK
970062	3	52		304		6.3		379		7.3		ST
970151	3	52		292		6.3		375		7.4		LG
2002 experiment												
980003	4	42	7	272	365	6.1	7.1	305	395	6.2	7.0	ABDL
990025	3	41	8	270	384	6.1	7.0	304	404	6.0	6.6	CDDL
991521	3	42	8	270	372	6.1	7.1	303	398	5.9	6.8	EFDL
991524	3	42	7	273	375	6.2	7.1	304	400	6.1	6.9	GHDL
58	2	42	8	270	374	6.1	7.1	311	408	6.1	7.0	AAS
75	2	42	8	270	386	6.1	7.3	305	403	6.1	6.9	BAS
107	2	41	8	269	384	6.1	7.1	310	418	6.2	7.0	CAS
1046	2	42	7	272	379	6.1	7.1	314	416	6.2	7.1	DAS

Table 72. Numbers and pre-mating and post-mating liveweights and body condition scores of heifers in the
various bull mating groups in 1999, 2000 and 2002

#: 2 bulls in one mating group

At the end of an extended dry period in 2002, 8 bulls (Table 72) were selected to each be mated to 49-50 non-lactating heifers aged 2 (85%) and 3 (15%) years. Heifers were allocated by stratified randomisation on weight within condition score and cohort. Mating was conducted in paddocks of approximately 100 ha, commencing on 13 March 2002 and continuing for 84 days. The design was 2 blocks of 4 paddocks. Within each block, bulls of similar age were used with a range of normal sperm from high to low being represented.

#### Measurements

The weight and condition score of females were recorded before and after each mating. At the end of each mating and 6-8 weeks later, females were pregnancy tested and foetal age estimated to calculate time of conception.

Prior to and after matings, all bulls were subjected to a BBSE with serving assessment not conducted. In the assessment of sperm morphology, abaxial tails, slight pyriform head shape, bent mid-pieces, segmental aplasia, and distal cytoplasmic droplets were all included as normal.

### Statistical analyses

#### 1999 observation

Statistical comparison was not possible due to confounding effects, ie, this study was an observation. However, cumulative pregnancy patterns were compared for the three bulls using a survival analysis and setting the multiple-sire group (high normal sperm) as the base-line hazard. The model included a mating group factor to correct for over-dispersion.

#### 2000 experiment

Cumulative pregnancy rate was analysed within bull cohort using the 1999 observation procedures.

#### 2002 experiment

Analysis of cumulative pregnancy rate was as for the 2000 experiment, though comparison was across all 8 mating groups. The following determined whether paddock differences contributed to conception rate differences:

- Weight and condition before and after mating, and changes over mating were analysed using a linear mixed model approach with fixed effects cohort (1999, 2000), paddock (8 levels) and their interaction and the random effect of heifer.
- To account for potential paddock differences, pregnancy at day 84 (0=non-pregnant, 1=pregnant) was analysed using a generalized linear model with binomial distribution and logit link. A series of models were investigated, some predictive and some explanatory. All models initially included paddock (8 levels), cohort (2 levels) and an additional term relating to weight or condition score and all possible 2-factor interactions (some interactions had missing cells and so were not included). The weight terms were factors based on 25 kg increments; weight change factors were 5 kg increments. Non-significant (P>0.05) terms were progressively removed from the full model while preserving marginality (ie any model containing the interaction between 2 terms, eg, A.B, must also contain the main effects A and B).
- Adjusted and simple pregnancy rates were compared.

#### 6.4.3.4 Results

#### 1999 observation

The bulls with low normal sperm had consistently demonstrated this trait over the previous year and before and after mating (Tables 73 & 74). At the end of mating, a semen sample could not be obtained from Bull # 951134 (samples were not collected from the multiple-sire group bulls).

Over mating, bulls with low normal sperm were sub-fertile, with significant (P<0.001) differences between the sub-fertile bulls (Table 75; Figure 79).

#### 2000 experiment

Semen quality was consistent, either very high or very low, from the start to completion of mating for four bulls (Tables 73 & 74).

Within both the 3- and 4-year-old bulls (numbers start with 97 and 96, respectively), bulls with lower normal sperm had lower cumulative pregnancy rates than the bull with consistently high normal sperm (P<0.05; Table 75; Figure 80).

Bull				Pre-ma	iting						Post-m	nating		
	Wt	Cond	Scrot	Tone	Semen	Mass	Motility	Wt	Cond	Scrot	Tone	Semen	Mass	Motility
	kg	1-9	cm	1-5	1-5	0-5	%	kg	1-9	cm	1-5	1-5	0-5	%
1999 obs	ervation													
921306	724	7	36.5	2		1	20	754	7	35.5	2		0	20
951134	776	7	40.0	3		1	10	865	7	39.5	3	Sample u	unable to b	e collected
951084	734	7	38.0	3		1	20	802	7	38.0	2		1	20
961203	550	7	34.0	3		1	80	640	6			No samp	le collecte	ł
970019	468	6	37.5	3		2	20	608	7			No samp	le collecte	ł
2000 exp	eriment													
960331	620	7	43.5	4		0	5	698	7	43.5	3		0	5
960349	565	6	35.5	4		1	80	616	7	35.5	3		3	80
961126	632	7	37.5	4		1	60	700	7	37.5	3		2	20
970062	546	6	33.5	3		2	70	662	7	34.5	3		5	90
970151	600	6	28.5	4		0	30	622	6	28.5	3		0	80
2002 exp	eriment													
980003		7	37.0	3	3	0	10	682	7	36.0	4	3	0	20
990025		7	38.5	3	4	3	80	672	6	37.0	4	3	1	70
991521		8	39.0	4	3	1	50	632	5	37.0	4	4	1	40
991524		7	36.0	3	3	0	40	602	6	34.5	3	3	2	50
58		7	34.0	3	3	1	40	450	7	33.5	3	5	4	70
75		7	32.0	3	2	0	30	514	6	31.0	3	4	2	70
107		7	33.0	3	3	1	20	432	7	31.5	3	3	1	40
1046		7	36.0	4	2	1	60	512	6	34.0	3	4	1	40

#### Table 73. Bull parameters pre- and post-mating, excluding sperm morphology

Table 74. Spermiogram of study bulls

Bull ID				Pre-mat	ing						F	ost-mat	ing			
	% Normal			%	Abnor	nal #			% Normal % Ab					onormal #		
		PD	MP	TH	Ру	KA	VT	SA		PD	MP	TH	Рy	KA	VT	SA
1999 obse	rvation															
921306	21	17	11	9	4	2	35	1	34	39	20	5	0	0	2	0
951134	13	69	6	5	4	1	2	0								
951084	14	5	26	34	0	1	19	1	9	7	12	23	2	1	46	0
961203	80	9	5	2	1	0	2	1								
970019	66	4	7	21	0	0	0	2								
2000 expe	riment															
960331	23	5	55	14	1	0	2	0	18	8	45	25	3	0	0	1
960349	91	0	5	0	2	0	2	0	86	0	12	0	1	1	0	0
961126	30	12	11	18	3	3	22	1	13	35	7	36	4	1	4	0
970062	90	3	4	0	1	0	1	1	93	1	3	0	2	0	1	0
970151	10	27	28	14	5	0	16	0	45	9	5	15	7	4	9	5
2002 expe	riment															
980003	69	6	15	5	2	0	3	0	48	2	18	30	0	0	2	0
990025	89	5	3	1	2	0	0	0	88	1	4	2	3	1	1	0
991521	14	31	33	15	2	1	4	0	82	5	9	3	1	0	0	0
991524	20	19	10	22	5	14	10	0	38	21	13	21	1	4	2	0
000058	80	5	8	6	1	0	0	0	91	0	5	2	0	2	0	0
000075	40	8	26	23	2	0	0	1	75	2	17	6	0	0	0	0
000107	32	13	23	28	2	0	2	0	78	8	9	4	1	0	0	0
001046	49	4	28	14	4	1	0	0	26	3	23	43	1	0	4	0

# PD: Proximal cytoplasmic droplets; MP: Mid-piece abnormalities; TH: Abnormal tails & loose heads; Py: Pyriform heads; KA: Knobbed acrosomes; VT: Vacuoles & Teratoids; SA: Swollen acrosomes

Bull ID	Average	e pregnant	Hazard ratio	95% confidence interval
	Weekly to Day 21	Per 21-day period##		
1999 observa	ation			
921306	3	20%	0.16 c	0.09 - 0.27
951084	1	9%	0.06 d	0.03 - 0.11
951134	5	33%	0.29 b	0.18 - 0.47
2 bulls	8	70%	1.00 a	
2000 experin	nent: 4-year-old bulls			
960331	3	17%	0.25 c	0.15 - 0.42
960349	9	55%	1.00 a	
961126	6	38%	0.55 b	0.35 - 0.87
2000 experin	nent: 3-year-old bulls			
970062	7	41%	1.00 a	
970151	<1	2%	0.04 b	0.01 - 0.12
2002 experin	nent			
980003	3	20%	0.55 ac#	0.29 - 1.04
990025	3	20%	0.53 ac	0.28 - 1.00
991521	2	17%	0.72 ac	0.40 - 1.23
991524	3	18%	0.73 ac	0.40 - 1.31
000058	3	23%	1.00 a	
001046	3	24%	0.96 a	0.55 - 1.68
000075	1	10%	0.43 c	0.22 - 0.82
000107	<1	1%	0.05 b	0.01 - 0.23

#### Table 75. Pregnancies rates and survival analyses of cumulative pregnancy rates

# Hazard ratios followed by a letter in common are not significantly different (P=0.05)

## Of females non-pregnant at the commencement of each 21-day period

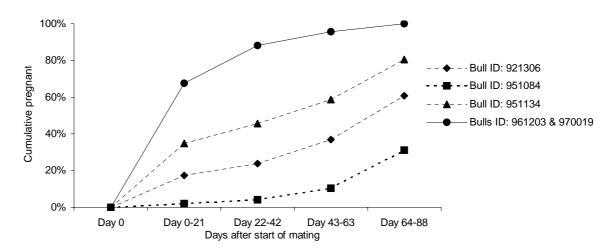
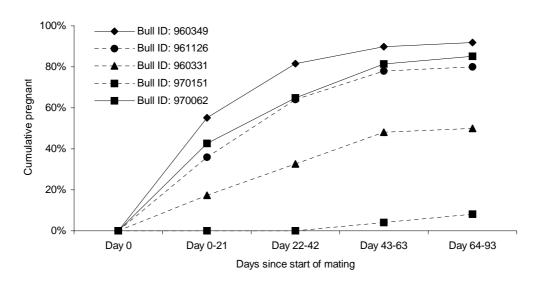


Figure 79. Cumulative pregnancy rates achieved by bulls mated in 1999



#### Figure 80. Cumulative pregnancy rates achieved by bulls mated in 2000

#### 2002 experiment

Poor seasonal conditions experienced prior to and during this experiment were reflected in loss of an average of one condition score and of 1.4 cm in average scrotal circumference (Table 73). Despite this, normal sperm was either maintained at pre-mating levels or increased in 6 of the 8 bulls; in 2 bulls the level decreased. In one bull, the normal sperm increased substantially over mating (14% to 82%).

Poor seasonal conditions prior to and during the trial resulted in relatively low weights and body condition scores of the females (Table 72, Figure 81); because of this, low cyclicity rates were expected. In an attempt to partially overcome this, mating was delayed by 2 months from the usual start of mating time (ie, from mid-January to mid-March), but seasonal conditions continued to be poor.

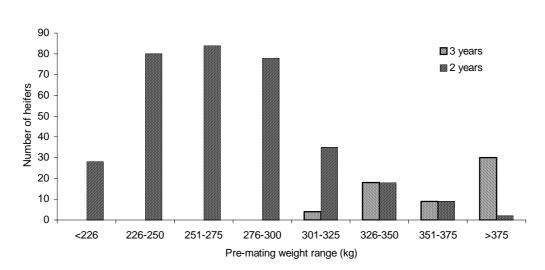


Figure 81. Distribution of female weights at the start of mating

A feature of the mating in 2002 was the low frequency of pregnancy (<1 pregnancy each 2 days), even at peak mating for the most fertile bulls (Table 75), because of the low weights, thus presumably low cyclicity rates of females. Cumulative pregnancy patterns did not differ significantly between mature bulls, despite one having consistently low normal sperm.

Two 2-year-old bulls achieved significantly (P<0.01) higher pregnancy rates than their two cohorts (Table 75, Figure 82). One of these (Bull ID 000058) had consistently high normal semen, and the other (Bull ID 001046) consistently low normal semen, though it was 49% at the start of mating. The normal sperm values in the two bulls with low pregnancy rates increased from below to above 50% over mating.

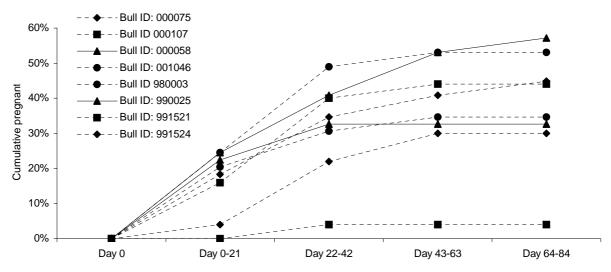
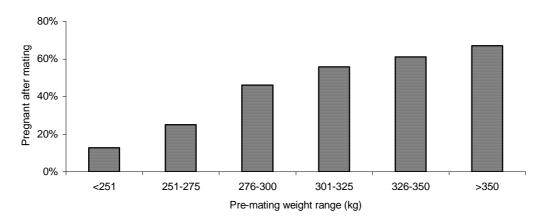


Figure 82. Cumulative pregnancy rates achieved by bulls mated in 2002

There were no differences between groups of heifers allocated to paddocks at the start of mating. However, by the end of mating there were differences, separate from the effects of bulls, indicated by the paddock differences for body condition score, average daily weight gain and body condition score change during mating. Pregnancy rate was higher in heifers heavier at the start of mating (Figure 83). Adjusting the paddock conception rates for growth parameters, eg, weights at the start or end of mating had little effect on paddock pregnancy rate differences, when compared with the simple pregnancy rates per paddock.





## 6.4.3.5 Discussion

The studies reported here were conducted under a range of seasonal conditions, which were reflected both in bull semen parameters and in mating loads available to bulls. Except for two bulls in the 2002 study, a consistent positive relationship appeared between normal sperm pre-mating, ie, bulls with fewer than 50% normal sperm were sub-fertile in the ensuing 3 months. Normal sperm post-mating was not consistently related to pregnancy rates. The relationship was less consistent under very poor nutritional conditions in 2002 that resulted in very low mating load for the bulls,.

Although there was evidence of nutritional differences between paddocks in 2002, these had little impact on the pregnancy rate of females as evidenced by the small difference between adjusted and unadjusted pregnancy rates. The conclusion is that differences in pregnancy rate between mating groups were due predominantly to bull differences or other unknown sources.

Hypotheses that may be considered for inconsistencies in relationship between normal sperm and mating outcome include: behavioural factors may contribute, especially in 2-year-old bulls that are sexually inexperienced; there may be factors in the semen of some bulls that contribute to either fertilisation failure and pregnancy failure, and are not related to sperm morphology or motility that currently are the prime indicators used to assess semen.

## 6.4.3.6 Conclusion

Overall, the data support the conclusion derived from multiple-sire matings, that bulls with fewer than 50% normal sperm pre-mating are sub-fertile in terms of impregnating cows.

## 6.5 The effect of herd dispersion and mating ratios on herd fertility

## 6.5.1 Background

In northern Australia, the majority of cattle graze at stocking rates of 1 adult to 5-30 ha. As a consequence cattle tend to be widely dispersed, although there tends to be concentrations of cattle at watering points. There is little scientific evidence to substantiate mating practices such as bull percentages for extensive herds although McCosker *et al.* (1989) indicated that there is considerable potential to reduce bull mating percentages. Reducing bull mating percentages will reduce the bull costs per calf, may reduce capital invested in bulls (Table 76), and will provide the opportunity to accelerate genetic improvement providing branding rates are not compromised.

Table 76. Significance of reducing mati	ng % throug	h use of bull breeding soundness evalu	uation
"Gulf country" station with previous manag	ement		
Females mated	16,500	Females per bull	25
Bulls onto station each year	185	Mating percentage	4.0%
Cost landed on station	\$3,000	Calves sired per bull per year	21
Veterinary cost: Year 1	\$0	Total calves sired in lifetime	85
Bulls culled after initial BBSE	0.0%	Bulls mated	657
Bulls culled after intermediate matings	2.5%	Total annual outlay on bulls	\$564,860
Annual husbandry costs	\$15	Bulls sold each year	165
Bull mortality/wastage rate	3.0%	Total salvage per bull sold	\$600
Seasons used	4	Net return on cull bulls	\$99,168
Branding rate	85%		
Peak mating period (months)	2.5	Av days/conception in peak mating	2.8
Calves sired in peak mating	85%	Annual bull cost/calf	\$33
Bull weight live @ salvage	600	Annual cost of bulls to station	\$465,692
Net \$/kg of bulls live @ salvage	\$1.00	Value of bulls mated	\$1,972,007
Potential scenario			
Females mated	16,500	Females per bull	33
Bulls onto station each year	165	Mating percentage	3.0%
Cost landed on station	\$3,000	Calves sired per bull per year	28
Veterinary cost: Year 1	\$20	Total calves sired in lifetime	113
Bulls culled after initial BSE	15.0%	Bulls mated	498
Bulls culled after intermediate matings	5.0%	Total annual outlay on bulls	\$505,775
Annual husbandry costs	\$15	Bulls sold each year	150
Bull mortality/wastage rate	3.0%	Total salvage per bull sold	\$600
Seasons used	4	Net return on cull bulls	\$90,030
Branding rate	85%		
Peak mating period (months)	2.5	Av days/conception in peak mating	2.1
Calves sired in peak mating	85%	Annual bull cost/calf	\$30
Bull weight live @ salvage	600	Annual cost of bulls to station	\$415,745
Net \$/kg of bulls live @ salvage	\$1.00	Value of bulls mated	\$1,494,994

There is enough evidence from both single- and multiple-sire results to suggest that bull mating percentages can be lowered below 5% without compromising herd fertility. Pexton *et al.* (1990) in Colorado found that, in Hereford and Angus bulls used as single sires, pregnancy rates of oestrus synchronised cows were not affected by bull-to-female mating ratios which ranged from 1:7 to 1:51. Bamualin *et al.* (1984), in a study of 27 years of mating records of Droughtmasters from Lansdown Research Station in north Queensland, showed that neither bull age or bull-to-cow mating ratios had any

effect on pregnancy rates. These results of Bamualin *et al.* (1984) were from both single- and multiplesire matings with bull-to-female ratios from 1:10 to 1:40. However the mating comparisons were not always contemporaneous but do suggest that bull mating percentages of 2.5% can be used without jeopardising herd fertility. The study by McCosker *et al.* (1989) at Mt Bundey in the Top End found that 3% tested bulls were superior to 5% untested bulls. McCool and Holroyd (1993) considered that there was scant evidence of lower bull percentages limiting herd fertility. In 20 years of investigating infertility in extensive herds, McCool (1993) found only 2 instances where low bull percentages could be suspected of causing lowered herd fertility. Fordyce *et al.* (2002) conducted observations on herd fertility in open savannah paddocks of 2000 ha with controlled waters at Swan's Lagoon in north Queensland. This study found that reducing bull percentages from 3.5% to 2.5% did not delay time of conceptions. Other work by Fordyce *et al.* (2002) in north-west Queensland compared bull mating percentages of 2% and 6% in paddocks ranging from 60 to 84 square km where waters were controlled. There was no difference in conception times although pregnancy rates were lower in the 2% group, this being attributed to poorer nutritional conditions in that paddock.

There were several other significant findings in these studies by Fordyce *et al.* (2002). Firstly, the variation between bulls in calf output, measured by DNA typing, was substantially lower when fewer bulls were used in the herd. Secondly, there was less bull wastage due to fighting and injury with lower bull percentages, and thirdly, the data refuted the perception that bulls with greater calf output ("harder – working") finished the mating season in poorer condition as there was no relationship between calf output and bull body condition.

The conclusions of Fordyce *et al.* (2002) were that a mating percentage of 2.5% reproductively-sound bulls is adequate for *Bos indicus* and *Bos indicus* derived cattle under conditions of low to moderate dispersion. However different bull percentages need to be to validated under conditions of different herd dispersion, particularly high dispersion created by topography, availability of water and pasture before we have confidence in recommending these low bull mating ratios.

Herd dispersion refers to the way that cattle spread out over their paddocks. Dispersion is modified by stocking rate and social behaviour of cattle but availability of water and to a lesser extent pasture quality and quantity are the major factors that influence dispersion. In paddocks where controlled waters such as dams or bores are the only water supply, then cattle have a common congregation point whilst the converse applies where there are large amounts of surface water. The location of cows in oestrus by bulls is critical for mating success particularly under conditions of high dispersion. It is a moot point whether bulls locate cows in oestrus or that the reverse applies. The work of Blockey (1976a) in southern Australia with *Bos taurus* cattle would suggest that cows in oestrus form a sexually active group and that bulls locate these females through cow mounting activity rather than by olfaction. However there appears to be little work done on the sexual behaviour of cattle under range conditions.

Based on normal levels of sperm production and normal sperm, and on likely sexual behaviour, it is expected that under low dispersion conditions in north Australia bull mating percentages of less than 1% might result in delayed times of conception. However, there is no data to support this.

## 6.5.2 Herd dispersion and mating ratio effects on conception patterns

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## 6.5.2.1 Summary

Four observations were conducted on north Queensland cattle stations that represented low, moderate, and high dispersion of cattle. Cumulative pregnancy rates as a function of bull:female mating percentages (low, <2%; moderate, 2-4%; and high, >4%) were assessed using Brahman and Brahman cross cattle. Except for some bulls used in the Canobie observation, all bulls passed a bull breeding soundness examination (BBSE), excluding serving assessment, prior to mating. Cumulative pregnancy rates of females were estimated by foetal ageing at pregnancy testing at the end of mating.

At Canobie Station, which had high dispersion of cattle, a high (4%) mating percentage (150 bulls mated to 3,766 heifers in 316 km<sup>2</sup>) was compared to a moderate (2.0%) mating percentage (10 bulls mated to 500 heifers in 103 km<sup>2</sup>). Flooding rains, resulting in hordes of savagely-biting sandflies during the peak conception period caused cattle to congregate in large groups with a median size of 200-300 animals, but these groups had dispersed into smaller groups with a median size of 30-50 animals by February 2001. The cumulative pregnancy rate was lower in the paddock with fewer bulls; the small difference occurred over 2 months of mating (December and January) when there was an estimated average of one pregnancy per bull every 6 days in both paddocks. This small difference was sustained for the following 3 months during a period when an average of one pregnancy was being achieved per bull each 2-3 weeks.

Observations conducted in consecutive years at Bow Park Station (low dispersion) compared low (1.2-1.4 %) mating percentages with moderate (2.7%) mating percentages in groups of between 170 and 220 maiden 2-year-old heifers grazing paddocks of approximately 2,000 ha. No differences in cumulative pregnancy rates occurred in 2001, but in 2002, lower rates occurred with the moderate mating percentage despite slightly heavier heifers being mated in the moderate mating percentage herd. Bulls caused on average 1 pregnancy each 1 or 2 days in the first 6-10 weeks of mating at low and moderate mating percentages, respectively. Aerial inspections of the cattle during the 2001 observation confirmed low dispersion in both paddocks.

An observation at Swan's Lagoon (moderate dispersion) of three groups of rebreeding cows (n=137 to 462) mated with either low (1.2-1.5%) mating percentage or moderate (2.4%) mating percentage demonstrated no effect on cumulative pregnancy rate attributable to mating percentage. One paddock with low stocking rate, and cows in better body condition, in which a low mating percentage was used, had the highest cumulative pregnancy rates. There was an average of 2 (1.5% bulls), 2 (1.2% bulls), and 5 (2.4% bulls) days per pregnancy per bull in the first 3 months of mating (February to April). Behavioural observations showed that bulls in the low mating percentage herds had higher movement ranges. In all paddocks, most bulls were generally found in the company of cows, but about half were also in the company of other bulls.

These studies showed that pregnancy rates were not reduced as a direct result of reducing bull percentages from high to moderate under high dispersion grazing management in north Australia, or from moderate to low under low or moderate herd dispersion. This provides further support to the recommendation that mating percentages of 2.5% reproductively-sound bulls (moderate % bulls) are adequate under most north Australian cattle management conditions.

## 6.5.2.2 Aims

The aim of the studies was to assess if conceptions are delayed when bull mating percentages (bulls:cycling females) were reduced from high (> 4%) to moderate (2-4%) under high dispersion grazing management in north Australia, or from moderate to low (<2%) under low or moderate herd dispersion.

## 6.5.2.3 Materials and methods

#### **Canobie observation**

#### Location, animals and allocation

Canobie Station is located in the Gulf region of north-west Queensland, approximately 200 km south of the Gulf of Carpentaria. The station is mostly flat, with small rises in some areas. The paddocks used were predominately Downs with some forest, traversed by large rivers that were up to 3 km wide because of multiple channels. The available pasture generally provides moderate-quality nutrition during the wet season (usually December to May), and poor nutrition during the dry season (usually June to November). The adjacent paddocks used (Figure 84) were very large; eg, the largest paddock used was 35 km in length. During the mating period (wet season), the rivers flooded, causing cattle to be isolated from each other. During the wet season, river flows prevented access to the study paddocks, except by helicopter. The paddocks were expected to provide similar nutrition and cattle dispersion.

HAWKES NEST DUFFERS NEST BORE SANDY BORE TEN MILE BI RGUMEN ARDINERS NEST MEST BOTTOM GARDINERS CANOBIE W.H RIVER CHERN'S NEST ALLE CANOBIE SPELL No 10 BORE TOP FUND CANOBIE BORE RIVER STANLEY NEST CORELL CORELLA BORE STANLEY STANLEY WATERHOLE No 11 BORE STUD BEECHAMPS BORE BLOODWOOD BORE PADDOCK

Figure 84. Paddocks used in the Canobie observation: Ten Mile & Top Canobie River (316 km<sup>2</sup>); Stanley (103 km<sup>2</sup>)

Two-year-old Brahman heifers weighing in excess of 240 kg were selected for mating in late 2000. It was presumed these heifers were non-pregnant, as bull control was not absolute. Mating commenced on 26 October 2000. Bulls remained with the heifers until mustering and pregnancy diagnosis in early June 2001.

Comparison in this high-dispersion situation was of:

- High mating percentage (4% bulls) where 3,766 heifers were mated with 150 bulls in Top Canobie River and Ten Mile paddocks (Figure 84).
- Moderate mating percentage (2%) where 500 contemporary randomly-selected heifers were mated to 10 bulls in Stanley paddock; 500 steers were also in the paddock. At the end of mating, 3 extra bulls were found to have moved from the high mating percentage paddocks to this paddock.

The bulls used in this study were 2-year-old Brahman x Red Angus (n=104) and randomly selected untested Brahman bulls (n=56) available on the station. The Brahman x Red Angus bulls were selected from 139 that had been relocated from central-western Queensland to Canobie in mid-September 2000. Prior to relocation their average weight had been 540 kg. In October 2000, these Brahman x Red Angus bulls were subjected to a bull breeding soundness evaluation (BBSE) of physical, semen, and sperm morphology traits and data on 104 that were mated in the observation are presented in Table 77. Not all bulls used in this study passed the BBSE.

Mating percentage	Condition. score	Testicular tone	Testicular tone Scrotal circumference		Normal sperm
	(1-9)	(1-5)	(cm)	%	%
Moderate (n=10 bulls)	5.1 (4-6)	3.3(3-4)	34.7 (31.5-38.0)	39 (10-70)	81 (68-96)
High (n=94 bulls)	5.1 (4-6)	3.3 (2-4)	34.2 (28.0-41.0)	48 (0-80)	75 (50-98)

Table 77. Means and ranges of physical and reproductive traits of Brahman x Red Angus bulls selected for the Canobie observation

#### Measurements

Dispersion of the cattle during mating was assessed twice by aerial reconnaissance using GPS. This took approximately 15 minutes in the small paddock and 100 minutes in the large paddock. The cattle had previous experience of being mustered by aircraft. Observations were conducted from 1,000 ft and caused some disturbance to cattle, thus there was some overestimation of grazing group size and underestimation of dispersion.

In early June 2001, the cattle were mustered, rectally palpated and foetal age estimated. From this data, dates of conception were estimated, allowing an estimation of cumulative pregnancy rate in each paddock over mating.

#### Bow Park observations

#### Location, animals and allocation

Bow Park is located 100 km north of Julia Creek in the lower Gulf country of north Queensland. The topography is flat and the trial paddocks were open, tree-less, fertile black-soil downs watered by bore drains. Nutrition derived from the pasture was generally moderate to high throughout the year; however nutritive value was limited in dry periods or in floods that cover some of the paddocks during the wet

season. Our observations commenced after heavy flooding followed an extended dry season. In the second year, rainfall was below average, and this pattern continued through the study period. Despite this, all cattle appeared to have moderate to good nutrition throughout the study period.

The females used were maiden 2-year-old Beefmasters (Brahman crossbred selected on performance). All bulls and heifers used were bred on the station. All bulls had passed a BBSE including physical, semen and sperm morphology, but excluding serving assessment (Table 78).

Date	Weight	Condition. score	Testicular tone	Scrotal circumference	Sperm motility	Normal sperm
	(kg)	(1-9)	(1-5)	(cm)	%	%
15-Jan-01	441 (386-510)	6.7 (6-7)	3.4 (3-4)	34.1 (31.5-36.5)	53 (10-90)	79 (61-98)
11-Apr-01		6.4 (6-7)	3.1 (3-4)	33.5 (31.5-36.0)	32 (0-80)	
22-Jan-02	399 (392-408)	6.0 (6-6)	3.2 (3-4)	33.4 (31.0-35.5)	73 (50-90)	56 (39-70)
9-May-02		5.8 (5-7)	3.0 (3-3)	32.8 (30.5-34.0)	58 (30-80)	

 Table 78. Means and ranges of physical and reproductive traits of bulls mated at Bow Park

In 2001, the comparison using two adjacent paddocks that were approximately 4 km square in size in this low-dispersion situation was of:

- Low mating percentage (1.4% bulls) where 3 bulls were mated to 220 heifers in Frontage paddock (1888 ha),
- Moderate mating percentage (2.7% bulls) where 5 bulls were mated to 183 heifers in Wingera paddock (1790 ha).

An estimate of pre-mating weight of the heifers in Frontage was 420 kg, 20 kg heavier than those in Wingera. Mating extended from 15 January 2000 to 11 April 2001. Four weeks into mating, one bull in Wingera paddock (moderate mating percentage) was accidentally hit by a vehicle, and was replaced immediately. In the third month of mating a stray bull was found in Frontage (low mating percentage).

In 2002, the comparisons conducted were:

- Low mating percentage (1.2 % bulls) where 2 bulls were mated to 170 heifers in Wingera paddock (1790 ha)
- Moderate mating percentage (2.3% bulls) where 4 bulls were mated to 175 heifers in Downs paddock (2570 ha)

The paddocks used each had heifers of different average weight (Table 79); however all were heavy and in forward condition and expected to have similar fertility. Prior to mating, real-time ultrasound assessment of ovarian function was conducted on a random group of 60 of the Downs heifers.

All bulls used in the study were 2 years of age and passed a BBSE (excluding serving assessment) (Table 79). Mating commenced on 22 January 2002 and continued to 09 May 2002 when pregnancy diagnosis was conducted. During mating, one bull in Wingera disappeared, but a second bull which had previously been assessed as sound, replaced it.

#### Measurements

Dispersion of the cattle during mating was assessed on 23 February during the 2001 mating by aerial reconnaissance and GPS. This took approximately 10 minutes for both paddocks. The cattle had

previous experience of mustering by aircraft. Observations were conducted from 1,000 ft, and caused some disturbance to cattle, thus there was some overestimation of grazing group size and underestimation of dispersion.

	Low % bulls	Moderate % bulls
Paddock	Wingera	Downs
Area (ha)	1790	2570
Bulls	2	4
Heifers	170	175
Heifers pre-mating		
Weight (kg; visual estimate)	400 (300-500)	400 (300-500)
Condition score(1-9)	7.4	7.2
Heifers post- mating		
Weight (kg; visual estimate)	450 (350-550; most 400-500)	420 (340-500; most 380-470)
Condition score (1-9)	7.4	7.2

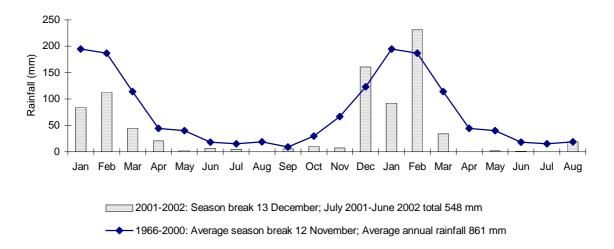
Table 79.	Paddock size,	cattle numbers a	nd estimated	pre- and	post- mating	liveweight and co	ondition
scores of	heifers at the 2	002 mating at Bov	v Park				

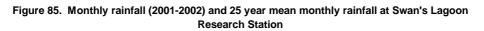
Pregnancy diagnosis and foetal ageing was conducted at the end of mating. During this procedure in 2001, a gate was accidentally opened between the two groups resulting in a mixed group of predominantly heifers from the moderate mating percentage group. These animals were excluded from the results.

#### Swan's Lagoon observation

#### Location, animals and allocation

The vegetation at Swan's Lagoon is open eucalypt savannah woodland with native pasture. Low-fertility duplex soils predominate on this relatively flat area of the station. Rainfall was below average over the study period with a late break to the dry season, followed by a short wet season (Figure 85.)





The mating comparisons were:

- Low mating percentage (1.5% bulls) where 2 bulls were mated to 137 cows in BNSE paddock of 910 ha (low stocking rate of 1 AE per 6.5 ha) (Figure 86),
- Low mating percentage (1.1% bulls) where 3 bulls were mated to 276 cows in HRYD paddock of 950 ha (high stocking rate of 1 AE to 3.4 ha),
- Moderate mating percentage (2.4 % bulls) where 11 bulls were mated to 462 cows in SDWE paddock of 2160 ha (moderate stocking rate of 1 AE to 4.6 ha).

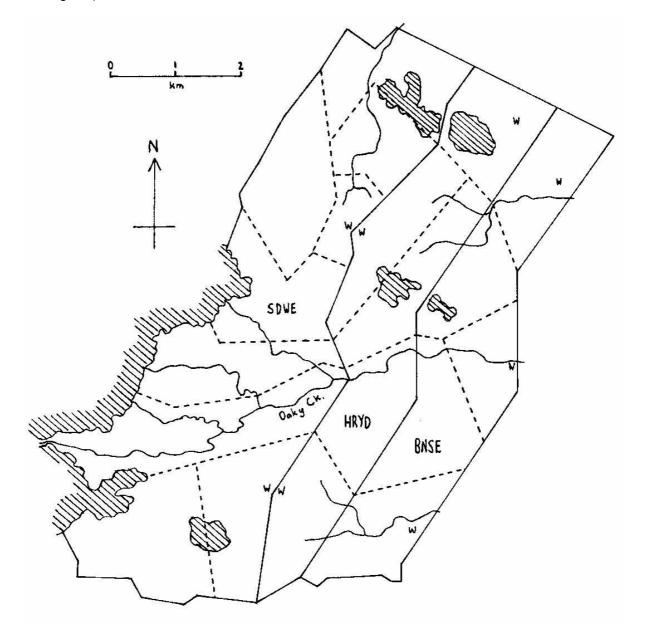
In each paddock the cow herd comprised 5 age groups of the same proportions with 22%, 23%, 21%, 18%, and 16% being 4, 5, 6, 7 and 8 years of age, respectively.

Bulls used were 2, 3, 4 and 5 years and average weight was 380, 518, 641, and 674 kg pre-mating, respectively (Table 80). All had passed a BBSE, including physical, semen and sperm morphology assessment, but excluding serving assessment. The bulls were allocated so a range of ages were represented in each paddock.

	Weight	Condition score	Testicular tone	Scrotal circumference	Sperm motility	Norm	al sperm	ı (%)
	(kg)	(1-9)	(1-5)	(cm)	%	Average	50%+	70%+
Jan 02	553	5.9	3.3	36.0	69	73	94	69
	(380-703)	(5-7)	(3-4)	(29.0-44.0)	(30-90)	(49-87)		
May 02	641	6.7	3.9	35.2	65			
	(458-778)	(6-7)	(3-4)	(29.5-41.0)	(30-90)			
Sep 02	583	5.9	3.1	34.3	65	70	92	67
	(453-718)	(5-7)	(3-4)	(28.0-42.0)	(30-90)	(15-87)		

#### Table 80. Means and ranges of physical and reproductive traits of bulls at Swan's Lagoon

Figure 86. Swan's Lagoon mating percentage observation site (W" indicates trough water; dotted lines indicate arbitrary divisions for cattle observations; hatched areas indicate steep hills that cattle tend not to graze)



Mating commenced on 19 January 2002 and continued to early September. In early May over a oneweek period, weaning of all calves over 100 kg was carried out. At this time, 30% of cows were removed from the paddock (60% of these were the oldest age group) and a new young age group introduced. Some cows were also transferred between paddocks to achieve even stocking rates across the whole trial area.

#### Measurements

At the May and September musters in both 2001 and 2002, weight and condition score of all cows was recorded, as well as lactation status and stage of pregnancy to estimate time of conception, thus cumulative pregnancy rates, during the 2002 mating.

Each 2-4 weeks between the start of mating and the May weaning muster, and thereafter at 3-7 week intervals, the location and behaviour of each bull was recorded, as was the size and location of each cow group. These observations were conducted mid-morning on each date. A movement range index (MRI) of between 0 (random) and 1 (always observed within the same 250 ha area) was calculated for each bull in the following manner. In SDWE, the 9 areas were ranked from that where a bull was found most frequently to that where he was found least frequently. The most-frequently occupied area was allocated a value of 1, the least-frequently occupied area a value of -1. The allocated values for intermediate ranking were calculated as 1 - (Ranking - 1) x Range of allocated values / (Number of levels - 1); eg, for the third ranked area the allocated value were multiplied. The MRI was then calculated as the sum of these products divided by the total number of observations.

## **Statistical analyses**

The Canobie and Bow Park studies were observations. Data collected at these sites were only summarised rather than compared statistically.

For the Swan's Lagoon observation, mating outcome (0 = not pregnant, 1 = pregnant) variables were analysed using a generalised linear model with binomial distribution and logit link. For each analysis, the main effects of paddock (n = 3), cohort (5 age groups), reproductive status in August 2001 (5 levels: nonlactating x 3 trimesters of pregnancy; non-pregnant; lactating and pregnant), and either weight (factor using 25-kg increments) or body condition score (5 levels: 2-3, 4, 5, 6, 7-8) in Aug 01 and or May 02 were included as well as two-factor interactions which did not include paddock. Higher order interactions were not included since there were generally many missing cells. The two points selected for analyses were 42 days into mating (2 reproductive cycles), and 96 days into mating (May 2001 at weaning). The full model was fitted initially and non-significant interactions progressively removed to arrive at the final model.

## 6.5.2.4 Results

## Canobie observation

#### Herd dispersion

On 08 January 2001, 450 mm of recent rain resulted in flooded rivers and plains. It also resulted in hordes of savagely-biting sandflies that persisted for weeks. These sandflies caused cattle to move constantly and hold together in tight groups, ie, there were a small number of large groups in both paddocks (Figure 87). Median group size was 200-300 animals.

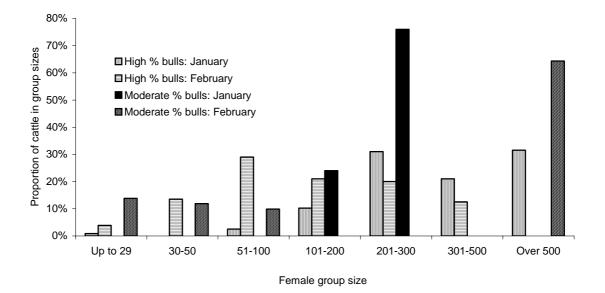


Figure 87. Distribution of cattle as indicated by group sizes during the 2001 observations at Canobie

At the next inspection on 12 February 2001, there had been a little more rain, and there were fewer sandflies. Median group size was 30-50 animals; in the small paddock, the aircraft caused about 2/3rds of the herd to form one group, though observations from a distance indicated these had mostly been in groups of median size prior to this occurring.

#### Mating outcome

For pregnancy diagnosis, 98% of heifers from the high mating percentage herd and 88% from the moderate mating percentage herd were present. Of those mustered, 9% were found to have already been pregnant at allocation. This resulted in bull to non-pregnant female mating percentages of 4.5% and 3.25%. A small difference in cumulative pregnancy rate occurred in the second and third months of mating (Figure 88). This difference was sustained for the following 3 months.

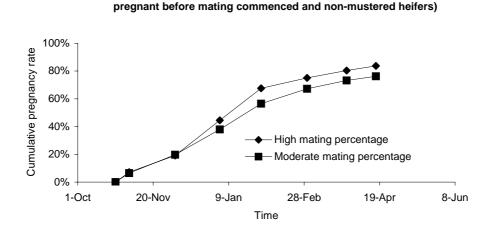


Figure 88. Cumulative pregnancy rates of Canobie heifers (excludes heifers

Whatever the differences were between paddocks, the rate of pregnancies achieved by bulls was low (Table 81). The data presented suggest that approximately 25% of heifers were cycling at the start of mating, increasing to 75% by the end of January and 80% by April.

	High mating percentage	Moderate mating percentage
26 October to 31 January	6	6
31 January to 16 April	21	13

#### **Bow Park observations**

#### Herd dispersion in 2001

Observations at a distance, before disturbance, suggested group sizes of 5 to 80 with a median of 30. The groups (n) recorded were:

- 5, 10, 35, 45, 130 in Frontage paddock (Low % bulls).
- 5, 10, 10, 70, 130 in Wingera paddock (Moderate % bulls).

These data indicate no difference in dispersion between paddocks.

#### Mating outcome

Cumulative pregnancy rates for the first 6 weeks of mating as predicted from foetal age indicated there was no difference between the groups in 2001 (Figure 89). Bulls in Frontage (Low mating percentage) paddock achieved an average of 1 pregnancy per bull per day whilst the rate was half this in Wingera (moderate mating percentage) paddock.

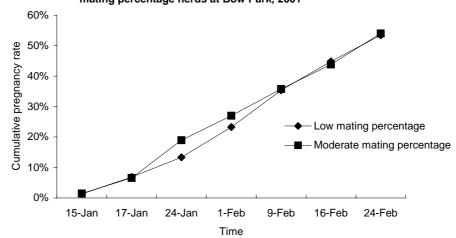
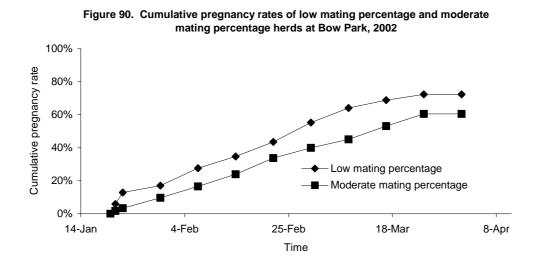


Figure 89. Cumulative pregnancy rates of low mating percentage and moderate mating percentage herds at Bow Park, 2001

Pre-mating in 2002, the most advanced ovarian structure found in 50%, 20%, 18%, and 12% of heifers were a CL, and follicles >11 mm, 9-11 mm, and less than 9 mm, respectively. This indicates that at least 60% of these heifers were cycling at that time. These heifers had moderate to high growth rates over mating, and most were expected to cycle over the 12 week mating. Cumulative pregnancy rates in 2002 were higher in the low mating percentage herd than in the moderate mating percentage herd (Figure 90). The rate of 1 pregnancy per bull per day was the same as in 2001.



## Swan's Lagoon observation

#### Cattle growth

The cattle in BNSE (low mating percentage) had higher body condition scores (~+0.5) than in the other two paddocks in both September 2001 and May 2002. BNSE cattle were also heaviest, with HRYD (Low mating percentage) lightest and SDWE (Moderate mating percentage) intermediate; the liveweight advantage of cattle in BNSE paddock over HRYD paddock was approximately 40 kg and 60 kg in September 2001 and May 2002, respectively.

#### Cattle behaviour

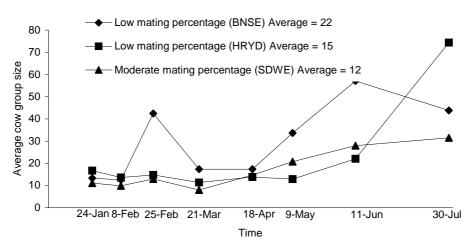
Mean MRI for bulls in SDWE paddock (Moderate mating percentage) was 0.79, but was 0.69 for bulls in BNSE paddock (low mating percentage) and was 0.67 for bulls in HRYD paddock (Low mating percentage) (Table 82). There was a trend for older bulls to have a more restricted movement range than younger bulls. In SDWE paddock, 7 bulls were always found in the northern half of the paddock and 1 always in the southern half; 3 bulls were each found once in the northern half, and the rest of the time were in the southern half of the paddock. In HRYD paddock, one bull remained in the northern half of the paddock, but the other 2 bulls and both bulls in BNSE paddock were found in both the northern and southern halves of the paddock.

SDWE P	addock	HRYD P	addock	BNSE P	addock
(Moderate matir	ng percentage)	(Low mating	percentage)	(Low mating	percentage)
Bull ID	MRI	Bull ID	MRI	Bull ID	MRI
970059	0.97	970054	0.63	970085	0.69
981184	0.91	980027	0.88	980061	0.69
990030	0.72	990055	0.50		
990051	0.84				
990061	0.81				
990081	0.78				
990090	0.78				
990139	0.64				
991518	0.75				
000065	0.69				
000104	0.81				

#### Table 82. Movement range index (MRI) of study bulls

In SDWE paddock, there was a trend for bulls with high MRI (smaller range) to be less frequently found in a bull group ( $r^2$ =0.23). Over the study period, a median of 5 bulls was generally found with cows but no other bulls; a median of 4 bulls was found in groups of bulls and cows. A median of two bulls was found grazing together away from cows, with a bull being found without the company of either bulls or cows only once. In BNSE and HRYD paddocks bulls were always with cows. In BNSE paddock, the bulls were found together only about a third of the time. In HRYD paddock, bulls were alone about half the time; at other times, 2 of the 3 bulls were together.

In all three paddocks, 40% of the cows were found consistently in the southern half. The group size for cows tended to be higher in BNSE paddock than in the other two paddocks, though in the period up to the first weaning, the only difference occurred at one observation (Figure 91).



#### Figure 91. Average cow group size during the Swan's Lagoon observation

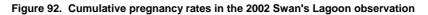
#### Mating outcome

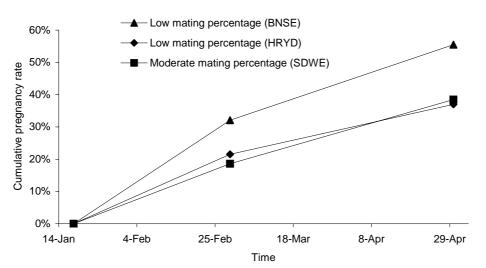
Pregnancy rate was unaffected by cohort, but was related to reproductive status in August 2001 and condition score in April 2002 (P<0.05; Table 83). Several significant interactions between factors in their effects on the April 2002 pregnancy rate appeared to be random effects caused by unbalanced numbers.

Adjusted cumulative pregnancy rate after 96 days of mating was higher in BNSE (Low mating percentage) than in both SDWE (Moderate mating percentage) and HRYD (Low mating percentage) (Figure 92). The average number of days per pregnancy per bull in BNSE, HRYD and SDWE in the first 8 weeks of mating was 2, 2 and 5, respectively; in the next 6 weeks this increased to 3, 9, and 12 days, respectively.

	n	01 Mar	30 Apr
Reproductive status			
1-3 months pregnant, Non-lactating	155	3% a#	7% a
4-6 months pregnant, Non-lactating	302	15% b	36% b
7-9 months pregnant, Non-lactating	159	23% c	58% c
Non-pregnant	200	48% d	67% c
Pregnant, Lactating	49	11% b	11% a
Condition score 30 April 2002			
3 to 4	95	2% a	3% a
5	232	9% a	23% b
6	215	21% b	40% b
7 to 8	324	38% c	65% c

# Means within factor and time with different post-scripts differ significantly (P>0.05)





## 6.5.2.5 Discussion

In the studies reported here, and in two observations previously reported (Fordyce *et al.* 2002), there was variation in whether higher mating percentages increased or decreased cumulative pregnancy rate (Table 84). In almost all cases where differences occurred, these were generated during a short peak-conception period, early in mating and the differences achieved generally remained constant. In most cases, the differences seen could be related to confounding differences in nutrition between paddocks. In the one observation conducted under high-dispersion conditions, the specific reason for the difference between mating groups was not apparent, but given the very low mating loads on bulls during peak mating, the difference was not obviously attributable to bull to female mating ratio.

Station	Dispersion	Mating year	Effect on cumulative pregnancy rate of higher	Days per pregnancy per bull during peak mating			Confounding with higher mating percentages
	mating percentage	High mating percentage	Moderate mating percentage	Low mating percentage			
Kamilaroi	Low	1995	+8%	5	3		Better nutrition
Kamilaroi	Low	1996	+6%		4	2	Better nutrition
Canobie	High	2001	+8%	6	6		
Bow Park	Low	2001	Nil		2	1	Heavier heifers
Swan's Lagoon	Moderate	2002	Nil		5	2	
Swan's Lagoon	Moderate	2002	-18%		5	2	Poorer nutrition
Bow Park	Low	2002	-11%		2	1	Heavier heifers

#### Table 84. Summary of studies of mating ratio effects on mating outcomes

## 6.5.2.6 Conclusion

These studies showed that conceptions were not delayed when bull mating percentages were reduced from high to moderate under high dispersion grazing management in north Australia, or from moderate to low under low or moderate herd dispersion. However the data provides further support to the recommendation that mating percentages of 2.5% reproductively-sound bulls (moderate mating percentages) are adequate under most north Australian cattle management conditions.

## 6.6 Factors influencing beef cattle libido and paternity

## 6.6.1 Background

Single-sire studies at Swan's Lagoon showed that 8% of bulls that passed breeding sounding examinations produced very few progeny after a 3-month mating period (Holroyd et al. 1993). In most instances there were no obvious reasons and these failures could have been related to libido or serving ability problems.

The studies by Holroyd et al. (2002) that examined predictors of fertility of bulls in multiple-sire herds in northern Australia, found a number of relationships between traits measured prior to mating and calf output. Sheath and testicular traits such as scrotal circumference were generally not related to calf output and neither was dominance hierarchy. In contrast, measures of semen quality based on sperm morphology were. However the pre-mating traits in the multiple regression models explained only between 35 and 57% of the variation in calf output of individual bulls. Other factors such as the behaviour of bulls in the paddock may have been contributing to calf output. Further, studies by Fordyce et al. (2002) showed that some bull behaviours were related to high calf output. These behaviours included restricted movement range, being with females at the majority of observations, stable social behaviour and social dominance.

Most information on cattle sexual behaviour relates to animals in confined areas such as serving capacity tests and there is limited information published on the sexual behaviour of rangeland cattle. A review of factors influencing beef cattle libido and paternity was undertaken to identify gaps in our knowledge and, thus, provide a sound basis for future behaviour research in this area.

## 6.6.2 A literature review of factors influencing beef cattle libido and paternity

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## 6.6.2.1 Introduction

This review focused on the factors that determine whether male and female beef cattle will come together and mate, making the assumption that males and females are physically capable of producing viable sperm and ova, of mating and producing offspring. The review concentrated on free-ranging cattle, where there is minimal human interference. The full literature review is attached as Appendix 2 and was reported in Milestone 7, November 2002.

In the review, libido referred to sexual motivation, revealed through behaviours such as mate seeking, detection, selection and courtship, and mating and ejaculation. It was not the intention of the review to detail the neurophysiological and neuroendcrinological mechanisms of libido, but rather to concentrate on behavioural constraints and factors that may affect the expression of libido, and, thus, influence which individuals will mate and achieve conception.

The review covered the following areas:

- Social organisation of animals within a herd
- Sexual behaviour including mate seeking, detection and selection by males, and females, courtship and mating

- Sperm selection and competition
- The assessment of libido, in both males and females
- Factors affecting libido and conception including herd dispersion, male:female ratios, social interactions between males, age of males, sexual experience of males, genetics and breeds, climatic and thermal environment, nutrition, relocation and temperament and novelty
- The implications for management including the conduct of serving capacity tests, single-sire verses multiple-sire groups, introducing new bulls into an existing new bull group and composition of bull groups.

## 6.6.2.2 Summary of main findings of factors affecting the expression of libido

Bull libido is assessed in a variety of ways, all of which determine the sexual responsiveness of bulls to females. The sexual activity of bulls is influenced by test conditions and measures of libido obtained in test situations may not reflect sexual activity in the mating paddock. Furthermore, fertility is multifactorial in nature. Consequently measures of libido obtained in tests do not necessarily predict bull fertility.

Genotype plays a role in determining libido, and inherent fertility differences exist between individual bulls. Generally, *Bos taurus* breeds show higher and less variable levels of libido in test situations than do *Bos indicus* breeds, but this appears not to translate into fertility differences between the breeds. There is anecdotal evidence that bulls and females mate preferentially with their own breed with implications for fertility.

Multiple males increase the expression of libido in bulls, but it is uncertain whether this translates into improvements in herd fertility. However, there are consequences for individual bull fertility, as there is ample evidence of inherent differences between bulls. Although data are limited, it appears that male:female ratios have minor effects on libido and fertility, although in libido tests too much competition between bulls increases aggression and reduces sexual activity. Social relationships between bulls can, also, affect the expression of libido, with subordinate bulls being inhibited by the presence of dominant bulls. There is evidence that dominant bulls may achieve more matings at pasture, but this is not necessarily reflected in their fertility. Anecdotal evidence indicates that multiple matings with the same or different bulls may reduce the duration of oestrus, and the data on the consequences of multiple matings for herd fertility are equivocal.

Older bulls show greater expression of libido in tests and appear more efficient in serving, although these changes may reflect greater sexual experience. Provided bulls are sexually mature and physically able to mate, age *per se* appears not to affect fertility, but age interacts with dominance, which can influence fertility.

Limited research on thermal and nutritional effects indicate some adverse consequences for libido of climatic extremes for unadapted bulls and of over-feeding, but not under-feeding.

Limited research has investigated the effects on libido and fertility of multiple stressors associated with relocation; relocation to environments very different in photoperiods and temperatures has long-lasting detrimental consequences for fertility, but there is no information on the effects of relocation to less 'extreme' environments.

Too few studies have been conducted to draw conclusions about the effects of topography and herd dispersion on libido and fertility.

Temperament is likely to affect the expression of libido when animals are put into new situations, and highly fearful bulls appear to show low libido, although this has not been critically researched.

## 6.6.2.3 Areas requiring further research

Some obvious features of the literature on cattle sexual behaviour are: its age, with the majority of the foundation work being conducted in the 1950s to 1980s; the very limited numbers of studies conducted on some aspects (often one or two); the descriptive nature of much of the work; the lack of experimental details; a paucity of robust data; and the repeated reference to the same few papers. There appears to be the need for the conduct of some rigorous hypothesis testing in this subject area. A consequence of these features is that there is the potential that many "accepted facts" about aspects of cattle sexual behaviour are far from proven. It is probably timely to re-examine many aspects of libido to ensure that previous findings are, indeed, sound. In addition there are a number of areas that are, apparently, either unexplored or have given rise to equivocal results to date. The following are some examples and the elucidation of these areas has the potential to have significant benefits on the efficiency of bull use and the speed at which herd genetic improvement occurs.

#### 1 Mate choice

The apparent preference for 'like breeds' requires more work. If it is, indeed, the case that males and females prefer to mate with their own genotype then bull genotype needs to be carefully considered in light of the female genotype. This is unlikely to be an issue where there is a single bull, but may play a role where there are multiple males of differing genotypes. We do not know what other factors are involved in mate selection and this needs to be researched further. There is evidence in other species that choice is made on gene quality and compatibility between genotypes, being revealed through features such as symmetry and secondary sexual characteristics. We have no idea if such systems operate in cattle. Given that producers attempt to introduce 'superior genetics' and exploit hybrid vigour in their herds by using bulls of genotypes very different to that of the females, there could be significant consequences of mate choice on herd improvement.

## 2 Sperm selection and competition

Even if a bull mates with a cow, paternity is not assured due to sperm selection. Furthermore, in many species where there is mating by multiple males, sperm competition occurs in the female's reproductive tract. As we do not know if sperm selection and competition operate in cattle, these are areas that warrant further research, particularly as this would provide an indication of the relative importance to fertility of mate selection and sperm competition.

## 3 Courtship and the timing of mating

It has been suggested that bull courtship behaviour may actually induce oestrus in females, but there is, to date, little evidence to support this hypothesis. Also, as timing of insemination in relation to oestrus appears to be a crucial factor in males achieving conception, we need to know more about the effects of courtship and guarding by individual bulls in multiple-sire groups and whether these behaviours are related to the number of offspring produced by individual bulls. Furthermore, it appears that older, more experienced bulls may be much more adept at determining the 'optimum' timing of mating. It would be very useful to determine the mechanism by which they do this in order to improve the efficiency of bull use.

#### 4 *Multiple matings and termination of oestrus*

There are equivocal data relating to the effects on fertility of multiple matings with the same and different bulls. Some studies report that females will accept multiple serves from the same bull, others state that females will accept serves from different males, but not repeated serves from the same male. Furthermore, there are differing opinions as to whether multiple matings improve fertility, and there are interesting observations that indicate that multiple matings reduce the length of oestrus or even terminate it. This whole area warrants further investigation given that it has the potential to have very significant effects on fertility.

#### 5 Pre-coital stimulation and sperm quality

There appears to be some evidence that pre-coital stimulation of bulls may result in improvements in sperm quality and fertility. However, as this hypothesis and the data on which it is based are rather tenuous, it is an area that warrants further investigation.

#### 6 Dispersion

It has been suggested that cattle dispersion (and the factors that contribute to it) will affect the ease with which bulls locate females, and, thus, influence herd fertility. The emphasis in work on libido has been on that of the bull and the role of the female in seeking the male has been given little attention. Further research is needed in the area of mate-seeking and the effects that cattle dispersion has on this.

#### 7 Relocation of bulls

Given the high probability that bulls will continue to be purchased at considerable distances from where they will be used, there is a need to determine the effects of relocation on fertility and the relative contributions of the multiple stressors to any fertility problems.

#### 8 Serving capacity tests and sexual competitiveness

Bulls assessed in yard tests as having high serving capacity scores appear to demonstrate the same levels of sexual activity, in single-sire mating systems, as those scored as having low serving capacity scores. However, in multiple-sire mating systems, high serving capacity bulls perform greater levels of sexual activity than do low serving capacity bulls. Furthermore, high serving capacity bulls, also, may have a greater tendency to serve females previously served by another bull, compared to lower serving capacity bulls, but this requires confirmation. These various behaviours may be related to the social relationships between bulls, or, alternatively, serving capacity tests may actually be measuring an aspect of sexual competitiveness, rather than libido *per se*. If serving capacity tests are to be continued to be used as a predictor of bull libido then these issues need to be resolved so that tests can be refined to give more meaningful results.

## 6.7 Delivery of useful, practical information

#### 6.7.1 Scientific papers from NAP.104 and new scientific papers from NAP3.117

- Bertram JD, Fordyce G, McGowan MR, Jayawardhana GA, Fitzpatrick LA, Doogan VJ, De Faveri J and Holroyd RG. (2002) Bull selection and use in northern Australia 3. Serving capacity tests. *Anim Reprod Sci* 71:51-66
- Chenoweth PJ, Petherick JC and Bertram JD (2002) Sexual behaviour of cattle. In "Bull fertility: Selection and Management in Australia" pp 4.1-4.9, (Geoffry Fordyce, editor), Australian Association of Cattle Veterinarians, PO Box 30 Indooroopilly Qld 4068, ISBN 0-9585654-3-0.
- Fitzpatrick LA, Fordyce G, McGowan MR, Bertram JD, Doogan VJ, De Faveri J, Miller RG and Holroyd RG. (2002) Bull selection and use in northern Australia 2. Semen traits. *Anim Reprod Sci* 71:39-49
- Fordyce G, Fitzpatrick LA, Cooper NJ, Doogan VJ, De Faveri J and Holroyd RG. . (2002) Bull selection and use in northern Australia 5. Social behaviour and management. *Anim Reprod Sci* 71:81-99
- Gardiner B and Fordyce G (2002) Evaluation and certification of bull fertility. In "Bull fertility: Selection and Management in Australia" pp 11.1-11.4, (Geoffry Fordyce, editor), Australian Association of Cattle Veterinarians, PO Box 30 Indooroopilly Qld 4068, ISBN 0-9585654-3-0.
- Holroyd RG, Taylor E and Galloway D (2002) Physical examination of bulls. In "Bull fertility: Selection and Management in Australia" pp 3.1-3.19, (Geoffry Fordyce, editor), Australian Association of Cattle Veterinarians, PO Box 30 Indooroopilly Qld 4068, ISBN 0-9585654-3-0.
- Holroyd RG, Bertram DJ and Holmes R (2002) Managing young bulls from weaning to mating. In "Bull fertility: Selection and Management in Australia" pp 6.1-6.8, (Geoffry Fordyce, editor), Australian Association of Cattle Veterinarians, PO Box 30 Indooroopilly Qld 4068, ISBN 0-9585654-3-0.
- Holroyd RG, Doogan VJ, De Faveri J, Fordyce G, McGowan MR, Bertram JD, Vankan DM, Fitzpatrick LA, Jayawardhana GA and Miller RG (2002) Bull selection and use in northern Australia 4.Calf output and predictors of fertility of bulls in multiple-sire herds. *Anim Reprod Sci* 71:67-79.
- McGowan MR, Bertram JD, Fordyce G, Fitzpatrick LA, Miller RG, Jayawardhana GA, Doogan VJ, De Faveri J and Holroyd RG. (2002) Bull selection and use in northern Australia 1. Physical traits. *Anim Reprod Sci* 71:25-37
- Petherick JC (in press) A review of some factors affecting the expression of libido in beef cattle and individual bull and herd fertility. *Appl Anim Behav Sci*
- Perry V, Phillips N, Fordyce G, Gardiner B, Entwistle K, Chenoweth P and Doogan VJ (2002) Semen collection and evaluation. In "Bull fertility: Selection and Management in Australia" pp 5.1-5-19, (Geoffry Fordyce, editor), Australian Association of Cattle Veterinarians, PO Box 30 Indooroopilly Qld 4068, ISBN 0-9585654-3-0.
- Wade CM, Bertram JD, Pullen B and Perry V (2001) Heritability of mating behaviour traits in beef bulls. Proc. Assoc. Advmt. Anim. Breed. Genet. 14: 341-344.

## 6.7.2 Conference papers

- Bertram J, McGowan MR, Doogan VJ and Phillips NJ (2000). Preliminary studies of criteria for selection of young tropically adapted bulls. Proceedings of the Australian Association of Cattle Veterinarians 2000 Perth Conference pp 130-132.
- Burns, BM, Bell GT, Bertram J, Fawcett G and Holroyd, RG (2001). Enhancing Tropical Beef Cattle Genetics, Reproduction and Animal Breeding Skills as Applied to Tropical Beef Industry Supply

Chain Systems, Progress Report, Phase II - In Vietnam, 25 May 2001 - 20 June 2001, AusAID Vietnam-Australia Development Cooperation Program, Capacity-Building for Agriculture and Rural Development Program (CARD), pp. 1-46.

- Doogan Vivienne, Foster Scott, Swain Tony and Holroyd Dick (2002) Modelling growth of young bulls Australasian Genstat Conference, 4-6 December 2002, Busselton WA
- Fordyce G and Kenneally G (2002). Dry season body condition loss temporarily suppresses breeding soundness of bulls in north Queensland. In "Bull fertility: Selection and Management in Australia" Short submissions pp 1-3, (Geoffry Fordyce, editor), Australian Association of Cattle Veterinarians, PO Box 30 Indooroopilly Qld 4068, ISBN 0-9585654-3-0
- Holroyd RG, Fordyce G, Bertram JD, Fitzpatrick LA, Doogan VJ, Turner LD and McGowan, MR (2001). Use and evaluation of *Bos indicus* bulls in extensive and semi-extensive management conditions: an update of current activities in northern Australia. In "4<sup>th</sup> Simposio Internacional de Reproduccion Animal" pp 9-24. (Mariana Caccia, editor), IRAC (<u>irac@iracbiogen.com.ar</u>), Cordoba, Argentina.

## 6.7.3 Books and saleable publications

Evaluating and Reporting Bull Fertility (Keith Entwistle and Geoffry Fordyce, editors). Australian Association of Cattle Veterinarians (Publisher), Indooroopilly, Qld 4068.

Bull Selection – An aid for beef producers on buying better bulls (2003) 2nd ed. (authors John Bertram, Geoff Fordyce, Richard Holroyd, Vince Edmondston, Rebecca Farrell, Kay Taylor, Rick Whittle, Mick Tierney and Keith Entwistle). Training series QE 94009 Dept Primary Industries.

MLA Breeding EDGE Male Reproduction section - (Product development and pilot workshop).

## 6.7.4 Press releases

Numerous-no records kept

## 6.7.5 Video material

"Bull Selection, using the BBSE measures with confidence" was launched at Beef 2003 on 30 April by JD Bertram.

## 6.7.6 CD and course material

"Structure, semen and sperm abnormalities". Course material for Emerald Agricultural College.

## 6.7.7 Articles in newsletters, magazines and periodicals

Title	Source	Edition
Increasing your bull power	MLA Feedback	October 2000, p18
Bull testing is for everyone	MLA Feedback	October 2000, p19
Select early maturing bulls for high production	Farming Ahead	May 2001
Function delivers performance	Beef Improvement News	April 2002

Location	Торіс	Staff	Date
Rockhampton, Beef 2000	Seminar on Bull Management	JD Bertram and G Fordyce	10 April 2000
'Mondah', Prairie	Bull fertility and assessment	G Fordyce	17 July 2000
'Compton' Downs, Hughenden	Bull fertility and assessment	G Fordyce	18 July 2000
'Bylong', Richmond	Bull fertility and assessment	G Fordyce	19 July 2000
'Mt Ravenswood', Charters Towers	Bull fertility and assessment	G Fordyce	3 August 2000
Murdoch University	Bull fertility testing	J Bertram and E Taylor	30 June 2000
'Belmont' Beef 2000	Bull assessment	RG Holroyd, JD Bertram and G Fordyce	13 April 2000
'Pialaway', Meandarra	Bull selection	JD Bertram	10 August 2000
'Gyranda', Theodore	Bull assessment	JD Bertram	17 August 2000
Cattle Council	Code of practice for serving capacity testing	JD Bertram	18 August 2000
'Caiwarra', Charters Towers	Bull selection	JD Bertram and G Fordyce	5 September 2000
Clermont	Bull fertility workshop for veterinarians	G Fordyce	20 January 2001
Rockhampton, MLA workshop on Market Analysis	Breeding, Genetics and Reproduction	JD Bertram	5 February 2001
'Belmont'	Reproductive physiology and bull selection – Vietnamese AusAid participants	JD Bertram and RG Holroyd	2 May 2001
'Swan's Lagoon' Open Day	Bull selection and management	G Fordyce	14 June 2001
Dalby, Braford Society	Results of Bullpower 1 and 2	JD Bertram	7 July 2001
'Glenlovely', Yelarbon. Border Beef Marketing Co-operative	Sire selection and bull fertility testing	JD Bertram	23 August 2001
Emerald Pastoral College Beef School	Bull selection	JD Bertram	18-20 September 200
"Lakeland Downs" Mareeba, Field Day	Bull selection	G Fordyce	20 November 2001
Mt Garnet	Bull selection	G Fordyce	21 November 2001
Darwin, AACV conference on Bull Fertility	Physical traits and semen assessment	JD Bertram and V Perry	13 July 2002
Win TV	Relocation results	RG Holroyd	24 January 2003
AgForce North Regional Council meeting, Cloncurry	Bullpower results	G Fordyce	09 April 2003
Win TV	Breeding soundness examinations	JD Bertram	01 May 2003
'Belmont' – Cattle Council	Bullpower results	RG Holroyd	07 March 2003
'Belmont' – Beef 2003	Bullpower results	RG Holroyd	28 April 2003
Rockhampton – Beef 2003	Bull evaluation and reporting	G Fordyce, KW Entwistle and RG Holroyd	28-29 April 2003
'Cardowan'	Bull selection using BBSE criteria	JD Bertram	28 May 2003
'Belmont' 50 <sup>th</sup> anniversary	Results from Bullpower studies on Belmont	RG Holroyd	12 June 2003

## 6.7.8 Field days, workshops and short presentations

## 7 Success in achieving objectives

## Objective 1. Quantifying the impact of relocation on subsequent fertility

The experimental studies showed that, providing bulls are transported and then managed post-relocation under favourable conditions, there appears to be no effect of genotype (either Brahman or Composite), concentrate feeding prior to relocation or the relocation process per se, on semen traits or sperm morphology either in the short term (1.5-3 months) or long term (12 months) post-relocation. The observational studies identified that about 50% of bulls failed a BBSE up to 3 months after sale and relocation. We were unable to determine the cause of these failures but they appeared to be independent of genotype, age, source and prior nutrition. We speculate that many bulls in the industry are being relocated and managed post-relocation under less than ideal conditions and this contributes to many bulls failing a BBSE post-relocation.

# Objective 2 Identifying traits whereby bulls can be selected for fertility at an early age

At about 14 months of age, bulls can be selected on a number of physical traits, which reflect their performance as 2-year-olds. There were generally high repeatabilities for liveweight and scrotal circumference, moderate to high for sheath depth, umbilical thickness and rosette score. However sheath score, a subjective measurement, had only low to moderate repeatability.

The studies also showed that selection for reproductive traits in yearlings was a poor predictor of reproductive traits, particularly semen morphology, as a 2-year-old. Most repeatabilities for semen traits and sperm morphology were low, as were correlations between times for normal sperm. Where comparisons could be made, in the *Bos indicus* genotypes at 14 months of age only 12% of bulls produced semen with at least 50% normal sperm. In the Composites at the same age, 59% of bulls produced semen with at least 50% normal sperm.

## Objective 3 Identifying traits that predispose bulls to early breakdowns in their working life

The predisposing factors for preputial eversion appear to differ between *Bos indicus* derived breeds and *Bos taurus* breeds. Although the literature showed that eversion in *Bos taurus* bulls was seen mostly in polled bulls and these polled bulls have a decreased size of the preputial retractor muscles, there was no evidence of poorer preputial muscle development or more preputial eversion in polled bulls compared to horned bulls that were examined.

Preputial prolapse in polled Santa Gertrudis bulls could be due to the significantly reduced size and development of the preputial retractor muscle complex in polled bulls. No obvious cause of prolapse in the horned bulls was determined from dissection studies.

From the study examining position of the penis and length of preputial eversion we concluded that preputial eversion in *Bos indicus* derived bulls was related to the position of the penis within the sheath.

## **Objective 4** Developing knowledge of sexual behaviour of cattle

A comprehensive literature review of factors influencing beef cattle libido and paternity was submitted as Milestone 7, 22 November 2002. This was modified for general distribution. The literature review has been refined and accepted by *Applied Animal Behavioural Science* for publication. The submitted review covers the following areas; the assessment of libido both in males and females; the effects on the expression of libido of genetics and breeds, male:female ratios, multiple males and multiple matings, social relationship between males, age of males, sexual experience of males, the climatic/thermal environment, nutrition, multiple stressors associated with relocation, topography and herd dispersion, temperament in novel environments; and, implications for management practices and areas requiring further research.

# Objective 5 Defining the effects of bull percentages on conception patterns and pregnancy rates at different levels of herd dispersion

These studies showed that pregnancy rates were not reduced as a direct result of reducing bull percentages from high to moderate under high dispersion grazing management in north Australia, or from moderate to low under low or moderate herd dispersion. The data provided further support to the recommendation that mating percentages of 2.5% reproductively-sound bulls are adequate under most north Australian cattle management conditions.

## Objective 6 Delivering useful, practical information to industry.

Data from this project have been incorporated in 7 scientific papers, 5 conference papers, 4 magazine articles and 2 saleable publications. Three of the team (RG Holroyd, G Fordyce and JD Bertram) were lead presenters at the AACV Conference, Darwin 2002, on "Bull fertility". A video on "Bull selection, using the BBSE measures with confidence" was launched at Beef 2003 and a CD on "Structure, semen and sperm abnormalities" is now part of the teaching material for the Emerald Agricultural College. There have been 27 presentations at field days and workshops and 2 television interviews.

## 8 Impact on the Meat and Livestock Industry

The study demonstrated that bulls could be relocated without any adverse effects on reproductive traits providing the following procedures are followed: minimise time in yards prior to relocation; minimise transport time; transport animals that are familiar with each other and place bulls on good nutrition on arrival. However these conditions cannot always be followed particularly if bulls are sold through a sale yard. Fifty percent of bulls in the observational studies failed a BBSE after relocation and these failures seemed to be independent of genotype, source, age and prior nutrition. Until the causes of failure of BBSE are identified, relocation will continue to have an adverse effect on the reproductive performance of herds in the northern cattle industry.

Providing bulls are not overfed and become fat prior to relocation, feeding high-energy supplements prior to relocation does not affect subsequent reproductive traits. However feeding high energy supplements to bulls should be discouraged from a production perspective because of the associated risks of laminitis and increased flight speed which may result in animals that are more difficult to handle. Further most of the benefits of increased liveweight and scrotal circumference in fed bulls were eroded after 12-months.

If bulls have to undergo a BBSE whilst in poor body condition, the evidence from our observations is many of these bulls may not pass. Culling of these failed bulls should be treated cautiously as the ranking of bulls on scrotal circumference and sperm morphology is similar to that when body condition has improved, thus giving an indication of future relative breeding soundness.

The observational studies also demonstrated that poorly adapted genotypes can be introduced into some tropical areas without any detrimental effect on reproductive traits providing bulls are managed adequately post-relocation. This has important implications for the northern cattle industry for introduction of diverse genotypes so that producers can target a wider range of markets. However one observation clearly showed the disastrous consequences of introducing poorly adapted cattle into a harsh tropical environment when the bulls were not provided with adequate nutrition post-relocation.

Mating yearling bulls has the advantage of accelerating genetic improvement through reducing the generation interval. As well, younger bulls are much easier to handle than older bulls. Even though yearling 5/8 Brahmans bulls passed a BBSE pre-mating, their mating outcome, as measured by pregnancy rates, was still less than those of 2-year-old bulls. This would result in reduced branding rates in northern herds. Our limited data for Composites indicate that mating yearling bulls would not impact on herd reproductive performance.

At about 14 months of age bulls can be selected on a number of physical traits such as liveweight, scrotal

circumference and sheath and umbilical measurements that reflect the relative value of these traits at 2 years of age. However sperm morphological assessment as a yearling is no indication of its morphological status as a 2-year-old. As sperm morphology is a more important pre-mating indicator of mating outcome in multiple-sire herds than physical traits, caution should be exercised in culling bulls as yearlings based on sperm morphological assessment.

The data showed that once bulls reached sexual maturity, the repeatability of normal sperm is high. As well the data demonstrated that pregnancy rates of females mated to bulls with less than 50% normal sperm in single-sire matings is reduced particularly under high mating loads. These two findings reinforce the importance of the assessment of sperm morphology as part of a thorough breeding soundness examination of a bull and will provide veterinarians and producers with further confidence in the relative contributions of various bull reproductive traits to calf output.

Polled bulls should be selected from herds that do not have prolapse problems, as herds of polled bulls with developed preputial muscles are available. Producers should avoid selecting bulls with extremely large sheaths as these bulls have a greater propensity to evert more than bulls with small sheaths. Although the direct link between eversion and prolapse has never been proven, eversion must still be a consideration in selection, as the larger the eversion the greater is the chance of injury. The anatomical studies have put into perspective the prevalence of preputial prolapse in *Bos indicus* genotypes. Only one in 200 bulls will develop preputial prolapse under northern Australian conditions. Under commercial conditions, the main loss associated with the development of a prolapse would be the loss of use of the bull and a reduced pregnancy rate if the onset of prolapse occurred whilst the bull was in a single-sire mating or as part of a multiple sire mating with high mating loads

The results from the study of the effect of herd dispersion and bull mating percentages on herd fertility add support to the current recommendation that bull mating percentages can be reduced to 2.5% providing bulls have passed a BBSE. This will allow considerable savings for producers, as less bulls need to be either bred or purchased as replacements. Alternatively producers can spend the same amount of money and purchase fewer bulls of superior genetic merit for traits such as growth, yield and meat quality.

In previous studies (Holroyd *et al.* 2002a), premating measurements of physical and reproductive traits of bulls only explained, at best, 57% of the variation in calf output of different bulls in multiple sire herds. It has been speculated that the reproductive behaviour of both bulls and females in the paddock may be more important than previously considered in contributing to calf output. The literature review on factors influencing beef cattle libido and paternity highlighted a number of areas of that may affect the expression of libido thus influencing which bulls achieve mating and conception. A better understanding and application of bull reproductive behaviour could improve reproductive rates of beef cattle in extensive areas of northern Australia.

# 9 Conclusions and Recommendations

# 9.1 The effect of relocation on bull reproductive traits

# 9.1.1 Conclusions

The perception held by veterinarians and pastoral companies was that relocation of bulls had an impact on their subsequent reproductive performance. However, many of the respondents found it difficult to quantify the extent of the problem, as most bulls were not re-examined until many months or years after relocation. The observational data, presented in Section 6.1.3, on bulls sold and relocated to tropical Queensland would suggest that there is a problem with some bulls and some cohorts, as approximately 50% of bulls failed a BBSE up to 3 months after sale and relocation, although up to 80% of these bulls passed a BBSE within 6 months under favourable nutritional and management conditions. In contrast, the two detailed experiments showed that there were only minor effects of relocation on changes in reproductive traits, either in the short term (1.5 - 3 months) or long term (12 months). There are number of plausible reasons for the differences between the observational and the experimental studies.

• In retrospect, in both experiments, bulls were relocated under favourable conditions; bulls were

not exposed to a sale yard process; bulls were familiar with each other; time in the yard before transport was minimised, water was on offer and in both cases the relocation process was relatively short. On arrival bulls were placed onto good pasture and were not mixed with other bulls already on the property. We assume that our procedures of relocation were less stressful than exposing bulls to sale yards but we have no evidence to support this.

- The observational data had very few pre-relocation assessments of bulls. Attempting to obtain these and then tracking the bulls post-relocation would have been a logistically difficult. Presumably many of these had passed a BBSE by the examining veterinarian but the procedures for BBSE particularly for sperm morphology had not been standardised until 2004. Many of these bulls would not have been examined for sperm morphology and would have passed the semen aspect of a BBSE on the basis of mass activity and motility only.
- The bulls in the experiments were not mated post-relocation whereas many of the bulls in the observations were. However we have no data comparing ejaculates of mated and non-mated bulls. The expectation would be that mated bulls would have ejaculates of lower volumes, be less concentrated and thus have lower mass activities compared with non-mated bulls. We are not aware of any evidence for differences in sperm morphology between mated and non-mated bulls apart from non-mated bulls tending to have higher levels of "stale" ejaculates. Stale ejaculates have a high volume, high sperm concentration and low sperm motility and are associated with bulls that have not ejaculated recently.
- Different technicians performed the morphological assessments. Our experience is that, although differences exist between technicians, the relative rankings of bulls remain the same, regardless of the technician. Whilst this does not influence the results in the two experiments, as the primary variable was change in absolute morphological values, it could influence the results in the observations, as an arbitrary 50% normal sperm was used as a pass/fail of a BBSE in some of the investigations. There was a higher prevalence of head abnormalities, particularly pyriform heads, with the observational data. These abnormalities, especially mild cases of pyriform heads have a questionable role in inhibiting fertilisation. As a consequence to these discrepancies between technicians, the AACV has introduced an accreditation procedure for technicians for sperm morphology assessment.
- The nutritional conditions experienced by the relocated bulls in the two experiments appeared to be not as severe as experienced by many of the bulls in the observational data. The relocated bulls at Toorak were heavier than the non-relocated bulls at Brigalow. The relocated bulls at Swan's Lagoon, although lighter than non-relocated bulls at Belmont, appeared to have better growth rates than many of the relocated bulls in the observations.

There were minor changes in haematological and biochemical parameters and these changes indicate that relocation was marginally stressful. Whilst biochemical parameters indicated that relocated bulls at Toorak were not coping as well as the non-relocated bulls at Brigalow, this was not reflected in growth, scrotal circumference or differences in semen quality or sperm morphology. At Swan's Lagoon, none of the biochemical changes could be attributed to relocation *per se* apart from an elevation of L-lactate on arrival.

The studies provided evidence to dispel many of the perceptions associated with reproductive problems post-relocation i.e. more prevalent in *Bos taurus* or non-adapted genotypes, more prevalent in good condition or fat animals and more prevalent in bulls that had been fed concentrates prior to relocation.

The observational data on bulls relocated to tropical Queensland showed that 50% of the bulls failed a BBSE and this was independent of genotype. Also the effect of poor nutrition increasing the likelihood that a bull failed a BBSE was often, but not always, greatest in *Bos taurus* genotypes. There was no effect of breed on sperm morphology in the bulls relocated to Swan's Lagoon. The Composite bulls in the Belmont-Swan's Lagoon study were comparable with the bulls moved to property S8 in the observational study. However in the latter study, Composite bulls experienced very severe nutritional stress and there were dramatic declines in body condition, scrotal circumference, normal sperm and proportion of bulls passing a BBSE. Therefore it would appear that providing bulls do not experience severe nutritional

stress, then genotype is not an important factor in whether bulls experience reproductive problems postrelocation.

When Brahman bulls were relocated from Brigalow to Toorak, there was no effect of feeding prior to relocation on reproductive traits of bulls post-relocation even though the fed bulls were 150 kg heavier and 2 units of body condition better than non-fed bulls. Whilst the fed bulls averaged 7 in body condition, there were not any bulls that were very fat. The observational studies found that prior concentrate feeding did not appear to influence whether bulls passed a BBSE.

It was not possible to precisely determine the impact of nutrition post-relocation on the reproductive traits of bulls. The observational studies indicated that nutritional stress accompanied by a significant loss of condition was associated with reduced scrotal circumference and lowered normal sperm. The same degree of body condition loss did not occur in the bulls in the relocation experiments, and there was no effect of relocation on semen quality or sperm morphology. The result from the observational data when bulls were in poor body condition, showed that the rankings of scrotal circumference and sperm morphology would be similar to that when body condition was recovered. Thus there is some indication that future breeding soundness can be evaluated in bulls when they are in backward condition.

The experimental studies showed that providing bulls are transported and then managed post-relocation under good nutritional conditions, there appears to be no effect of genotype, feeding or relocation on semen traits or sperm morphology. The observational studies identified that about 50% of bulls failed a BBSE up to 3 months after sale and relocation and this appeared independent of genotype, age, source and prior nutrition. We speculate that many bulls in the industry are being relocated and managed post-relocation under less than ideal conditions. This presumably is having an adverse impact on the reproductive performance of the northern cattle industry.

# 9.1.2 Recommendations

# Recommendation 1. Further research would better quantify the effects of relocation on bull reproductive traits particularly bulls relocated through sale yards.

Measurements are required on bulls that have been exposed to larger insults than those imposed in these experiments such as following bulls through sale yards as part of the relocation process. The observational studies highlighted that 50% of relocated bulls fail a BBSE within 3 months of relocation and this failure appears not to be related to genotype, age, location and prior feeding.

# Recommendation 2. Further research on the effects of high-energy diets on physical, behavioural and semen traits would define thresholds for levels and duration of feeding.

Whilst there have been some reduction in levels of high-energy diets fed to bulls, feeding bulls for sale is an entrenched practice of the beef cattle industry particularly the stud sector. There appears to be very few reproductive benefits of this feeding, apart from possibly increasing the onset of puberty and it is used mainly as a marketing tool for the sale of bulls. There is considerable interest from bull sellers on cost effective levels to feed bulls to maximise growth but without compromising physical and reproductive traits and sale price.

# 9.2 Increasing the working life of bulls – Identifying criteria whereby bulls can be selected for fertility at an early age and evaluation of yearling mating

## 9.2.1 Conclusions

These studies focussed on examining the potential outcome of mating bulls as yearlings. As mating is a function of seasonal conditions and time of the year, bulls would be first mated at about 14 months of age. In northern Australia this age coincides with most start of matings for herds that have a restricted mating length or peak conceptions for all-year round mated herds. A comparison of the mating performance of yearling bulls and 2-year-old 5/8 Brahmans bulls was achieved only at Swan's Lagoon. It had been our intention to do this at 2 other locations but drought conditions prevented this. The mating

performance of yearling Composite bulls at Narayen was documented.

Many of the bulls had not reached puberty by 14 months of age. There was a large range in the age at which bulls reached puberty in both genotypes at Belmont with the average (and ranges) for Brahman and Composites being 17.4 (13.5 - 24) months and 15.4 (12.5 - 18.5) months respectively. In the Bos *indicus* genotypes at 14 months of age, only 12% of bulls produced semen with at least 50% normal sperm. In the Composites, 59 % of bulls produced semen with at least 50% normal sperm.

The mating performance of yearling bulls appeared to be dependent upon genotype. Yearling 5/8 Brahman bulls mated as single-sires can cause females to become pregnant but the overall pregnancy rate was about 18% units lower than for 2-year-old bulls. As well the rates of conception were lower (calculated rates of conception being 4.3 v 5.2 conceptions per week respectively for 1- and 2-year-old bulls). Although there was no comparison with 2-year-olds, the yearling Composite bulls at Narayen mated as multiple sires achieved a 95% calving rate as multiple sires. However their rate of conception was not known.

Supplementing yearling bulls with moderate levels of concentrates had a variable effect on scrotal circumference but there was no effect on semen quality or sperm morphology. However there were a number of detrimental effects. At Brigalow, 3 of the 38 bulls developed lameness from acidosis with one bull having to be removed from the experiment. The 150 kg liveweight difference of the fed bulls at the end of feeding period was reduced to a 3 kg difference 12 months later. Similarly the 4.3 cm difference in scrotal circumference of fed bulls was reduced to 0.7 cm in the same period. As well flight speeds of fed bulls were higher and libido scores of fed bulls at Narayen were lower than non-fed bulls.

Yearling bulls can be selected on a number of physical traits, which reflect their performance as 2-yearolds. There were generally high repeatabilities for liveweight and scrotal circumference, moderate to high for sheath depth, umbilical thickness and rosette score. However sheath score, a subjective measurement had only low to moderate repeatability. Repeatabilities for behavioural traits (apart from serves) tended to be low to moderate. However most repeatabilities for semen traits and sperm morphology were low as were correlations between times for normal sperm. Thus, a semen and sperm morphological assessment as a yearling is a poor indicator of its status as a 2-year-old.

These results indicate that with *Bos indicus* genotypes, few yearling bulls will pass a BBSE at 14 months and if even if they do, producers can expect reduced pregnancy rates if these bulls are mated as yearlings. However, limited data from Narayen indicate that this reduction in pregnancy rates will not occur if yearling Composite bulls are mated. An initial selection of bulls based on physical traits can be done as yearlings. However if bulls are being first mated as 2-year-olds, final selection of bulls should be left until then in order to evaluate behavioural and semen traits and sperm morphology.

## 9.2.2 Recommendations

# Recommendation 3. Further research may develop efficient ways to improve the proportion of Bos indicus derived bulls that pass a BBSE at 14 months of age.

There are a number of advantages of mating yearling bulls including accelerated genetic improvement and ease of handling. Less than 20% of yearling *Bos indicus* and *Bos indicus* derived bulls produce sufficient quality semen to pass a BBSE at 14 months of age. One option is to examine the impact of high-energy diets from weaning on subsequent reproductive traits and calf output.

# 9.3 Increasing the working life of bulls - Traits that predispose bulls to preputial prolapse

## 9.3.1 Conclusions

Preputial damage is a significant reason for culling *Bos indicus* and *Bos indicus* derived bulls in northern Australia.

The predisposing factors for preputial eversion may differ between Bos indicus derived breeds

and *Bos taurus* breeds. The literature shows that eversion in *Bos taurus* bulls is seen mostly in polled bulls and these polled bulls have poor development of the preputial retractor muscles. There was no evidence of poorer preputial muscle development or more preputial eversion in polled bulls than in horned bulls examined. The relationship between more preputial eversion and larger sheaths may justify selection against *Bos indicus* derived bulls with pendulous sheaths if preputial eversion leads to an increase in preputial injury or infection.

Preputial prolapse in polled Santa Gertrudis bulls could be due to the significantly reduced size and development of the preputial retractor muscle complex found in the polled bulls that were examined. No obvious cause of prolapse in the horned bulls was determined from the dissections.

From the study examining position of the penis and length of preputial eversion we concluded that preputial eversion in *Bos indicus* derived bulls was related to position of the penis.

# 9.3.2 Recommendations

# Recommendation 4. Polled bulls should be selected from herds that do not have prolapse problems and producers should avoid selecting bulls with extremely large sheaths.

Herds with polled bulls that have adequate preputial muscle development are available but these herds can be difficult to identify. Access to detailed herd records may indicate if prolapse problems are present in a herd. Bulls with large sheaths will evert more than bulls with small sheaths and if the sensitive mucosa of the prepuce is protruded further then it may have more chance of injury.

# 9.4 Qualifying the role of sperm morphology as a criterion for bull selection and calf output

## 9.4.1 Conclusions

Data from the yearling bull studies (sections 6.2.1 - 6.2.6) showed that normal sperm was lowly repeatable in bulls that had reached puberty and were undergoing sexual maturation (14 through to 24 months of age). In contrast the data from sections 6.1.4 and 6.1.5 of the relocation studies showed that the repeatability of normal sperm was high (r=0.69-0.78) in 2 to 3-year-old Brahman and Composite bulls that had reached sexual maturity. However the repeatability of normal sperm was only moderate (r=0.41) in the 2 to 5-year-old Brahman and Composite bulls at Belmont. The data sets of the relocation experiments were an ideal group to estimate repeatability as the same bulls were examined at 6-weekly intervals over a 12-month period. The data from the older Belmont bulls (Section 6.4.2) was not ideal for estimation of changes in sperm morphology because the bull population was not stable as a number of bulls were sold or introduced during the experimental period. Overall the repeatability of normal sperm was at least moderate in the mature bulls.

Pregnancy rates will be reduced if bulls with less than 50% normal sperm are single-sired mated, particularly under high mating loads.

## 9.4.2 Recommendations

# Recommendation 5. Further research will allow the estimation of the heritability of male reproductive traits and the genetic relationships between these traits and other productive traits including female fertility.

There is little information on the heritability of male reproductive traits and phenotypic and genetic correlations between male reproductive traits and other productive and female traits, particularly in tropically adapted beef cattle genotypes. There is merit in looking for new traits that might be better predictors of male fertility at both the phenotypic (bull's own performance) and genetic level (male and female progeny) than ones such as scrotal circumference that are commonly in use. Scrotal circumference is a poor indicator of calf output both in single-sire matings and multiple-sire matings Once bulls have reached threshold values for scrotal circumference, there appears to be little value in

selecting bulls for larger scrotal circumference to improve calf output. However scrotal circumference is still an important trait for selection of bulls as there is a negative genetic correlation between scrotal circumference and the female trait, days to calving. Measures of semen quality based on sperm morphology are important contributors to calf output in AI, single-sire and multiple-sire situations. It is a trait that is easily measured and it is moderate-highly repeatable in bulls once they have reached sexual maturity. There is phenotypic variation in the trait with about 35% of bulls having less than 70% normal sperm and 15% less than 50% normal sperm. If semen traits are heritable, then EBVs can be developed for these traits. This recommendation has been identified as a component of the project "Gene discovery and expression for improved female reproductive performance" in the CRC for Beef Genetic Technologies renewal application.

Recommendation 6. North Australian studies of the incidence of bulls with less than 50% and 70% normal sperm and the relationships between sperm morphology and calf output should be repeated in southern Australia in Bos taurus bulls to enhance the national adoption of sperm morphology assessment as a component of a BBSE.

# 9.5.1 The effect of herd dispersion and mating ratios on herd fertility

# 9.5.1 Conclusions

These studies showed that pregnancy rates were not reduced as a direct result of reducing bull percentages from high to moderate under high dispersion grazing management in north Australia, or from moderate to low under low or moderate herd dispersion. The data provided further support to the recommendation that mating percentages of 2.5% reproductively-sound bulls are adequate under most north Australian cattle management conditions.

# 9.5.2 Recommendations

# Recommendation 7. Herd dispersion effects need to be investigated using detailed behavioural research in controlled situations.

The herd dispersion studies have highlighted the managerial difficulties of doing these sorts of studies on commercial properties. At best the studies were observations rather than replicated research thus making the interpretation of results questionable. Even with the best intent, the researcher is at the mercy of the elements i.e. flooding, sandfly plagues, cattle being boxed etc. However, the extrapolation of results from small groups on research stations to large commercial properties is also questionable.

# 9.6 Factors influencing beef cattle libido and paternity

## 9.6.1 Conclusions

The main findings from the literature have been presented in Section 6.6.2.2. These can be summarised as follows:

Bull libido is assessed in a variety of ways, all of which determine the sexual responsiveness of bulls to females. The sexual activity of bulls is influenced by test conditions and measures of libido obtained in test situations may not reflect sexual activity in the breeding paddock.

Genotype plays a role in determining libido, and inherent fertility differences exist between individual bulls. Generally, *Bos taurus* breeds show higher and less variable levels of libido in test situations than do *Bos indicus* breeds, but this appears not to translate into fertility differences between the breeds.

Multiple males increase the expression of libido in bulls, but it is uncertain whether this translates into improvements in herd fertility. However, there are consequences for individual bull reproductive performance, as there is ample evidence of inherent differences between bulls in reproductive performance. Although data are limited, it appears that male:female ratios have minor effects on libido and fertility, although in libido tests too much competition between bulls increases aggression and

reduces sexual activity.

Social relationships between bulls can, also, affect the expression of libido, with subordinate bulls being inhibited by the presence of dominant bulls. There is evidence that dominant bulls may achieve more matings at pasture, but this is not necessarily shown in their calf output.

Older bulls show greater expression of libido in tests and appear more efficient in serving, although these changes may reflect greater sexual experience. Provided bulls are sexually mature and physically able to mate, age *per se* appears not to affect their calf output, but age interacts with dominance, which can influence calf output.

Limited research on thermal and nutritional effects indicate some adverse consequences for libido of climatic extremes for unadapted bulls and of over-feeding, but not under-feeding.

Limited research has investigated the effects on libido and fertility of multiple stressors associated with relocation; relocation to environments very different in photoperiods and temperatures has long-lasting detrimental consequences for individual bull calf output, but there is no information on the effects of relocation to less 'extreme' environments.

Too few studies have been conducted to draw conclusions about the effects of topography and herd dispersion on bull libido and calf output.

Temperament is likely to affect the expression of libido when animals are put into new situations, and highly fearful bulls appear to show low libido, although this has not been critically researched.

# 9.6.2 Recommendations

Some obvious features of the literature on cattle sexual behaviour are: its age, with the majority of the foundation work being conducted in the 1950s to 1980s; the very limited numbers of studies conducted on some aspects (often one or two); the descriptive nature of much of the work; the lack of experimental details; a paucity of robust data and the repeated reference to the same few papers. There appears to be the need for the conduct of some rigorous hypothesis testing on cattle sexual behaviour. A consequence of these features is that there is the potential that many "accepted facts" about aspects of cattle sexual behaviour are far from proven. It is probably timely to re-examine many aspects of libido to ensure that previous findings are, indeed, sound. In addition there are a number of areas that are either not explored or have given rise to equivocal results to date.

# Recommendation 8. Research into cattle reproductive behaviour may provide some valuable key concepts in developing more efficient and profitable mating management of bulls.

The following are some examples and the elucidation of these areas has the potential to have significant benefits on the efficiency of bull use and the speed at which herd genetic improvement occurs. These areas have been prioritised on a combination of likely economic impact, probability of obtaining useful data and ease of conducting the research.

#### 1 Mate choice

The apparent preference for 'like breeds' requires more work. If it is indeed the case that males and females prefer to mate with their own genotype then bull genotype needs to be carefully considered in light of the female genotype. This is unlikely to be an issue where there is a single bull, but may play a role where there are several males of differing genotypes. We do not know what other factors are involved in mate selection and this needs to be researched further. There is evidence in other species that choice is made on gene quality and compatibility between genetics, being revealed through features such as symmetry and secondary sexual characteristics. We have no idea if such systems operate in cattle. Given that producers attempt to introduce 'superior genetics' and exploit hybrid vigour in their herds by using bulls of genotypes very different to that of the females, there could be significant consequences of mate choice on improving reproductive rates of extensive beef herds.

#### 2 *Multiple matings and termination of oestrus*

There are equivocal data relating to the effects on calf output of multiple matings with the same and different bulls. Some studies report that females will accept multiple serves from the same bull, others state that females will accept serves from different males, but not repeated serves from the same male. Furthermore, there are differing opinions as to whether multiple matings improve fertility, and there are interesting observations that indicate that multiple matings reduce the length of oestrus or even terminate it. This whole area warrants further investigation given that it has the potential to have significant effects on fertility.

#### 3 Dispersion

It has been suggested that cattle dispersion (and the factors that contribute to it) will affect the ease with which bulls locate females, and, thus, influence herd reproductive rates. The emphasis in work on libido has been on that of the bull, and the role of the female in seeking the male has been given little attention. Further research is needed in the area of mate-seeking and the effects that cattle dispersion has on this.

#### 4 Courtship and the timing of mating

It has been suggested that bull courtship behaviour may actually induce oestrus in females, but there is, to date, little evidence to support this hypothesis. Also, as timing of insemination in relation to oestrus appears to be a crucial factor in males impregnating females, we need to know more about the effects of courtship and guarding by individual bulls in multiple-sire groups and whether these behaviours are related to the number of offspring produced by individual bulls. Furthermore, it appears that older, more experienced bulls may be much more adept at determining the 'optimum' timing of mating. It would be very useful to determine the mechanism by which they do this in order to improve the efficiency of calf output of bulls.

#### 5 Sperm selection and competition

Even if a bull mates with a cow, paternity is not assured due to sperm selection. Furthermore, in many species where there is mating by multiple males, sperm competition occurs in the female's reproductive tract. As we do not know if sperm selection and competition operate in cattle, these are areas that warrant further research, particularly as this would provide an indication of the relative importance of mate selection and sperm competition to calf output of a bull.

#### 6 *Pre-coital stimulation and sperm quality*

There appears to be some evidence that pre-coital stimulation of bulls may result in improvements in sperm quality and fertility. However, as this hypothesis and the data on which it is based are rather tenuous, it is an area that warrants further investigation.

#### 7 Serving capacity tests and sexual competitiveness

Bulls assessed as having high serving capacity scores (in yard tests) appear to demonstrate the same levels of sexual activity, in single-sire mating systems, as those scored as having low serving capacity scores. However, in multiple-sire mating systems, high serving capacity bulls exhibit greater levels of sexual activity than do low serving capacity bulls. Furthermore, high serving capacity bulls, also, may have a greater tendency to serve females previously served by another bull, compared to lower serving capacity bulls, but this requires confirmation. These various behaviours may be related to the social relationships between bulls, or, alternatively, serving capacity tests may actually be measuring an aspect of sexual competitiveness, rather than libido *per se*. If serving capacity tests are to be continued to be used as a predictor of bull libido then these issues need to be resolved so that tests can be refined to give more meaningful results.

# 9.7 Delivery of useful, practical information

# 9.7.1 Recommendations

# Recommendation 9. The main findings of this project should be incorporated into a saleable publication "Management of bulls in northern Australia".

The publication could be a sequel to "Bull Selection – Buying Better Bulls" and can incorporate results from the two Bullpower projects. The target audience will be beef producers. The publication will cover the measurement and importance of reproductive traits, health, puberty and sexual maturity, relocation, nutrition, bull percentages and sexual behaviour of cattle.

Recommendation 10. The importance of sperm morphology assessment in enhancing profitable use of bulls, especially in genetic improvement, indicates the need for targeted extension to achieve widespread adoption of full bull breeding soundness examinations. Key findings from this project could be included in updates of the Breeding Edge program.

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# Appendix 1 Survey Form

# Possible relocation effects on bull fertility

The **BULLPOWER** group consists of researchers from various institutions (Department of Primary Industries, University of Queensland, Royal Veterinary College; London and James Cook University) and is currently undertaking research into various aspects of bull fertility and management. Over the years we have had feedback on the effects of relocation of sale bulls on fertility and the range in criteria used by veterinary practitioners to assess breeding soundness. 'Relocation' of bulls may be to a new location in the immediate district or over long distances to a different environment. We would appreciate your input into this survey regarding your perception of the effect of relocation on bull fertility.

We would like you to read this survey form and we will complete it with you over the phone at a time convenient to you. All information is strictly confidential with no specific identification of individuals or businesses.

**Q 1**. How do you describe your business? (*please*  $\sqrt{}$ )

Insurance company

Veterinary practice

Pastoral company

**Q 2.** Do you believe there is reduced fertility of bulls following relocation? (*please*  $\sqrt{}$ )

Yes

No

Sometimes

Unsure

If you answered No to Q2, Go to Question 8;

**Q 3**. How have you formed this opinion? (*please*  $\sqrt{}$ )

Hear-say

Pre and post relocation fertility evaluations

Post relocation fertility evaluations

Q 4. In which of the following situations do you most frequently see reduced fertility? (*please* √)
Bulls relocated to a hotter district
Bulls relocated to a more humid district
Associated with outbreaks of various infectious diseases
Bulls placed on a lower plane of nutrition
Not Applicable
Other (*please specify*) .....

**Q 5**. In which breeds have you encountered depressed fertility associated with relocation? (*please*  $\sqrt{}$ )

Bos indicus

Bos indicus cross

Bos taurus

Sanga

Not Applicable

Q 6. In which category of relocated bulls is depressed fertility more prevalent? (*please* √)
Paddock reared and fed bulls
Bulls supplementary fed with grain in self-feeders
Bulls sold off crop / Oats
Bulls raised in feedlots / grain fed
No Difference
Unsure

Other (please specify).....

**Q 7**. What was the body condition score of bulls you have identified as having a fertility problem? (*please circle the number/s*)

1	2	3	4	5	6	7	8	9
Poor/emaciated			Store					Over Fat
Not Applicable								

**Q 8**. What attributes do you record in a Bull Breeding Soundness Evaluation (BBSE)? (please  $\sqrt{}$ )

Scrotal circumference

Testicular tone

Semen gross motility

Semen percent progressively motile

Semen percent normal

Mating ability

Numbers of mounts and serves

Skeletal structure

Eyes

Internal Accessory Sex Glands

I do not conduct a BBSE

**Q 9.** If semen attributes are identified as being recorded in Q8, how are they assessed? (*please*  $\sqrt{}$ )

Crush-side only

Sent to a laboratory for a detailed spermiogram

As a prepared slide / Placed in transport medium for later examination by a veterinarian

**Q 10**. Of the traits identified below, which do you believe are the important measures influencing bull fertility? (*please*  $\sqrt{}$ )

Scrotal circumference

Testicular tone

Semen gross motility

Percent progressively motile sperm

% normal sperm

Mating ability

Numbers of mounts and serves

Skeletal structure

Eyes

Internal Accessory Sex Glands

Sheath structure

Other (please specify).....

**Q 11.** What percentage of all bulls do you believe fail pre-selection and post-relocation fertility tests for the following reasons? (*please place a percentage value in the boxes for both categories*)

Pre-selection: Structural problem

Semen quality problem

Penile problem

Serving ability/capacity

Other(please specify)

Post-relocation:

Structural problem

Semen quality problem

Penile problem

Serving ability / capacity

Don't know

Other(please specify).....

Thank you for your time in completing the questionnaire.

# Appendix 2 Milestone Report 1

# Operational plan finalised by project team.

# Strategy 1. Relocation followed by depressed fertility (RDF)

The studies will more precisely define the prevalence and severity of RDF (what proportion of bulls are affected, for how long and how many recover without intervention); factors that predispose bulls to RDF and methods to manage and overcome the problem.

#### 1.1 A survey of practitioners, industry advisers, producers and breed societies.

This will be conducted in the first 18 months of the project. It will be based on the collation of informed opinion from an initial mailed survey with following up by telephone contact and, where possible, by interview. This activity will be a minor component of the project.

#### Resources

Staff: John Bertram (activity leader), Vivienne Doogan, Dick Holroyd, Geoff Fordyce.

Financial: \$2000 (operating)

Time Frame: July 2000 - June 2001

#### 1.2 Experimental studies

Studies will focus on changes in sperm morphology as the critical parameter determining presence or absence of RDF.

#### 1.2.1 Longitudinal studies on the repeatability of morphology of spermatozoa.

Studies will measure the repeatability of sperm morphology, at 1-2 month intervals, of clinically normal bulls bred on the property and thus adapted to the environment. The data will provide a benchmark for normal variability in sperm morphology of bulls adapted to and coping with a particular environment and not subject to the stressors associated with relocation. 3 cohorts of Brahman and Belmont Red bulls will be followed at Belmont.

- 30 No 2 8 bulls from January 2000 to April 2002 (Experiment 1297.99.01)
- 64 No 9 bulls from January 2000 to December 2000 (Experiment 1297.00.01)
- 70 No 0 bulls from January 2001 to December 2001.

#### Progress to date

4 examinations have been made on the older bull group and 6 examinations have been conducted on the No 9 yearling bulls.

#### Resources

Staff: Dick Holroyd (Activity leader), Carol Petherick, Bronwyn Venus

Physical: Belmont Station bulls

*Financial:* \$4000 (operating)

*Time Frame:* January 2000 to April 2002

#### 1.2.2 Effect on breeding soundness of relocating bulls after sale (Experiment 1297 BP2 05).

The studies will quantify the prevalence and severity of the effect of relocation on bull reproductive soundness. The work will be conducted on up to 25 commercial properties throughout Queensland. Bulls bought by co-operators will be monitored for up to 1.5 years from the time of purchase. Within property, bulls of different classes will be compared. Up to 4 replicates of each comparison will be made and each class will be represented by at least 4 bulls. The study will be conducted in 2 regions:

- sub tropics with about 100 bulls of 7 classes on 8 properties to be monitored
- tropics with about 200 bulls of 29 classes on 10-15 properties to be monitored.

#### Progress to date

One co-operating property, 'Flora Valley' has been identified. Further properties over the next 3-6 months will be identified in conjunction with practitioners.

#### Resources:

*Staff* Geoff Fordyce (Activity Leader), John Bertram, Dick Holroyd, Lee Fitzpatrick, Vivienne Doogan, Neil Cooper, Tim Schatz, Dallas Baker, Bronwyn Venus

Physical: Up to 25 commercial properties in north Australia

*Financial:* \$41,000 (operating)

Time Frame: July 2000 to December 2002

#### 1.2.3 Effect of relocation on bull fertility (Experiment 1297.BP2.03).

The objective of this experiment is to measure the changes in the fertility of bulls in the 12 months after their location. The design is a 2x2 factorial of 4 treatments each with 18 bulls. 80 weaner Brahman bulls will be transferred from Fletcherview Station, Charters Towers to Brigalow Research Station at Theodore in July 2000. From then until June 2001, bulls will be grazed as a common group. In July 2001, bulls will be allocated into 4 treatments: not relocated, grazing pasture only; not relocated, grazing pasture with additional grain and concentrates; relocated, grazing pasture only; relocated, grazing pasture with additional grain and concentrates. Bulls will be relocated in November 2001 to north-west Queensland.

#### Progress to date

The experiment has received approval by the TBC Animal Ethics Committee. 80 weaner bulls have been selected at Fletcherview, pending their transfer to Brigalow.

#### **Resources:**

*Staff:* Dick Holroyd (activity leader), Lee Fitzpatrick, John Bertram, Bronwyn Venus, Vivienne Doogan, Dallas Baker, Chris Coleman and Nancy Phillips

Physical: Brigalow and Toorak Research Station

Financial: \$64,000 (operating)

*Time Frame*: July 2000 to November 2002

## Strategy 2 Increasing the working life of bulls

#### 2.1 Identifying criteria whereby bulls can be selected for fertility at an early age

The objective will be firstly, to measure physical and reproductive traits in yearling bulls and link these with fertility when mated and secondly, to compare the mating outcome of bulls mated as either as yearlings or as 2-year-olds.

A longitudinal study will be conducted on 120 weaner bulls at 3 sites; 40 Santa Gertrudis at Gyranda, Theodore (Experiment 1297 BP2 06), 40 Droughtmaster bulls at Dilga, Glenmorgan (Experiment 1297 BP2 07), and 40 5/8 Brahman bulls at Swan's Lagoon (Experiment 1297 BP2 04). Bulls will be mated initially as yearlings for 2 consecutive matings and compared with bulls mated initially as 2-year-olds.

#### Progress to date

Animal ethic applications have been submitted for all 3 sites. Experimental protocols are undergoing revision. Bulls have been drafted off at Swan's Lagoon.

#### **Resources:**

*Staff:* John Bertram (activity leader), Geoff Fordyce, Lee Fitzpatrick, Viv Perry, Bronwyn Venus, Vivienne Doogan, Dallas Baker, Chris Coleman and Nancy Phillips

Physical: 'Gyranda' Theodore; 'Dilga' Glenmorgan; Swan's Lagoon Research Station

Financial: \$30,000 (operating)

*Time Frame*: June 2000 to November 2002

#### 2.2 Identifying traits that predispose bulls to premature physical breakdown

Abattoir studies will identify anatomical reasons for development of preputial problems. A survey of cooperating property managers will determine causes of breakdown and extent of bull culling. This survey has commenced in one large northern pastoral company and 2 other pastoral companies are planning to implement a survey this season. Data from these surveys will quantify the major reasons for bull wastage and give an overall perspective of the importance of this trait in bull selection. This study will be completed by March 2002.

Penile movements and resting positions in the sheath have been highlighted in the previous abattoir studies as possible factors in preputial eversion leading to an increased risk of preputial injury. A video study on live bulls is planned to observe penile movements in bulls with everting and non-everting sheaths and then compare the anatomy post-slaughter. This study will commence in the latter part of 2001.

#### Progress to date

The following activities are part of Lex Turner's MSc and have not been funded by MLA. However the studies will be used as indicators for direction of activities in NAP3.117. The preputial structures of a uniform group of 40 reproductively normal bulls have been dissected and measured. This was to establish normal variation. The preputial structures of a number of bulls with prolapses have been dissected to highlight differences between normal and prolapsed bulls to determine predisposing indicators of prolapse. Both studies are being prepared for publication.

#### Resources:

Staff: Lex Turner (activity leader), Dallas Baker, Vivienne Doogan and John Bertram

*Physical:* Various pastoral companies, Pinjarra Hills (UQ).

Financial: \$7,000 (operating)

*Time Frame*: January 2000 to June 2002

#### Strategy 3 Herd dispersion and bulls:female mating ratios

Herd dispersion refers to the way cattle spread out over their paddocks. It is modified by stocking rate, social behaviour, water availability and pasture quality and quantity. Previous work from Bullpower 1 indicated that under conditions of low to moderate dispersion, mating reproductively-sound bulls at a ratio of 2.5% of cycling females was adequate for *Bos indicus* cattle. This is to be validated under different dispersion conditions. The study will be done on 5 commercial properties in northern Australia.

#### Progress to date.

One site has been established at 'Corfield Downs', Corfield that will look at low dispersion, low bull percentages (1%) v low dispersion, medium bull percentages (2.5%). Other sites under consideration are 'Lara Downs', Richmond (2.5% v 5.0% under high dispersion) and 'Canobie', Julia Ck.

#### **Resources:**

*Staff:* Geoffry Fordyce (activity leader), Lee Fitzpatrick, Neil Cooper, Chris Coleman and Vivienne Doogan

*Physical:* 'Corfield Downs', Corfield, 4 other properties in northern Australia.

*Financial:* \$44,000 (operating)

*Time Frame*: June 2000 to November 2002

#### Strategy 4 Review of sexual behaviour of cattle

Most information on sexual behaviour relates to animals in confined areas such as serving capacity tests with little published information on the sexual behaviour of rangeland cattle. This desktop study will provide objective information on the sexual behaviour of cattle and will provide a sound basis for behaviour research if required.

#### Progress to date

A literature search has commenced.

#### Resources:

Staff: Carol Petherick (activity leader) Dick Holroyd, Geoffry Fordyce and John Bertram

Financial: \$2,000 (operating)

Time Frame: June 2000 to December 2001.

#### Strategy 5 Extension and Education activities

Widespread and coordinated extension of accurate and consistent information on reproductive management of bulls will be carried out for all sectors associated with calf production, recognising the different requirements for producers, extension officers, veterinarians and other industry-support personnel. The information will be derived both from Bullpower 1 and as results become available from this project.

#### Progress to date

A video on bull breeding soundness has been partly completed. A series of field days have been planned on 5 properties for mid July 2000.

#### **Resources:**

Staff: John Bertram (activity leader) plus rest of project team as appropriate

Financial: \$9,000 (operating)

Time Frame: June 2000 to November 2002

# **Budget Summary (operating)**

#### Strategy 1. Relocation

1.1	A survey of practitioners, industry advisers, producers and breed societies.		\$2,000		
1.2	Experimental studies				
1.2.1	Longitudinal studies on the repeatability of morphology of spermatozoa.				
1.2.2	Effect on breeding soundness of relocating bulls after sale				
1.2.3	Effect of relocation on bull fertility		\$64,000		
Strategy 2 Increasing the working life of bulls					
2.1	Identify	ring criteria whereby bulls can be selected for fertility at an early age	\$30,000		
2.2	Identifying traits that predispose bulls to premature physical breakdown		\$7,000		
Strategy 3		Herd dispersion and bulls:female mating ratios	\$44,000		
Strategy 4		Review of sexual behaviour of cattle	\$2,000		
Strategy 5		Extension and Education activities			
Total			\$203,000		

# Appendix 3 Milestone Report 4.

## Interim report on whether to proceed with following bulls pre-and post-relocation.

**Date** 30<sup>th</sup> June 2001

Achievement Criteria Endorsement by reference group

#### Progress

The evidence is increasing that relocation followed by depressed fertility (RDF) is a real problem as indicated in the following case studies and observations:

A collation of 420 examinations of bulls recently relocated from bull breeding herds in central Queensland to commercial herds in north Queensland indicated that, although there is often a lack of good data prior to relocation, about 46% of these bulls had at least 50% normal spermatozoa. This compares to about 80% of acclimatised bulls that have adequate normal sperm.

Twelve, 2-year-old bulls were relocated about 1000 km from central Queensland to north Queensland. All of these bulls, prior to relocation, had at least 50% normal sperm and mean % normals of the group were 66%. However when these bulls were examined 2 and 4 months later, only 27% and 17% of bulls had at least 50% normal sperm with mean percent normals of all bulls being 41% and 37% respectively.

We are having major problems working with industry where we can obtain sufficient numbers of the same bulls that we can access both prior to and post-relocation. It is important to have this sequential information otherwise we don't have good comparative information on the changes of sperm morphology associated with relocation. Also there is difficulty in accessing bulls frequently post-relocation in order to determine severity and recovery of RDF.

These issues were discussed at both the Bullpower team meeting 26-28<sup>th</sup> March 2001 and at the Bullpower Reference group meeting at Rockhampton, 22<sup>nd</sup> May 2001. At both of these meetings, it was agreed that the project team should scale down the effort working with industry herds and concentrate on working with experimental groups where we have control on access to bulls pre-relocation and for up to 12 months post-relocation at 6-8 weekly intervals.

Since the 22<sup>nd</sup> May meeting, one of the team members (Geoffry Fordyce) has approached 2 north Queensland bull sellers pursing the possibility of pre-sale through to post-sale evaluations with their clients. It would appear very unlikely that we will be able to achieve anything of experimental use.

Therefore the experimental studies will follow 2 sets of bulls:

Experiment 1297 BP2.03 "Effect of relocation on bull fertility". This is a 2 x 2 factorial of 4 treatments each with 18 bulls. From August 2000 to June 2001, weaner Brahman bulls (ex Fletcherview) were grazed as a common group at Brigalow Research Station. In June 2001, half of these bulls were allocated to 2 groups, supplemented with commercial bull pellets or not supplemented. We aim to have a difference of 150 kg and at least one body score difference between the 2 groups by November 2001. At this time bulls will be allocated into their 4 treatments; 1 not relocated, pasture only, 2 not relocated, pasture and supplemented, 3 relocated, pasture only and 4 relocated pasture and supplement. Bulls will be relocated to Toorak Research Station in NW Qld and the others will remain at Brigalow. Bulls in all treatments will be subjected to a physical and reproductive examination at 6-8 weekly intervals until November 2002.

42 AXBX and 38 Brahman bulls at Belmont were selected in July 2001 for use in relocation studies.

These will be allocated into 2 treatments of relocated or not relocated. The non-relocated bulls will remain at Belmont. Potential sites for the relocated bulls are Toorak or Swans or possibly a commercial property. However both relocated and non-relocated bulls must be accessed at 6-8 weekly intervals until November 2002. This is currently under negotiation within the project team and with the station managers.

RG Holroyd

Project leader NAP3.117

20<sup>th</sup> July 2001

# Appendix 4 Milestone 7

A review of the factors influencing beef cattle libido and paternity

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Milestone Report 7 for Meat & Livestock Australia

NAP3.117 Bullpower: delivery of adequate normal sperm

to site of fertilisation

November 2002

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# 1. Introduction

This review focuses on the factors that determine whether male and female beef cattle will come together and mate, making the assumption that males and females are physically capable of producing viable sperm and ova, of mating and producing offspring. The review will concentrate on the situation of freeranging cattle, where there is minimal human interference.

In this review, libido refers to sexual motivation, revealed through behaviours such as mate seeking, detection, selection and courtship, and mating and ejaculation. It is not the intention of this review to detail the neurophysiological and neuroendcrinological mechanisms of libido, but rather to review the factors that may affect whether males and females achieve mating and conception. Some factors may, indeed, reduce or eliminate libido, such as may occur when there are other conflicting motivations (eg between libido and fear, or hunger). Other factors may not affect libido per se, but rather interfere with and/or prevent its full expression, that is, animals may still be sexually motivated, but circumstances prevent some aspect(s) of sexual activity, such as mating. For example, a bull may be motivated to mate, but the presence of another bull may prevent him from doing so.

The word 'fertility' is widely used in the literature and apparently with various meanings. In this review fertility means the ability to produce progeny.

In the literature the emphasis is very much on male libido, with the behaviour and role of the female being largely ignored. As a consequence of this lack of information on females the emphasis of this review is, also, on the male and, in particular, on factors affecting male libido, as a particular aim of the review is to identify factors that could influence whether particular bulls will achieve paternity. This is important because natural mating accounts for the vast majority of pregnancies in the beef industry and there are the economic considerations of the capital costs per calf produced, which are dependent on the purchase price of the bull, its management costs, the number of calves it produces and its salvage value (McGowan et al., 2002). Another aspect of the economics relates to the genetics imparted by the bull (McCosker and Gatenby, 1993), which make a much greater contribution to accelerating the genetic improvement of herds than do females. According to McGowan et al. (2002), there are major financial gains from using bulls whose offspring have improved fertility, growth, temperament, survival and carcase attributes.

# 2. Social organisation

According to Hafez and Bouissou (1975) the usual grouping in feral cattle is a matriarchal herd derived from the continued association between mothers and female offspring. The mating system is polygynous, which means that individual males can inseminate many females. Mature males are either solitary or in so-called 'bachelor herds' that join the females only during the breeding season. Where cattle are managed by humans, such social organisations are simulated by 'controlled' breeding (ie the bulls are placed with the females for a limited period of the year). However, Hall (1989) reports that in the Chillingham White cattle (a herd of unmanaged, feral cattle in a 134 ha area of parkland in England) males and females are in association at all times and breeding occurs all year. Bulls had home ranges with each home range being shared by two or three bulls. Aggression between bulls that shared a home range was frequent, but rare between those with different home ranges. Social behaviour showed seasonal patterns and Hall (1989) proposed that maintenance behaviours, such as feeding, were the main priority during winter and, as a result, matings were less hotly contested at that time.

Within any group, cattle interact and develop relationships with one another and this leads to the rapid development of what has been termed the 'social order' or 'dominance hierarchy' (eg see Hafez and Bouissou, 1975; Fraser, 1980). This social order remains stable for long periods of time providing that the composition of the group is not changed. In the case of mature animals, the introduction of a stranger will temporarily disrupt the social order until the newcomer establishes itself in the order, but violent fights are rare (Hafez and Bouissou, 1975).

In a wide range of species, the social order is reported to determine order of access to resources and this apparently includes sexual access to females by bulls (Rupp et al., 1977; Chenoweth, 1981; Price, 1987, and see section 6.3.2). Blockey (1979) described a method of forcing bulls to compete for space in

yards that produced a similar social order as when the bulls were competing for females during mating in the paddock. In this study, Blockey found that bulls of mixed ages had a more defined social order than a group of 2 year-old bulls. The stability of the social order affected the sexual behaviour of the bulls, and the social order of a group of 2 year-old bulls was more unstable than that of the mixed age group. In the mixed age group there was a significant correlation between the social rank of an individual and sexual activity, with the most dominant animal being most sexually active. This pattern was not seen in the group of 2-year old bulls. Blockey (1979) found that social rank was determined by the length of time that the bull had been in the herd, and pointed out that in many cases this was the same as the age of the bull.

However, as will be discussed later in this review, access to females and sexual activity does not necessarily assure paternity.

## 3. Sexual behaviour

## 3.1. Seeking and detection by males

Bulls that are not actively seeking-out oestrous cows are unlikely to produce offspring. When no females are in oestrus, bulls spend most of their time on their own resting and grazing, and spend only a small proportion (3.3%) of their time investigating females (Blockey, 1978a). It is reported that bulls locate and follow cows for several days before observable (to humans) oestrus (Reinhardt, 1983; Wodzicka-Tomazewska et al., 1981). When grouped with oestrous females, bulls spend about 12% of their day in seeking females (Raadsma et al., 1983/84) and in that time appear to walk about 10-12 km/day (Raadsma et al., 1983/84: Boyd et al., 1989: Godfrev and Lunstra, 1989), However, Raadsma et al. (1983/84) found that the distance declined to about 7 km/day after the first 21 days. Although in the aforementioned studies it is not possible to distinguish between distances moved during mate seeking and those during other activities (eg grazing), Raadsma et al. (1983/84) found a significant relationship between the distance walked and the number of females mated. Other workers have, however, suggested that libido, as measured during serving capacity tests (see section 5.1.1), and locomotory behaviour are unrelated; Boyd et al. (1989) and Godfrey and Lunstra (1989) fitted bulls of high and low serving capacities with pedometers and grouped them, at pasture, with oestrous females. No difference was found between the two serving capacity groups of bulls in the distance that they moved, or in conceptions achieved.

Time spent in seeking and detection could differ with social organisation, as the figures reported by Raadsma et al. (1983/84) was in a single-sire situation, whereas in a multiple-sire situation bulls may spend about 50% of their time seeking, tending and mating during the first 21 days of joining, with a decline to about a third of their time in these activities in the following 28 days (Fritz et al., 1999).

Visual and olfactory cues appear to be of greatest importance in the detection of females by bulls (Hafez and Bouissou, 1975; Blockey, 1976a, 1980a; Geary and Reeves, 1992), although auditory stimuli are important too (Hafez and Bouissou, 1975; Arnold and Dudzinski, 1978). Blockey (1976a; 1980a) suggests that the relative importance of visual or olfactory cues depends on female activity; if there is female-female mounting then bulls will use this as a visual cue for detecting oestrous females, but if mounting is absent then bulls will sniff at the vulva to detect oestrous females. Blockey (1978a; 1980a) cited some of his work to illustrate this; in 17 of 22 observations, the bull walked to join a group of females immediately after there had been female-female mounting within the group. In four of the 22 instances the female group moved to the bull and in only one instance did the bull detect oestrus by sniffing the vulvae of the females. The findings of Mattner et al. (1974) confirm this suggestion. In contrast, Fraser (1980) states that nosing the perineum and hindquarters of females is a fairly continuous male activity.

There is evidence that the perineal skin glands of cows are a source of an oestrous pheromone (Blazquez et al., 1988) and the cervico-vaginal mucus from peri-oestrous females contains substances (pheromones) that sexually stimulate bulls (Klemm et al., 1987). However, Klemm (1989) suggested that there may be two pheromones, one for olfaction and that acts as an attractant for the bull, and one for taste or vomeronasal organ sampling that elicits a sexual response. From data obtained during serving capacity tests (see section 5.1.1), it appears that bulls do not use olfactory cues to distinguish between pre- and post-ovulatory females (Garcia et al., 1986).

The use of different senses is likely to, also, be dependent on paddock size and topography; femalefemale mounting could only serve as a visual attractant if the bull was able to see the females. Thus, bulls must use other cues (probably auditory stimuli) to initially locate females.

# 3.2. Seeking and detection by females

Mate seeking is not an activity conducted solely by males; females play an active role (Arnold and Dudzinski, 1978; Blockey, 1980a; Fraser, 1980) and will even solicit attention from a bull by licking, following and mounting him (Blockey, 1979). When in oestrus, cows become hyperactive, with the amount of time spent in eating and resting reduced and the time spent in locomotion, investigation and vocalisation is increased (Hafez and Bouissou, 1975; Fraser, 1980). Raadsma et al. (1983/84) reported that heifers in a single-sire mating system walked about 5 km/day, although it was not possible to determine what proportion of this distance was attributable to mate seeking. The social order (see section 2) between cows is ignored by oestrous cows and, as a result, agonistic interactions become more common (Hafez and Bouissou, 1975). Cow-cow mounting is seen (Fraser, 1980) and whilst non-oestrous cows will mount oestrous cows they will not tolerate being mounted. This contrasts with the oestrous cow, which will stand for mounting (Hafez and Bouissou, 1975; Blockey, 1980a).

Blockey (1978a, 1980a) describes the formation of a sexually active group (SAG), comprising oestrous and pro-oestrous females, which appears to serve as a visual attractant to bulls. However, paddock size was relatively small (2 ha) and, so, it would be easy for the bull and females to be in visual contact.

Oestrous heifers spent 97% of their time in the SAG (Blockey, 1978a, 1979) and pro-oestrous animals joined and left it, although spending long periods of time within it for several days before showing signs of oestrus (Blockey, 1978a). This author also states that the females that form the SAG congregate in a small area of the paddock and that the bull or bulls will spend a large proportion of their time with them (Blockey, 1980a). However, Blockey (1978a) also reports that the SAG is very mobile, moving, on average, 1 km/h. Most of the time (90%) only a single SAG is formed, but multiple SAGs (two or three) may form, but only when out of sight of each other (Blockey, 1979). As these studies were conducted in small paddocks (2 ha) it is not surprising that single SAGs predominated. However, multiple SAGs would be much more likely in large paddocks with uneven terrain and tree cover, and multiple SAGs would provide the opportunity to allow bulls to form associations with female groups with reduced interaction and competition between the bulls. Indeed, Rupp et al. (1977) recommended the use of paddocks with natural barriers to reduce the congregation of bulls at the same group of sexually active females. However, there is little information on the distribution of bulls and females in large paddocks and effects on fertility (see also section 6.1).

Hall found cow-cow mounting to be a rare occurrence in a herd of "wild" Bos taurus cattle inhabiting parkland in England and suggested that cow-cow mounting is an artefact of cattle husbandry (Hall, 1989). Hall (1989) found that oestrous cows were guarded by a bull against both bulls and cows. Mounting between oestrous and non-oestrous cows was also found to be rare in a group of semi-wild Bos indicus cattle because attention from multiple males tended to isolate the oestrous cow from the other cows, although pregnant cows occasionally mounted one another (Reinhardt, 1983).

## 3.3. Mate selection

Males and females of any species cooperate in order to reproduce successfully. However, the reproductive strategies of each sex are very different. For the male, the level of investment in reproducing is relatively low compared to the female. The male has only to produce sperm and deposit it in a female in attempting to pass on his genes. In contrast, females have very high levels of investment in their offspring during pregnancy and lactation. For a male, the best strategy is usually to attempt to mate with, and fertilise as many females as possible, whereas the female needs to ensure the survival of the offspring. The female's best strategy to achieve this is to ensure high quality offspring. Thus, given the opportunity, females tend to be very selective about their mating partners and try to mate with the best one available (see Trivers, 1972).

There is evidence across a range of species (although cattle are not specifically mentioned) that females select good quality males with which to mate on the basis of features such as symmetry (Thornhill, 1993)

and/or secondary sexual characteristics (Zahavi, 1975), which are reported as reflecting the quality of that male (Thornhill and Moller, 1998). For example, to grow and carry horns is energetically expensive. Thus, a bull with very large horns is demonstrating that he is of high quality because he can "afford" to have those horns.

#### 3.3.1. Male

We know next-to-nothing about mate preference and selection in beef cattle. Certainly there are observations that bulls may preferentially serve one or two females over others (Reinhardt, 1983; Garcia et al., 1986; Pexton et al., 1990; Price and Wallach, 1991). In the study by Pexton et al. (1990) the range of services given to females was one to 27, although 86% were serviced five times or less and 23.5% were serviced once only. There is apparently, also, variation in the number of bulls mounting (and possibly serving) a particular female in multiple-sire groupings. For example, Rupp et al. (1977) found, in one trial, that the proportions of heifers marked by one, two, three or four bulls were 27, 39, 28 and 6% respectively. Pexton et al. (1990) observed a large number of matings being directed to a few select females in single-sire situations when (a) there were few females in oestrus at the time the bull was introduced (b) an inexperienced (young) bull was used at the beginning of the joining period (c) a physically fatigued bull was used towards the end of the joining period or (d) a 'permissive' female was present. It is understandable that situations (a) and (d) would result in preferential mating, but the other situations suggest that inexperienced and/or bulls with low libido may not distribute matings across females. However, we are far from certain as to why such behaviours should occur (see also section 3.5).

Preference for particular females may be related to the stage of oestrus, as in the context of serving capacity tests (see section 5.1.1) using peri-oestrous females, those females that had most copulatory responses directed to them were those pre-oestrus rather than post-oestrus (Garcia et al., 1986). It is possible, also, that a female that has been mated by one bull makes her attractive (perhaps pheromonally-induced) to be served again by different bulls, although this concept has not, apparently, been examined. Such a mechanism would allow the operation of sperm selection and competition (see section 4).

There appears to be some evidence (discussed in section 6.6) that cattle have a preference to mate with members of their own breed, but we do not know how this is modified by rearing experience, or the consequences of the preferred breed being unavailable.

#### 3.3.2. Female

In relation to mating, females have long been deemed to be the passive partner. However, the focus changes markedly with our current knowledge of cryptic female choice (Eberhard, 1996) and sperm competition (Birkhead and Hunter, 1990). We now understand that females can (and do) control paternity through a number of mechanisms, and these have been reviewed by Birkhead and Moller (1993). In relation to mate selection there are two possible mechanisms: (i) copulating only with a preferred male and (ii) determining whether or not ejaculation occurs. We simply do not know whether beef cattle demonstrate mate preference in these ways, although Stookey (JM Stookey, pers. comm.) has documented a situation where a cow has determinedly avoided being separated from a group of cows and a bull by a low-ranking bull. Further, it is reported that there is large variation between females in the number of services accepted by any one bull.

Blockey (1980a) reports studies showing large individual variation between cows in the number of services accepted by any one bull; some will allow a bull to serve them once, others will stand for more than 10 services. Blockey (1980a), also, states that, once mated, a cow will not accept mating from that same bull for some unspecified period of time, but will accept being mated by a different bull. Rupp et al. (1977) found that more than 70% of females were serviced by more than one bull. We are not at all sure if these behaviours reflect preferences for particular bulls, although there seems to be some evidence that oestrus females may prefer dominant bulls (Blockey, 1979).

A study by Ancalmo and Warnick (1968) found the average number of matings per oestrous female to be 1.73, but it is not clear whether this work was conducted in a single-sire or multiple-sire situation. Reinhardt (1983) reported one to four copulations per oestrus within a few hours, but did not specify if this was from the same bull or different ones. Price (1987), on the other hand, reports that four to eight

copulations will terminate receptivity, and after serving a particular female a number of times, males seek out new females. This has the obvious advantage that a larger number of receptive females will be found and inseminated.

Whatever the mechanisms, such effects provide an opportunity for different bulls to copulate with the same cow, and for individual cows to mate with a number of bulls, both strategies having obvious consequences for the paternity of the offspring, particularly in view of the potential for sperm competition or cryptic female choice (see section 4).

# 3.4. Courtship

Once a bull has located a pro-oestrous female he will remain in close proximity to her; an activity referred to as 'guarding' (Hafez and Bouissou, 1975) or 'tending' (Fraser, 1980). Both the male and female contribute towards this association. During this time the bull 'drives' the female in various directions (Fraser, 1980). 'Nudging' is also performed, which appears to stimulate the female to stand stationary and this firm stance reciprocally stimulates the bull (Fraser, 1980). The bull will also place his chin over or on the hindquarters of the female. Raadsma et al. (1983/84) stated that more guarding and following of oestrous females occurred at night than during the day.

As the female approaches oestrus the bull will sniff and lick the area around the vulva and display the flehmen response, which involves extending the neck, raising the head and curling the upper lip (see Hafez and Bouissou, 1975). It is believed that this response draws odours through the vomeronasal organ, allowing the bull to detect specific pheromones that identify the stage of oestrus of the female (see section 3.1).

Chenoweth (1983) refers to the nuzzling, nudging and licking of the female by the bull during courtship as 'biostimulation' and suggests that this, combined with genital stimulation during intromission, can induce oestrous behaviours in the female and increase pregnancy rates. The bases for this assertion appear to be reports by other authors, and studies on artificial insemination and on dairy cows. There appear to have been no rigorous experiments to test this assertion.

During courtship, bulls will perform threat displays, although these may also be performed in the absence of females. The display involves arching of the neck and protrusion of the eyeballs, erection of the hair along the back and turning the shoulder towards the object of threat (Fraser, 1980). Bulls will also indulge in "challenging behaviour" (Fraser, 1980) that has three main aspects: roaring, pawing the ground, and horning the ground or other objects. The latter is often performed in a kneeling position. Fraser (1980) suggests that the bare earth patches and craters that result from pawing and horning are the "claiming of territory" by the bulls.

# 3.5. Mating

Providing that the female is receptive, courtship will be followed by mating (also referred to as servicing or serving). Immobility is a key characteristic that encourages serving (Blockey, 1981a; Garcia et al., 1986) and Hafez and Bouissou (1975) state that bulls are primarily attracted to the "inverted-U" shape that resembles the rear of an animal. Hence, the successful use of steers and dummies for semen collection, although it is likely that a certain amount of training and positive reinforcement (ejaculation) encourages the persistence of mounting steers and dummies by bulls. It appears that relative immobility of the female is a greater cue for eliciting mounting by Bos taurus bulls than is stage of oestrous cycle of the female, but stage of oestrous cycle may have a greater effect on the sexual response of Bos indicus x Bos taurus bulls (Price, 1987). Such differences have implications for the conduct of serving capacity tests (see section 5.1.1).

Copulation consists of a sequence of events: penile erection and protrusion from the sheath, mounting, intromission, ejaculatory thrust and ejaculation, and dismounting (Hafez and Bouissou, 1975). Hafez and Bouissou (1975) provide a description of mating behaviour: in mounting, the bull transfers his weight to his hindquarters, raises his front legs and moves forward to straddle the female with his forelegs close to the middle of the female's back. The end of the penis sways to and fro until the glans meets the vulva

and then the penis is inserted into the vagina. After further intromission the female's vulvar sphincter contracts around the penis, with the result that the bull's rectus abdominis muscle contracts suddenly. The bull's forelegs 'fix' on the female's pelvis (or more anterior) and the bull's pelvic region is rapidly brought into direct apposition to the female's genitalia and maximum intromission, leading to ejaculation, is achieved. The muscular contraction of the rectus abdominis muscle is so strong that often the bull's hind legs are drawn off the ground, giving the appearance that the bull is jumping. Intromission is performed quickly, the ejaculatory thrust is forceful with semen ejaculated as a single gush near the os cervix. The abdominal muscles then relax and the bull dismounts slowly.

Mounting and mating activity in Bos taurus cattle has been reported as occurring throughout daylight hours with peaks between 0600 to 1100h and 1800 to 2100h, with little sexual activity during the hours of darkness (Boyd et al., 1989). These findings contrast with those of Mattner et al. (1974), also working with Bos taurus animals, who found that during daylight, in single-sire mating groups, few matings occurred between 07.00 and 11.00 and most were between 14.00 to 19.00h and in multiple-sire groups, matings were evenly distributed throughout the daylight hours. However, these workers also stated that the bulls were more sexually active at night than during the day, as only 30% of mounting marks could be attributed to observed mounts and matings (those in daylight hours). Also working with Bos taurus animals, Raadsma et al. (1883/84) found the peak of mounting activity in both cows and bulls to be during the three hours after sunrise and the three hours prior to sunset, with least between 13.00 and 16.00 h. However, these workers conducted only limited observations during darkness. It is likely that the differences in the patterns of activity between these studies are due to methodological differences in conducting observations, seasonal and thermal effects, although there are reports that Bos indicus cattle are 'shy' breeders (Chenoweth, 1994) and, as a result, mate in the hours of darkness. However, as Bos indicus cattle are the predominant breed in hotter environments this observation could also result from thermal effects. Furthermore, Crichton and Lishman (1988) found no difference between the sexual activity of Bos indicus bulls during serving capacity tests conducted during the daytime and at night.

Copulations are most frequent in the early part of oestrus and become less frequent as oestrus progresses (Fraser, 1980). In serving capacity tests, pre-ovulatory cows elicited more copulatory responses from bulls than did post-ovulatory females (Garcia et al., 1986). Interestingly, these data are in contrast to the recommendations for artificial insemination, which state that best results are obtained when insemination is performed at, or near the end of oestrus (Boothby and Fahey, 1995). The reason given by these authors is that ovulation can occur two to 26 hours after the end of oestrus. Presumably this refers to overt, standing oestrus.

It has been suggested that Bos indicus bulls are more discriminating with respect to stage of oestrus than are Bos taurus bulls, which is reflected in their relative responses to oestrous or non-oestrous females during serving capacity tests (see section 5.1) (Garcia et al., 1986; Crichton and Lishman, 1988).

As would be expected, mating activity is reported to increase with the number of oestrous females (Rupp et al., 1977; Pexton et al., 1990), but also in multiple-sire groupings compared to single-sire (Mattner et al., 1974). Chenoweth (1983) cited one study in which bulls achieved an average of 55 services (range 14-101) in a 30-hour period. Variability between bulls in number of services in a 19-day pasture situation was reported as being two to 105 by Blockey (1981a), thus demonstrating the huge individual variation in libido.

The so-called 'serving capacity' of a bull is regarded as a measure of libido, and is the number of services it achieves in a paddock mating period (Blockey, 1976b). Serving capacity can be very variable, for example Blockey (1976b) reports a range of zero to 15 serves in a 7.5 h period. This serving capacity actually appears to reflect a combination of libido and mating ability in bulls. Serving capacity is reported to be highly variable between bulls (Blockey, 1976b); 1981a) and correlated with the proportion of oestrous females mounted and served (Blockey, 1976b), and achievement of first oestrous conception, but not pregnancy rate (Blockey, 1978a). However, because of the way in which these data were obtained (Blockey, 1976b) serving capacity appears not to be a useful concept. In the experiment, groups of bulls were put with groups of oestrous heifers for a 7.5 h period and the sexual activity recorded. Serving capacity was defined as the number of services a bull completed during the 7.5 h period and within a bull herd the animals were ranked on serving capacity. The upper, middle and lower one thirds were classified as high, medium and low serving capacity bull groups respectively. Serving capacity class was, therefore, a relative classification within herds only and had no meaning across

herds. This is evidenced by the data showing that in one herd the high serving capacity bulls averaged 10.5 serves compared to 5.0 in another herd, and the medium serving capacity bulls averaged 4.3 serves in one herd compared to 1.3 in another. Thus, depending upon which herd a bull that achieved four or five serves happened to be in, he could be classed as having a high serving capacity or a medium serving capacity. Thus, the large variation in serves achieved by individuals means that the concept of serving capacity is unhelpful in classifying bulls according to libido. Furthermore, it is obvious that high serving capacity bulls would mount and serve more females than low serving capacity bulls because that was the way in which the classification was defined in the study (ie bulls that did more serving were classed as having a higher serving capacity then those that did less). Females that are not served cannot conceive and, so, it is, also, obvious that, because of the way in which the classifications were defined, bulls with high serving capacities are much more likely to achieve a greater number of conceptions.

According to Blockey (1976b) the predictive value of a serving capacity test is a result of the way in which individual bulls distribute their services during paddock mating ie each bull will distribute its services equally to the oestrous females it serves and, as a consequence, a bull will rarely serve a female more than once. This certainly contradicts other findings (see section 3.3).

When the data, from this study, for the serving classes were compared across herds they showed that, for the high serving capacity bulls, a high proportion of females were served more than once (approximately 47% on average), a lesser proportion for the medium serving capacity bulls (about 21% on average) and none at all for the low bulls. These data appear to indicate that high serving capacity bulls have a greater tendency to service females that have been previously serviced by a different bull, compared to bulls with medium and low serving capacities. This finding raises the possibility that serving capacity tests are measuring "sexual competitiveness" rather than the libido of an individual bull ie that high serving capacity bulls appear to have high libidos as a result of being in a competitive situation with other bulls (see also section 5.1.1). Such a bull in a single-sire mating system may not actually have high libido. This hypothesis certainly warrants further investigation by conducting serving capacity tests with bulls in groups and alone, and then observing the number of matings and conceptions that they achieve under both multiple- and single-sire mating systems. Such a study appears not to have been conducted.

Libido would be largely academic if cows had to be served once only to achieve conception. However, Lindsay (1980) suggests that, in the field situation, cows need to mate several times, with one or more bulls, in order to accumulate sufficient semen to have a reasonable chance of conception. Blockey (1976a; 1976b; 1978a) cites work conducted in the 1950s using artificial insemination as demonstrating that repeated inseminations increased pregnancy rates. For artificial insemination it is recommended that semen be placed both in the uterus and cervix, as that placed in the uterus reaches the fertilisation site quickly, whilst that in the cervix survives longer (Boothby and Fahey, 1995). This dual placement method is said to help overcome the unpredictability of the timing of ovulation.

Chenoweth (1983) cites other work (a thesis by Farin, 1980) demonstrating that pregnancy rates increased with increased numbers of serves. Pexton et al., (1990) also found higher pregnancy rates in females served twice or more (range 55.5% to 68.6%) compared to once only (47.4%). However, Boyd et al., (1989) cited studies indicating that pregnancy rates of heifers in single-sire mating systems did not differ between those being mated once and those mated multiple times. Given that the assertion that more matings result in increased conceptions is based on limited information and data, it would seem advisable to conduct work to confirm this. Indeed, judging from the work of Rupp et al. (1977) and Raadsma et al. (1983/84), even if this is the case for individual animals, it appears that herd fertility is unaffected by service activity, regardless of whether it is by the same or different bulls.

It is reported that following ejaculation there is an unspecified period of time during which the bull and cow will not mate again. Fraser (1980) states that this appears not to be a physical effect of 'exhaustion' on the part of the bull, but rather that the female loses her stimulation value for the bull. Blockey (1980a), on the other hand, states that it results from the female not accepting another service from the same bull, but that she will accept service from a different bull within minutes of the previous one. However, in a previous paper, Blockey (1976a) states that the cow will stand to be mated by the same bull, but that the bull must encourage the female by vulva-sniffing, courtship and mounting. Given that studies in this area have given apparently conflicting results, and the evident importance of multiple matings for conception and paternity, it would seem appropriate for further work to be conducted.

The social environment can influence mating activity. Sexual activity of bulls is increased if bulls have previously watched other bulls mounting and mating (Blockey 1981b; Chenoweth, 1983; Mader and Price, 1984), but sexual activity of individuals can be inhibited by the presence of other bulls. This will be discussed in more detail in section 6.3.2.

# 4. Sperm selection and competition

Sperm selection can operate through mate selection (see section 3.3) by the female copulating only with a preferred male. The female can then select sperm through a number of mechanism: (i) determining whether ejaculation occurs (ii) ejecting sperm immediately following copulation (iii) timing copulations, in relation to ovulation, with multiple males (iv) selecting sperm within the reproductive tract and (v) post-fertilisation selective resorption or abortion of embryos (see Birkhead and Moller, 1993 for further details of these mechanisms). Thus, copulation and ejaculation does not necessarily ensure paternity. We do not have the evidence for the operation of such mechanism in cattle, but given that there is evidence for these mechanism in other mammals (including humans), it seems likely that they exist.

Additionally, sperm competition can take place. Sperm competition has been defined as the competition between sperm from different males to fertilise the eggs of a single female (Parker, 1970), and encompasses both behavioural aspects and the physiological events in the female reproductive tract. Birkhead and Hunter (1990) have reviewed the various mechanisms by which sperm competition operates. In mammals, sperm competition is compressed into the short time around oestrus, as a result of sperm remaining viable in the female reproductive tract for a very short time (usually less than 24h) and females being receptive and able to be fertilised only during the short period of oestrus. Again, we are not certain that sperm competition occurs in cattle, although studies on heterospermic insemination (ie the female receives sperm from more than one male within a short period of time near ovulation) indicate that it probably does (see section 6.6).

A number of features have evolved in species in which sperm competition occurs. These are large testes relative to body size, production of large quantities of sperm and ejaculate containing high proportions of motile sperm. During copulation males deposit a large number of spermatozoa into the female reproductive tract, but only a small proportion is retained in the tract. Roldan and Gomendio (1992) describe the ways in which sperm move through and are selected in the reproductive tract, and cite references that provide evidence of the female reproductive tract selecting against unfit sperm morphs. These authors conclude that the first sperm to reach the vicinity of the ova are the ones most likely to fertilise them. Sperm dimensions (particularly, long tails) are important in relation to sperm competition because they determine swimming velocity and may also relate to the distance the sperm must swim (Roldan and Gomendio, 1992). It appears that no morphological changes take place in sperm during their movement through the reproductive tract or capacitation (ie develop the capacity in the female reproductive tract to fertilise ova). However, at the ova (vicinity or surface) sperm undergo a change (the acrosome reaction) that assists with egg penetration (Roldan and Gomendio, 1992). Thus, an assessment of morphology of fresh ejaculate will be indicative of whether or not sperm are of appropriate morphology to attain the vicinity of the ova and penetrate them; a high proportion of normal sperm with long tails may be indicative of a high likelihood of achieving conception. Thus, sperm quality and quantity are likely to be key factors determining the fertility of a bull and, although this review is not intended to cover sperm quality and quantity in any depth, it seems appropriate to cite some work indicating whether there is support for these factors affecting bulls' ability to produce progeny.

# 4.1. Sperm quality

Coulter and Kozub (1989) found the proportion of primary sperm defects to have a negative effect on bull fertility in multiple-sire mating groups, but it was the least important factor of those included in their model. Larsen et al. (1990) investigated semen quality in four cattle breeds (Angus, Hereford, Brahman and Senepol) and found that the factors affecting conception rate in single-sire herds were percent normal (% normal), proximal droplets, detached heads and semen score (motility plus % normal). In Brahman cattle, after the effect of breeding season length had been removed, the most significant factor on conception was percent-detached heads. In Angus, motility was correlated with all reproductive performance indices (total conception rate, conception in the first 21 days, mean calving date and mean calving date of the first half of the herd to calve). Holroyd et al. (1993) also found motility of sperm and

the proportion of normal sperm to be correlated with fertility, although in later work with various cattle genotypes and multiple-sire groupings, sperm motility was related to fertility only in 5/8 Brahman cattle (Holroyd et al., 2002). However, other measures of sperm quality based on spermatozoa morphology (% normal) were important contributors to fertility in Santa Gertrudis and Brahman bulls.

Studies on heterospermic insemination demonstrated that differences in the proportion of offspring from different bulls, using frozen semen, were found to be correlated with semen laboratory tests for estimating fertility (Saake et al., 1980). Semen characteristics correlated with fertility are reported to be motility, the amount of DNA damage and number of heparin binding sites (Marshall et al., 1988).

# 4.2. Sperm quantity

The amount of testicular tissue determines the quantity of sperm produced, with relatively large testes producing more sperm on a daily basis (Wildeus and Entwistle, 1982; Entwistle, 2002). Testes weight and volume are highly correlated with scrotal circumference (Gazzola et al., 2000; Entwistle, 2002), thus scrotal circumference is an important measure of the sperm-producing capacity of a bull and it is recorded as part of a bull's fertility assessment. However, the data on the relationship between scrotal size and fertility are equivocal, although some authors specify a minimal size (eg Blockey, 1980b and Smith et al., 1981 stated that a minimum scrotal circumferences ranging between 28.9 and 44.2 cm were joined with 28 or 29 cows (Raadsma et al., 1983/84). Holroyd et al. (1993) found no correlation between scrotal circumference or testes tone and fertility in four genotypes of Bos indicus bulls in single-sire mating groups. Coulter and Kozub (1989) found that scrotal circumference positively influenced bull fertility in multiple-sire mating groups, whereas Holroyd et al. (2002), also using multiple-sire herds, found that scrotal circumference was not a major contributor to the calf output of individual bulls.

The production of large quantities of sperm does not ensure paternity unless the bull can deliver large quantities of fertile sperm at the right time, although there is evidence from heterospermic insemination studies (see section 6.6) that quantity of sperm is important; changes in the proportions of sperm from different males can overcome inherent advantages in fertility of particular males (Dzuik, 1996).

# 4.3. Timing of mating

In birds the last male to mate is the one most likely to fertilise the next ovum, but there appear no clear order effects in mammals (Gomendio and Roldan, 1993). Rather, the male most likely to fertilise the ovum is the one that times copulation so that the sperm are ready to fertilise when ovulation occurs (Birkhead and Hunter, 1990; Roldan and Gomendio, 1992). In mammals the process of capacitation can take up to 6 hours (Gomendio and Roldan, 1993). The frequency of mating would, thus, influence the chance of mating at the most appropriate time (Dziuk, 1996). Dzuik (1996) gives details of studies on a variety of species that demonstrates that timing of insemination is critical for paternity, and even appears to over-ride any inherent fertility advantages that individuals may have. There is a paucity of data for cattle, but it is likely that the same mechanisms operate.

Thus, paternity in cattle is likely to be determined by the interaction between mating order, delays between matings, timing of matings in relation to ovulation, and number and duration of matings. Evidently, mate guarding (see section 3.4) could have a considerable effect on the timing of, and delay between matings. Bulls show olfactory investigation of females in the peri-ovulatory period and pheromonal information may tell the bull when is the optimum time to mate. Thus, a bull that is guarding and checking a female may permit other bulls to copulate with the female at times that are not optimal, but may exclude them and ensure that he is the one that mates at the optimum time.

Chenoweth (1983) also suggested that 'biostimulation' (see section 3.4) induces oestrous behaviours. Thus, it could be that a combination of guarding and biostimulation allows a bull to time insemination to match ovulation. It would be useful to determine whether bulls in multiple-sire groups that conduct most courtship are also the ones that produce most progeny.

# 5. Assessment of libido

The intensity or strength of libido is something that is extremely difficult, perhaps even impossible, to assess or measure because libido may remain strong even when its expression is eliminated. For example, Hafez and Bouissou (1975) say that if it was possible to collect semen from a bull until it was physically exhausted, it would be the exhaustion that would prevent the expression of sexual behaviour, but libido itself would be unaffected. For this reason they say that libido is unrelated to the frequency of copulation. Even if libido is, technically, impossible to assess it is still essential for cattle breeding enterprises to have some indication that cattle are likely to mate. Studies indicate that some bulls that are unfit to be used as sires can be identified through physical examination, but others are only detected in tests that require bulls to mount and attempt to mate (Price, 1987; Bertram et al., 2002).

In beef cattle, formal tests for libido tend to be conducted only in males, although females may be tested to check for standing oestrus in some situations (eg for artificial insemination).

### 5.1. Male

Bull libido tests attempt to measure the sexual responsiveness of bulls to females through measures of the latency for bulls to mount and/or ejaculate (Chenoweth, 1981), counts of 'interest' (such as sniffing at the vulva, time spent with females; Chenoweth et al., 1979; Coulter and Kozub, 1989; Bertram et al., 2002) and mounts and serves during a set period of time (Blockey, 1981a; Coulter and Kozub, 1989; Bertram et al., 2002). The so-called 'serving capacity score' of a bull is generally determined in a yard with a number of restrained non-oestrous, or unrestrained oestrous females and is reported to be highly positively correlated with the number of services a bull achieves during paddock mating activity (Blockey, 1981a) and conception rate (Blockey, 1989).

Tests of libido and/or serving capacity generally take place in situations very different to those under which mating will take place. Tests are generally conducted in pens, yards or lots, usually involve two or more bulls and a small number of females. Thus, the animals are confined in a relatively small area and are unable to avoid particular individuals. The bulls are generally not tested in the same social group as will be used for mating at pasture. In most tests there is no participation from females, as they are frequently restrained and even when unrestrained, confinement in yards means that females are unable to avoid particular individuals.

Additionally, the reliability of yard serving capacity tests in demonstrating the sexual activity of bulls can be heavily influenced by test conditions. Some of these factors are the same as those that affect libido in free-ranging situation (and are discussed in more detail in section 6). Young bulls may need to be given the opportunity to learn how to mount and mate prior to testing in order that their true libido is demonstrated (Silver and Price, 1986; Boyd and Corah, 1988; Crichton and Lishman, 1988; Bertram, 1999). 'Prestimulation' of bulls, by allowing them to view mounting and mating activity, prior to the test (Blockey, 1981b; Chenoweth, 1983; Mader and Price, 1984) appears to increase libido, as does competition between bulls (Chenoweth, 1983; Mader and Price, 1984). However, the number of bulls tested together and the ratio of bulls to females can influence serving capacity as a result of high levels of aggression between bulls, and this appears to vary with different breeds of bull (Price and Wallach, 1991). Whilst non-oestrous, restrained females are generally used in yard tests, these may not be an ideal stimulus for Bos indicus bulls, and the use of oestrous, unrestrained females has been recommended for them (Price, 1987; Crichton and Lishman, 1988; Chenoweth, 1991, cited in Chenoweth et al., 2002). However, the use of restrained or unrestrained females may affect the expression of some aspects of sexual behaviour; Bertram et al. (2002) found more mounts and combined mounts and serves were made when females were restrained than unrestrained, but interest, serves and libido score were not different. Blockey (1981a) reported that the use of unrestrained females could only discriminate high serving capacity bulls, but the use of restrained females produced a high correlation between pasture and yard serving capacity. If females are restrained, the distance between them can affect the expression of libido by some bulls, as a result of 'interference' between bulls (Lane et al., 1983). It has, also, been recommended that test pens be devoid of food that may distract the bulls (Price, 1987).

Price (1987) stated that serving capacity tests should be conducted as follows: bulls should be tested individually to prevent fighting and social inhibition (see section 6.3.2), but placid beef breeds may be

tested in groups of three or four, particularly when the groups are of young, same-age males that have been reared together and a ratio of 1:1 is used for males to females. Alternatively, simultaneous individual tests could be conducted, but so that bulls can see each other mounting. Restrained, nonoestrous cows should be used, except with Bos indicus bulls, and stanchions should be 8-10 m apart to reduce interference and fighting between bulls. The test pens should contain no food or grass that may distract the bull. The bulls should be prestimulated for 10 minutes prior to testing and young animals may need several tests before their true capacity is expressed.

#### 5.1.1. Libido tests and fertility

A question frequently asked is whether bull libido affects herd fertility. This is not really the question in point, as it is obvious that bulls that are not motivated to seek and mate will never produce offspring. The question that needs to be addressed is "are libido test results related to herd fertility" or, more specifically, "does a libido test result predict the fertility of a bull"?

Given the differences between test and mating situations and the large number of factors that can affect the mating activity of bulls during yard tests it is perhaps not surprising that libido/serving capacity test results do not reliably predict the matings and conceptions achieved by bulls at pasture.

Lunstra (1986), Crichton and Lishman (1988) and Blockey (1989) reported serving capacity score to be positively correlated with conception rate under single-sire mating conditions, whilst Makarechian et al. (1985) stated that serving capacity score was "useful in assessing bull fertility". Coulter and Kozub (1989) found that, in multiple-sire mating situations, bulls with low serving capacity scores produced fewer progeny than those bulls with medium to high serving capacity scores. However, a number of other authors have reported that the relationship between serving capacity and herd fertility is inconclusive. Whilst Larsen et al. (1990) found that serving capacity score was positively correlated with percentage conception in Brahman herds, and in 2-year old Hereford bulls (but not other ages), there was no significant relationship in Angus and Senepol herds. Bertram et al. (2002) and Holroyd et al. (2002) found that there were positive relationships between some measures of sexual behaviour recorded during serving capacity tests, but none were able to consistently predict fertility in multiple-sire mating groups. Boyd et al. (1989) observed the behaviour of yearling bulls of high and low serving capacities during single-sire matings. The high serving capacity bulls performed more services, but pregnancy rates were no different between the two treatment groups and the authors tentatively suggested that the effect may have been related to pregnancy rates not differing between heifers being served just once and those being served multiple times. Similar findings were reported by Godfrey and Lunstra (1989) whose study used single and double-sire mating situations. They found that conception rates were no different between high and low serving capacity bulls in single-sire and double-sire situations. Interestingly, these workers found that high serving capacity score bulls performed more services than low in double-sire pasture tests, but there was no difference between the groups in single-sire pasture tests. This perhaps suggests that serving capacity tests may actually measure "sexual competitiveness" rather than libido per se.

Other tests of libido have produced similarly inconclusive results. For example, Smith et al. (1981) found that high libido bulls (measured as the number of heifers mated by a bull compared to the number in oestrus) did not necessarily achieve high pregnancy rates, apparently due to poor semen quality.

Blockey (1989) and Galina et al. (1992) reported studies demonstrating a low correlation between serving capacity score and pregnancy rate in Bos indicus cattle. It is suggested that the reasons for this low correlation is a reluctance of Bos indicus bulls to perform sexual behaviour in the presence of humans and, if unrestrained cows that are not fully in oestrus are used, there can be interference from them. Certainly, Bertram et al. (2002) found that Bos indicus bulls performed fewer sexual behaviours during serving capacity tests compared to Bos taurus bulls.

Price (1987) emphasises that no single variable can compare libido of individuals, and expression of adult sexual behaviour does not correlate with most of the factors that may be considered to be relevant to producing offspring eg testosterone and luteinizing hormone levels, scrotal circumference, seminal vesicle size, semen characteristics, body size and age at maturity. However, a bull with low libido will produce fewer ejaculates than one with higher libido (Hafez and Bouissou, 1975), although it has been suggested that bulls with excessive libido may rapidly exhaust sperm reserves; a third of sperm reserves

were in the first three ejaculates and <sup>3</sup>/<sub>4</sub> in the first 10 ejaculates into an artificial vagina (Rupp et al., 1977). This may explain the finding of Coulter and Kozub (1989) that bulls with moderate to high serving capacity scores produced more progeny than bulls that had very high serving capacity scores. Rupp et al. (1977) also suggested, on the basis of some of their results, that, in some bulls, excessive mating may be associated with declining fertility as a result of inadequate sperm reserves.

In conclusion it appears that serving capacity/libido tests will identify bulls that are capable of mating, but they do not necessarily assess libido, because bulls may not express their sexual motivation for a variety of reasons (see also section 6). Further, such tests certainly do not provide a reliable measure of the bull's capacity to produce offspring.

# 5.2. Female

Female libido is generally assessed through the expression of oestrus. Normally, cycling cows and heifers experience oestrus, on average, every 20 to 21 days and it lasts 13 to 14 hours in situations where females are in association with bulls (Blockey, 1980a). There is also some evidence that the duration of oestrus may be shorter in cattle kept in tropical conditions compared to temperate ones (Galina et al., 1992). Breed appears not to influence oestrous duration, but younger animals tend to have a shorter oestrus than older animals (Blockey, 1980a). However, Holroyd (1985) cites a number of references indicating that duration of oestrus is shorter in Bos indicus than Bos taurus females.

The period of receptivity is reported to be decreased by as much as 8 hours when repeated matings take place (Fraser, 1980) and behavioural oestrus is reported to be terminated after four to eight services (Price, 1987). Further, cows that are grouped with vasectomised 'teaser' bulls have a shorter period of oestrus than those grouped without teasers bulls (Fraser, 1980). Interestingly, Rupp et al. (1977) reported that heifers grouped with a bull that infrequently completed copulation remained in 'standing oestrus' for a longer duration than did those grouped with bulls that did complete copulations frequently. All of these results suggest that the stimulation of several matings acts to reduce the duration of oestrus. This makes biological sense, as once the cow has received a certain amount of sperm, from one or more bulls, it would be unnecessary for her to continue to remain attractive to bulls, but the suggestion warrants testing.

The intensity of oestrus can be assessed by measures of the amount of soliciting, mounting and standing to be mounted performed by a cow, together with appearance of the vulva and vaginal mucus. The heritability and repeatability of these measures are low and the intensity of oestrus is not directly related to conception (Hafez and Bouissou, 1975). There is also some evidence that sexual behaviour at oestrus differs between Bos taurus and Bos indicus females; more specifically, the expression of oestrus in Bos indicus animals appears less obvious and overt to human observers (Galina et al., 1992).

## 6. Factors affecting libido and conception

### 6.1. Herd dispersion

The spatial distribution of cattle is likely to restrict the expression of libido, although animals would still be sexually motivated. Thus, the extent to which cattle are dispersed or grouped within a paddock is likely to influence the ease with which bulls and cows are able to locate each other for mating. Indeed, Arnold and Dudzinski (1978) state that contact between males and females is likely to be less in extensive compared to intensive conditions, but say that this has not been studied adequately. Intuitively this would seem to be the case, as physical barriers, such as steep hills and rivers, may prevent males and females from coming together. Rupp et al. (1977) recommended the use of paddocks with physical barriers as a method for reducing 'overlap' between bulls (multiple bulls associating with the same group of sexually active females) in multiple-sire mating groups. The implication is that physical barriers within a paddock could affect the detection of oestrous females by bulls. These workers found that large groups of sexually active bulls and cows congregated in the paddock and suggested that this resulted in inefficient use of bulls, although there was the advantage that fertile bulls could compensate for any infertile ones.

In a recent study, Fordyce et al. (2002) calculated a movement range index (MRI, a measure of a bull's

"home range") for bulls in a multiple-sire herd in a 22 km<sup>2</sup> paddock of open eucalypt savannah woodland, containing numerous gullies, some hills and two main water sources. These workers found that MRI was unrelated to either social dominance or bull age. However, the bull with the highest MRI sired two to four times the average number of calves sired per bull, and those with the lowest MRI sired no more than half the average. Further, pairs of bulls were frequently found together and in each pairing one bull was considerably higher in the social order than the other and, on average, sired four times the number of calves compared to the subordinate member of the pair. When the proportion of bulls was reduced from 3.7% to 2.8% there was no change in conception pattern, indicating that the fewer bulls were able to locate oestrus cows and impregnate them, apparently by increasing their MRIs. At a proportion of 3.7% the bulls were not evenly distributed throughout the paddock, although the cows were; more bulls were located at one end of the paddock and were found with females on about only half of the observations. In contrast, the bulls at the other end of the paddock were with cows on most observations.

Studies on cattle dispersion are limited, but some are reported by Arnold and Dudzinski (1978). In one study reported, over a 5-year period, a herd of 300 cows divided into sub-groups of less than 80 animals, with the average ranging between four and 11 animals. Van Rees and Hutson (1983) reported groups of six to 12 animals grazing close together (less than 5 m apart for 50% of grazing observations). Larger groups were formed when resting at camps (20 to 40 head). A number of factors are reported by Arnold and Dudzinski (1978) as affecting sub-group size, including the quantity and quality of feed; the weather; location of watering points and the extent to which cattle graze away from them; topography; camping sites; and presence of faeces and urine. These authors state that in central Australia it is reported that cattle densities averaged zero to nine animals/km<sup>2</sup>, but following localised rainfall and fresh pasture growth, densities were up to 40 animals/km<sup>2</sup> because cattle home-in on fresh, green growth. Arnold and Dudzinski (1978) report that there are "walkers" and "non-walkers" in cattle, with the latter grazing close to watering points. Most cattle graze within 4 km of water and only go beyond this distance if pasture conditions are poor. Van Rees and Hutson (1983) demonstrated how cattle distribution is influenced by their preference for particular vegetation communities; they grazed in grassland and closed heathland, but avoided mossbeds.

#### 6.1.1. Terrain

It is claimed by Arnold and Dudzinski (1978) that double the number of bulls are required to maintain calf numbers when cattle are on rough, rocky rangeland compared to level ground. They presume that this is because the bulls have greater difficulty finding the cows. However, they appear to ignore the role of the cows in seeking the bulls and their evidence is based on a single report by Jardine and Anderson in 1919, which contains no experimental data.

### 6.2. Male:female ratios

Libido per se is unlikely to be affected by male:female ratios, but the issue is whether there is a limit to the numbers of females that bulls are able to inseminate with sufficient sperm to achieve conception.

Blockey (1976a) cites a number of studies illustrating that the ratio of bulls to cows is a critical factor in achieving conception; when the ratio was 1:20 or 1:30 95-100% of oestrous cows were mounted, but the proportion declined to about 65% when the ratio was 1:60 and declined further to 51% when it was 1:100. These results differ somewhat to those found by Rupp et al. (1977). They reported equally good oestrus detection and pregnancy rates at ratios of 1:25, 1: 44 and 1:60 (excluding two bulls that were unable to complete service) and suggested that a ratio of 1:25 was inefficient because the full breeding potential of the bull was not used. Pexton et al. (1990) also found no difference in herd fertility when bulls were single-sire mated at ratios ranging from 1:7 to 1:51 and Chenoweth (1994) states that bulls are under-utilised at ratios of 1:20 or 1:30.

Fordyce et al. (2002) compared conception patterns and rates of females in multiple-sire mating groups with bulls at 6% and 2.5%. Although pregnancy rates were lower in the 2.5% bull herd, conception patterns together with observations on pasture quality and body condition scores of the cattle indicated that the reduced pregnancies were not a result of using smaller numbers of bulls. On another site, these same workers found no effect on conception pattern with a reduction from 3.7% to 2.8% bulls (see section 6.1).

# 6.3. Social interactions between males

Social interactions can both enhance and inhibit the sexual behaviour of bulls. It is probable that it is mainly the expression of libido that is changed through these interactions. There is a problem with many studies that describe the effects of social dominance because most do not explain the method by which dominance was determined. Even when the method is given, details are inadequate to determine if appropriate techniques were used. For example, Godfrey and Lunstra (1989) measured the so-called "competitive order" of bulls when access to water was restricted, but it's not clear how this method provided information about the social relationships between the bulls. The method appears to indicate that a group of six to eight bulls were observed for 20 minutes, which would appear inadequate to determine all of the relationships. No mention is made of how interactions were scored and how animals that did not interact were ranked. It is of little wonder, therefore, that the authors concluded that the relationship between sexual activity and social rank was unclear.

Blockey (1979) described a method of forcing bulls to compete for space in a yards that produced a similar social order as when the bulls were competing for females during mating in the paddock. Blockey (1979) found that bulls of mixed ages had a more defined social order than a group of 2 year-old bulls. The stability of the social order affected the sexual behaviour of the bulls. The social order of a group of 2 year-old bulls was more unstable than that of a mixed age group.

It must, also, be remembered that the dominance hierarchy determined in a test situation may not necessarily be the same as that in the paddock mating situation, as Fordyce et al. (2002) found. These workers suggested that other factors, such as injuries to, and maturing of bulls acted to change the social order.

#### 6.3.1. Sexual activity of individuals

There are a number of reports that sexual activity of bulls is enhanced by the presence of other males (Godfrey and Lunstra, 1989; Lunstra, 1981, cited in Price and Wallach, 1991; Chenoweth, 1994). This pre-coital stimulation of bulls (eg through visual, olfactory and auditory cues associated with sexual activity) is said to improve fertility, with greatest improvement in those bulls with the lowest sperm output prior to stimulation (Hafez and Bouissou, 1975). Chenoweth (1983) cites evidence that pre-coital stimulation influences sperm motility, survival rate and conception rate, but the studies cited appear to be mainly in dairy bulls and/or work done in the 1950s. It would be useful to determine whether these findings are applicable to beef bulls and using current technology for semen collection and examination.

High serving capacity bulls perform more serves in the presence of another bull, but low do not (Godfrey and Lunstra, 1989), which may indicate that high serving capacity bulls express heightened libido in response to mounting activity, or that the presence of another male stimulates them more (see section 5.1.1). Whatever, more sexual activity per bull would be expected in multiple-male situations, regardless of the serving capacity of the bulls.

There are also reports that the presence of a more dominant male (see also section 6.3.2) can inhibit and/or interfere with the sexual behaviour of others (Price, 1987; Rupp et al., 1977; Blockey, 1979, 1980a; Crichton and Lishman, 1988; Lopez et al., 1999). In an experiment by Lopez et al. (1999) the presence (without physical contact) of a 43-month old bull did not affect the sexual behaviour of 15-month old and 27-month old bulls during a yard serving capacity test, although time spent close to the stimulus female was reduced in the 27-month old bulls. When the older bull was physically present he spent all of his time close to the stimulus female and markedly reduced the time that the younger bulls spent close to the female. Thus, the presence of an older, more dominant bull certainly has the potential to restrict access to oestrous females by subordinate males. However, it must be born in mind that under natural pasture mating conditions it is unlikely that a single female will be in oestrus at any one time. Therefore, a dominant bull would need to be able to restrict or eliminate access to a number of females in order to prevent subordinate bulls from serving them. There is some evidence that this occurs (Fritz et al., 1999), although Blockey (1979) reported that with SAGs containing more than three oestrous females the dominant bull was unable to prevent subordinates serving. However, when there were less than three oestrous females in a SAG the dominant bull disrupted the serving attempts of the subordinates. This disruption by the dominant bull was more successful (87% of occasions) with groups of mixed age bulls than with a group of 2-year old bulls (20% success).

#### 6.3.2. Effects of social dominance order

Rupp et al. (1977) found that bulls in multiple-male mating groups tended to use the same areas of the paddock and the same SAG, and that the number of females mated was affected by the social rank of the bull, with a direct positive relationship between number of females mated and dominance. These authors found that in multiple-sire mated groups, about 70% of females were mated by more than one bull, but pregnancy rates were no different in multiple-sire and single-sire groups. In another study involving multiple-sire mating of 7 bulls to about 200 cows, 75% of matings observed in the first 21 days of joining were by the dominant (and oldest) three bulls (Fritz et al., 1999). In the next 28 days, 53% of the matings were to these same bulls. Overall, 67% of the matings were to these bulls, although they checked cows for oestrus much less frequently than the other bulls (355 v 1,050). The subordinate bulls appeared to act as 'oestrus detectors', but they were then displaced by the dominant bulls. The dominant bulls, also, spent more time guarding/tending cows.

With a single SAG, the dominant bull spent more time in its proximity (91%) than subordinate bulls (53%) (Blockey, 1980a). This contrasts with the findings of Rupp et al. (1977) who found that multiple bulls, regardless of dominance rank, associated with the same SAG. Fordyce et al. (2002) found that dominant bulls spent more time with females in one breeding season, but this had not been the case in the previous breeding season. However, in pairs of bulls that consorted, dominance rank was markedly different between pair members, and the dominant bull sired an average of four times the number of calves than the subordinate bull.

Reinhardt (1983) studied the sexual behaviour of a group of semi-wild cattle in Africa over a 2.5-year period. At the start there were 29 cows, their calves and one adult bull. No cattle were removed and males were not castrated. Young male calves from 4-15 months attempted to mount and copulate but aggression from the adult bull prevented copulation. However, from 16 months of age onwards the young males achieved copulations. Dominance relationships operated in another group of 'wild' cattle that are not managed by humans at all, the Chillingham White cattle. Whenever a cow was in oestrus dominance played a role, but only bulls that shared a home range were observed tending an oestrous cow, whilst those from different home ranges were never seen to do so (Hall, 1989).

When the social order is not clearly defined, as in a group of 2-year old bulls studied by Blockey, serving capacity was reported not to be affected by social dominance (Blockey, 1978a, 1980a). Ologun et al. (1981), however, found libido and social dominance to be negatively correlated in yearling bulls. In groups of mixed aged bulls, the dominant bulls achieved more services than subordinates (Blockey 1978a, 1980a). However, there were no differences between efficiency of oestrus detection or first oestrus conception rate. The differential effects of group composition on mating are likely to result from the fact that the bulls were of a similar, young age and were probably reared together; young age and long-term cohabitation tends to minimise aggression between bulls (Price and Wallach, 1991). Also, as reported by Blockey (1979), dominance tends to be determined by length of time in the herd, which is often the same as the bull's age (see section 2.). These findings could account for some of the inconsistencies in correlations between serving capacity scores and bull fertility. Bulls could be inhibited during serving capacity tests that are conducted using multiple males and females, or if other bulls are held close by for prestimulation. Such inhibition is more likely to occur if a range of ages of bulls is used, rather than if all bulls are of similar age (see also Lopez et al., 1999 and section 6.3.1).

There is some evidence that oestrous females demonstrate a preference for dominant bulls. Blockey (1979) reported that oestrous heifers solicited attention from bulls by following, licking and occasionally mounting them. Eighty-two percent of such activities were directed to the dominant bull. Such behaviour would have obvious consequences for paternity.

In a recent study by Fordyce et al. (2002), dominant bulls sired more calves than subordinates in one breeding season, but not in the previous one. Overall, there was no relationship between dominance status and calf output in 5/8 Brahman bulls (Holroyd et al., 20002). McCosker et al. (1989) showed social dominance to be only weakly (but significantly) correlated with fertility in multiple-sire herds. The dominant bull was the most fertile in two of three paddocks, but lower order rankings were not consistent with fertility. However, to assess dominance the behaviour of the mating groups of bulls was recorded in yards for just one hour, which appears an inadequate amount of time to determine all relationships. When same-age bulls are used rather than mixed age, overall conception rate has been found to be

higher (Blockey, 1979, 1980a) and Price (1987) suggests that this is because social dominance is not completely expressed in males of approximately equal size and age. Therefore, there would be less competition between males and less potential for inhibition of libido.

Social interactions can result in physical injuries (Rupp et al., 1977) that can evidently affect the mating ability of a bull. Kilgour and Campin (1973) compared the behaviour, at pasture, of groups of bulls of different ages. Animals of 2.5 to 3.5 years of age tended to synchronise their activities and their social interactions were mainly 'amicable'. A proportion of the group of 3.5 to 4.5 year old bulls indulged in a lot of aggressive and mounting behaviours, and demonstrated signs of an increasing tendency to become territorial. In contrast, the bulls aged 5.5 to 6.5 years old had well-defined territories and were independent from other group members. The number of social interactions tended to be small and those that did take place were mainly threats and displays. Kilgour and Campin (1973) suggested that these behavioural differences explained why injuries in bulls at pasture increase after about 2.5 years of age to a peak at about 4 years of age, and then decline.

Fritz et al. (1999) reported that, in a multiple-sire mating herd, the three older, dominant bulls interacted little with each other and with the younger, subordinate bulls. However, the young bulls still attempted to mate in the presence of the older, dominant bulls, which put them at risk of injury from the older, dominant bulls. Overall, one dominant and three subordinate bulls incurred injuries, and it was assumed this was due to social interactions. Fordyce et al. (2002) found that there was greater attrition of bulls (2 to 2.5 years of age) when a high proportion of bulls (6%) compared to a low proportion (2.5%) was used. The authors presumed that the attrition was due to increased competition and fighting between bulls and gave an anecdotal report of management problems, such as mustering problems and broken fences, with more than 3.5% bulls in paddocks.

# 6.4. Age of males

Age evidently affects libido; sexual motivation will be reduced or lacking in sexually immature animals. Furthermore, age will be related to many other factors including social dominance, sexual and other experiences, and sperm quality. As described in section 6.3.2, social dominance has the greatest effect on mating success when males of different ages compete for females (Price, 1987). Older males are reported as having a greater tendency for locomotor and genital abnormalities (Price, 1987). If this is, indeed, the case then there could be dramatic consequences for herd fertility if those older bulls are less able to mate, and, also, prevent younger bulls from doing so.

Libido score was found to increase with bull age between 16 and 31 months in tropical bulls (Perry et al., 1990), and Coulter and Kozub (1989) found that yearling bulls had lower libido scores and performed more mounts than older bulls. Libido scores generally increased with age in young (12-24 months) Bos taurus bulls, but not young Bos indicus bulls, which displayed lower libido than the Bos taurus bulls (Chenoweth et al., 1996). During yard serving capacity tests, Bos taurus bulls increased mounts and serves from 4.6 and 0.1 (during a 20 minute test) to 5.4 and 1.9 between 9 and 25 months of age respectively (Bertram et al., 2000). In this same study, the mean number of mounts increased between 9 and 14 months of age, but declined thereafter, whilst the number of services steadily increased. Recently, Bertram et al. (2002) found that Santa Gertrudis and Belmont Red bulls showed an increase in the number of serves and libido score in serving capacity tests, with increasing age from 2 to 3 years old. However, the numbers of expressions of interest tended to decrease from 2 to 3 years old and the number of mounts decreased from 3 to 4+ years old. All of these results, taken together, suggest that older bulls become more 'efficient' in serving capacity tests by reducing time spent in any detection and courtship. This is probably not an effect of age per se, but rather an effect of sexual experience (see section 6.5).

There appears to be some evidence that age is correlated with the number of calves that a bull sires (Makarechian and Farid, 1985). In this study, groups of five bulls were run with cows for a 6-week breeding period at a bull:cow ratio of about 1:25. For the yearling bulls, three groups of five bulls were used, each being run with the cows for one week and rested for two weeks. Another group of mature bulls (one 3-year old and four 2-year olds) were run with a different herd of cows for the six weeks. Whilst there was little difference in the number of calves born to the yearling and mature bull groups there were differences between bulls. Amongst the mature group the 3-year old bull sired about 41% of the calves,

whilst the 2-year olds sired between 9% and 20%. In two of three yearling groups one bull sired 44% of the calves and another bull failed to sire any. In the third group there was no difference between the bulls.

Chenoweth (1981) cited South African studies that were cited by Blockey in his thesis, showing that over a 5-year period the oldest bulls sired 60% or more of calves, compared to 15% by the youngest bull. Coulter and Kozub (1989) report similar findings. In this study, bulls of 1, 2 and 3 years of age were grouped with cows under a multiple-sire management system for an average of 46.6 days. The male to female ratios ranged between 1:14 and 1:30. On average the numbers of calves sired were 4.7, 8.2 and 10.5 for bulls of 1 year, 2 years and 3 years of age respectively. In contrast, Holroyd et al. (2002) found that bull age was not a significant contributor to fertility in multiple-sire herds. However, this was probably a result of bull ages differing by only about one year in most cases (RG Holroyd, pers. comm.).

There were no differences in herd fertility when bulls ranging in age from 2 to 6 years of age were singlesire mated (Raadsma et al., 1983/84). However, Pexton et al. (1990), using single-sire mating, did find an effect on fertility of age of bull, although the only effect on sexual behaviour was that older bulls were more efficient (less mounts to serves), probably as a result of experience. Pregnancy rates achieved by bulls in three age groups were 30.2% for yearlings, 40.3% for 2-year olds and 50.7% for those 3 years of age and older (up to 7 years old). Studies in northern Australia, using single-sire herds, demonstrated that pregnancy rates were higher when 3-year old bulls were used compared to 2-year old bulls (72.8% and 66.3% respectively) (Holroyd et al., 1993).

In studies where differences in fertility were found, numerous other factors were found to change with age, such as bodyweight, sexual behaviour, scrotal circumference and semen quality. Differences in calf output may have operated through improved semen quantity and quality, as well as higher libido and serving efficiency in the older bulls. One study has indicated that semen quality improves with increased age of a bull, with 1-year old animals having more abnormal sperm than animals of 2 years of age and older (Chenoweth et al., 1984). However, this same study indicated that libido, as assessed during serving capacity tests, did not differ with age, although 1 and 2-year old animals made more mounts than older bulls. Chenoweth et al. (1984) suggested that this was due to the relative inexperience of these bulls. Bodyweight may have also played a role in relation to social dominance, but in the study by Coulter and Kozub (1989) the authors simply assumed that 3-year old bulls would be dominant to 2-year olds and they, in turn, would dominate 1-year olds (this was not actually tested). A significantly higher pregnancy rate was found in females mated to 3-year old bulls compared to those with 2-year old bulls, but the average liveweight was significantly greater in the 3-year old bulls (Holroyd et al., 1993).

Mating management may impact on whether or not age affects calf output. Larsen at al. (1990) investigated bull factors affecting conception in single-sire mating systems in 155 herds over an 8-year period. Only amongst Angus herds was there a significant (negative) correlation between bull age and the proportion of cows pregnant. These results are in contrast to those from the studies by Coulter and Kozub (1989) and Makarechian and Farid (1985) in which multiple-sire systems were used. However, Larsen et al. (1990) stated that in all herds, bulls were mostly used only as 2-year olds. These results indicate that age per se is unlikely to be the factor affecting conception, but, rather, that social interaction between bulls of different ages is the underlying cause (see section 6.3.2). This is further supported by the work of Blockey (1979) who suggested that herd fertility would be affected using bull groups of mixed ages (2 x 2-year olds and a 5-year old) compared to groups of 2-year old bulls. He found no difference between the groups in the proportion of first oestrus' detected or first oestrus conception rate. However, second oestrus detection and conception rates were significantly lower in the mixed age groups compared to the 2-year olds. After 6 weeks the groups of 2-year olds had impregnated significantly more of the females (93.8%) than the mixed age groups (84.8%). The author suggested that this was a result of the mating loads and disruption of serving by the old bull in the mixed age group leading to poor serving capacities of the mixed age bulls. Initially there was a high mating load (34 females/week) and the old bull was not able to disrupt all servings by younger bulls. Later the mating load dropped to 11 females/week and at this load the old bull was able to disrupt servings of the younger bulls, resulting in lower fertility. If this explanation is correct, then a difference between the groups in the weekly distribution of conceptions would be anticipated (a reduction in the later weeks for the mixed age groups), but there was no evidence of this.

Age may also affect the ability of bulls to achieve conception indirectly through injury (Kilgour and Campin, 1973), as discussed in section 6.3.2.

In general, the results demonstrate an age effect on fertility in multiple-sire mating groups, but not singlesire ones, and with greater effects with bull groups containing mixed ages. This suggests that the social relationships between the bulls play a key role, with the older bulls dominating the younger and, as a consequence, achieving more copulations and impregnations.

### 6.5. Sexual experience of males

According to Price (1987) the qualitative aspects of male sexual behaviour are resistant to being modified by experience, and sexual behaviour, including performance in serving capacity tests and measures of libido, of bulls appears to be unaffected by limited contact with other bulls and females during rearing (Lane et al., 1983). However, Sambraus (1980) reported that rearing bull calves with no contact with heifers resulted in the bulls in later life having a tendency to prefer to mount other bulls and avoid cows.

There is some evidence that providing sexual experience to bulls increases sexual activity. For example, Carpenter et al. (1989) found that number of services was higher in 18-month old sexually experienced Bos taurus bulls compared to 12-month old virgin bulls. Evidently age and sexual experience were confounded in this study. In another study on serving capacity tests, Bertram et al. (2002), also, found that providing virgin Santa Gertrudis bulls with sexual experience prior to serving capacity testing improved libido scores and number of serves.

The age of bulls appears to affect efficiency of mating (mounts:serves) and pregnancy rate (eg Pexton et al., 1990) and may reflect the lack of sexual experience of yearling bulls. Young beef bulls learn the correct mount orientation through mounting experience (Silver and Price, 1986). In one study of the yard serving capacity test, Bertram (1999) found that libido scores and number of services increased when 2-year old Santa Gertrudis bulls were given sexual experience prior to the test.

As stated in section 6.4, Bertram et al. (2002) found some evidence that, with increasing age, bulls showed less of the preliminary sexual behaviours, such as interest and mounts, but more serves, indicating greater efficiency of mating with age. This improved efficiency is likely to be the result of greater sexual experience. If, with increasing age, bulls have been exposed to increasing numbers of serving capacity tests then this result may due to the bulls learning (becoming trained) to "perform" in a serving capacity setting; the bulls learn that the females are unable to avoid them and so do not spend time in testing their receptivity, but just serve them. On the other hand, if bulls of different ages have experienced similar numbers of serving capacity tests then the findings are likely to have resulted from the sexual experience of the bulls gained at pasture with females. As stated in section 6.3.2, Fritz et al. (1999) found that the younger, subordinate bulls appeared to act as oestrus-detectors and they were then displaced by the dominant bulls, which mated with the females. It appears that these older and more experienced bulls may have a greater ability to determine the 'optimum' time for mating than younger, less experienced bulls, and do not need to spend time and effort determining the receptivity of the females by repeatedly mounting them. If this is, indeed, the case then it would be useful to know the mechanism by which this superior discrimination is achieved.

## 6.6. Genetics and breeds

Hafez and Bouissou (1975) suggest that libido is under genetic control, with the numbers of ejaculations and latency to ejaculate being highly repeatable in individual bulls. They also cite studies that demonstrate the heritability of libido. These same authors cite a study that demonstrates considerable individual differences between bulls in the latency to ejaculate during successive weeks, at least during weekly semen collection (Hafez and Bouissou, 1975). According to Price (1987) there is evidence of a hereditary basis for mating competence and serving capacity, and breed differences in sexual performance.

Chenoweth (1981) cites studies from the 1950s that appear to demonstrate that genetics play a large role

in determining libido; monozygous twin bulls raised on differing nutritional regimes displayed greater similarity within pairs in mating behaviour than between pairs suggesting a strong genetic influence on this trait, and paternal half-siblings of Swedish bulls differed significantly in libido score with greater variation between sire-son groups than within them. Studies showing that cross-bred bulls generally exhibited higher libido scores in pen-tests than did their parental pure-breds indicate that genetic effects influence bull libido (Chenoweth and Osborne, 1975; Perry et al., 1990).

Differences in libido scores were observed between breeding lines and sires-within-lines in young bulls of British breeds (Ologun et al., 1981). Sire strongly influenced serving capacity in young Angus bulls (Boyd and Corah, 1988). A heritability estimate of 0.59±0.16 was obtained for serving capacity in a study of 157 paternal half-sibling bull groups in Australia (Blockey, 1978b), whilst in another Australian study of 251 Santa Gertrudis, 208 Belmont Red and 189 Hereford bulls, significant heritability estimates for mounts of 0.29±0.14 and 0.57±0.25 were found across all breeds and for Santa Gertrudis bulls respectively, but serves were not demonstrated to be heritable (Wade et al., 2002).

Some authors have suggested that Zebu bulls are slower in their mounting reaction of oestrous cows compared to Bos taurus breeds (Chenoweth and Osborne, 1975; Price, 1987). Brahman bulls and cows, under single-sire management for a 60-day breeding season failed to achieve conception rates similar to Angus and Hereford herds in the study by Larsen et al. (1990). The authors, however, stated that this was not due to the Brahman bulls, but rather, longer gestations and postpartum anoestrus periods preventing the cows being in oestrus early in the breeding season.

There is evidence that, generally, Bos taurus breeds show higher and less variable levels of libido in test settings than Bos indicus (Hardin et al., 1981; Bertram, 1992; Randel, 1994; Chenoweth et al., 1996; Chenoweth, 1997). In north Australian studies, Africander bulls and their crosses achieved the highest libido scores, Brahman and Brahman crossbreds the lowest and European genotypes were intermediate (Chenoweth and Osborne, 1975; Perry et al., 1991). Similarly, increasing Brahman content resulted in lower libido scores in serving capacity tests (Bertram, 1999; Bertram et al., 2002). Chenoweth et al. (1996) suggested that the commonly-used testing procedures for libido testing may disadvantage Bos indicus bulls and, like Price (1987), suggested modifications to improve the performance of such bulls by the use of unrestrained, oestrous females and the avoidance of extraneous distractions.

Chenoweth (1994) says Zebu/Brahman bulls have low serving capacities even at pasture due to the situation and the presence of humans. However, they can achieve comparable pregnancy rates, although they display less sexual activity. According to Chenoweth (1994) these types of bull are said to be selective and "shy" breeders with a tendency to only mount females that are in full oestrus, and they generally do not perform well in pen tests to assess libido (Chenoweth et al., 1996). However, despite such observations, a comparison of trials in which bulls were mated with oestrus-synchronised females indicates that Bos indicus bulls were as efficient as Bos taurus bulls in detecting, serving and impregnating available females, despite a lower service rate (Chenoweth, 1994). Certainly it is reported that some Bos indicus bulls will show levels of sexual activity in serving capacity tests comparable to Bos taurus bulls (JD Bertram, pers. comm.).

A report by Donaldson (1962) is repeatedly referenced in the early literature as demonstrating that bulls of certain breeds do not 'compete' for mating against those of other breeds. In this study 21 Hereford and 21 Brahman cross bulls were mated with 600 Hereford cows and only 10% of the calves were Hereford x Brahman. However, when only Brahman bulls were mated with Hereford cows similar numbers of calves were produced as when Hereford bulls only were mated with Hereford cows. The assertion, that these results indicate that Brahman bulls will not compete with Hereford bulls, appears incorrect. A more likely explanation for these data is, simply, that Hereford cows have a preference for Hereford bulls as mates (see also section 3.3). Similarly, James (1979) reported that when Sahiwal bulls were mated with a mixed group of Sahiwal and Shorthorn heifers, the pregnancy rate of the Shorthorns were much lower than would be expected. This worker had observed that the bulls appeared to spend much greater lengths of time in association with the Sahiwal females compared to the Shorthorn. Similarly, Sambraus (1980) recorded that Simmental bulls from Germany were sold to another country to be joined with the native cattle of that country. He reported that many of the bulls showed no sexual interest in the cows, although the cows mounted each other. Sambraus concluded that this was a result of the bulls imprinting on (developing a preference for) their own breed during rearing. Price (1987) states that males prefer to court and mate females of the species with which they are reared, and generally this is their own species.

Indeed, Goddard (1980) raised this point of mate preference as an explanation for the low pregnancy rates reported by Reynolds (1973) for Bos indicus x Bos taurus compared to purebreds.

There are, also, inherent fertility differences between individual bulls. For example, Chenoweth (1981) cites a study showing that, in a multiple-sire mating system, the bulls that were in the top third of the siring order sired 65-100% of the calves. Osterhoff (1966), also working with multiple-sire mating herds, showed that some bulls sired 70% of the calves, whilst others sired just 4%. These inherent differences are also illustrated through studies on heterospermic insemination and the review by Dziuk (1996) provides ample evidence of these differences. These differences appear to operate through differences in the speed with which sperm reach the ova, attach and penetrate the eggs, which may be mediated through the efficiency of capacitation. Dzuik cites a number of studies on bulls demonstrating this, and also that the differences between bulls were modified depending on whether semen was fresh or frozen.

Although it is not within the bounds of this review to discuss all reproductive traits in depth, it is worth noting that there is a wealth of literature demonstrating breed differences in reproductive traits, such as sheath measurements, scrotal circumference and semen quality (eg see McGowan et al., 2002; Fitzpatrick et al., 2002; Holroyd al., 2002). Such traits are often correlated with fertility and are likely to be confounded with libido effects on fertility (Chenoweth et al., 2002).

## 6.7. Climatic/thermal environment

Extremes of climatic and/or thermal environments probably operate to reduce the expression of libido. For example, Hall (1989) suggested that during the English winter, maintenance behaviours such as feeding were the main priority for the Chillingham White cattle and as a result there was less competition for matings at that time.

Bull libido is reported to be reduced at high temperatures and this 'disinterest' appears to be due to discomfort (Arnold and Dudzinski, 1978). Holroyd (1993) reviewed some of the literature on the effects of the thermal environment and suggested that during hot periods libido is not affected in Bos indicus bulls, in contrast to Bos taurus breeds, but there may be some depression during cold periods.

### 6.8. Nutrition

Chenoweth et al. (2002) cited some studies apparently demonstrating adverse nutritional effects on bull libido, for example negative relationships between libido and average daily gain and final liveweight, and the feeding of high levels of concentrate to crossbred bulls and prolonged nursing/suckling in Angus bulls compromising the normal expression of libido. However, according to a review by Holroyd (1993), nutritional levels appear to have a minimal effect on libido, although if animals are severely underfed fertility may be impaired.

The sexual behaviour towards a teaser was tested in twin bulls, one of each pair having been underfed (30% less crude protein in the diet than the recommended amount) for a period of about 3.5 years from 18 months of age (Wierzbowski, 1978). Despite being about 235 kg less in bodyweight than their control brothers, the underfed bulls were more efficient in their sexual behaviour. Latency to mount, the interval from mounting to copulatory thrust, the interval between first and second copulations and the number of mounts per ejaculation were all significantly lower in the underfed bulls. The author suggested that these results may relate to the physical effort and forces involved in bull mounting behaviour. Although semen was collected, no data were given on the effects of underfeeding on semen quality. There is some evidence, however, that excess nutrition may adversely affect sperm reserves and quality (Entwistle and Holroyd, 1993; Miller, 1993).

### 6.9. Multiple stressors associated with relocation

There is currently little definitive information on the effects of relocation of bulls to 'stressful' environments and the relative contribution of the various stressors (eg climatic, pathogenic, parasitic and nutritional) to changes in bull libido. The problem and some potential solutions were outlined by McCool and Holroyd (1993). A large-scale study in the USA (Godfrey et al., 1990) investigated the effects when pubertal Hereford bulls from Montana and Nebraska, as well as Brahman bulls from Texas, were relocated to each other's sites of origin. No changes in reproductive parameters were directly attributed to relocation. The Brahman bulls showed seasonal changes in gonadotropin releasing hormone-induced luteinizing hormone and testosterone in both Nebraska and Montana, while the Hereford bulls showed no seasonal variation in these hormones.

## 6.10. Temperament and novelty

Cattle temperament reflects the animals' fearfulness and ability to cope with stressors, including those associated with change and novelty (Petherick et al., 2002). Although apparently referring to semen collecting situations, Hafez and Bouissou (1975) reported that some bulls become apprehensive in new situations and their sexual behaviour decreases until they become accustomed to the new surroundings. The authors say that the length of time to adapt to new situations depends upon breed and age of the bull, with younger bulls adapting faster than old ones. This has implications for the management of breeding bulls as changes of property, paddock, handlers and management procedures are highly likely to temporarily affect sexual performance.

### 7. Implications for management practices

Many of the factors detailed above will have significant effects on the ways in which cattle are managed in order to optimise fertility. Some examples are given below.

## 7.1. Conduct of serving capacity tests

Whilst the use of multiple bulls may be desirable to increase sexual activity through competition it cannot be guaranteed that some bulls may not be sexually inhibited by the presence of others. Thus, their true libido will not be revealed. Furthermore, as the social dominance hierarchy relates to specific groupings of individuals, it is likely that serving capacity tests will reveal little about the paddock sexual activity of the bulls unless the same social grouping is used in both situations.

The conduct of serving capacity tests in yards restricts or completely prevents any participation from the females and, therefore, may be unrepresentative of what occurs in the paddock situation. We do not know the effect that being tested for serving capacity has on bulls, but if bulls learn that females cannot avoid them in serving capacity tests they may dispense with courtship. If this behaviour is carried over into the paddock context and, as has been suggested, courtship induces oestrus, there could be fertility consequences for bulls that fail to show courtship in the paddock situation. Furthermore, if mate preference is normally demonstrated by cattle, then there is usually no, or very limited opportunity for animals to demonstrate this is during serving capacity tests.

Serving capacity tests may provide some indication of the libido of bulls to be used in single-sire matings, and they can certainly indicate whether or not a bull is able to mate. However, they should not be promoted and used as a method for predicting the fertility of individual bulls.

## 7.2. Single-sire v multiple-sire groups

Single-sire mating systems overcome the problems associated with social relationships between bulls, and, hence, resolve the issue of the relative proportions of offspring that particular bulls will sire. However, fertility could be drastically affected if, in a single-sire mating system, the bull is sub- or infertile. Even in a multiple-sire system there could be adverse effects on herd fertility if the dominant male is sub- or infertile, as a result of his ability to prevent subordinate bulls from mating.

### 7.3. Introducing new bulls

When bulls are introduced to new situations there is likely to be a temporary inhibition of libido, simply as a consequence of the change in environment, and regardless of any stressors resulting from relocation. Bull temperament and age are likely to affect the speed with which a bull will adapt to change and, hence, the length of time during which libido will be reduced.

If new bulls are to be put into a multiple-sire mating system then it is likely that the new bull will be the most subordinate of the group, as length of time in the herd (which is often the same as age) is correlated with social dominance. Thus, a new bull will be at the bottom of the social order and is likely to sire few calves. Bulls develop home ranges and territories, so it is inadvisable to introduce a new bull will either be breeding paddock, as resident bulls will have their territories established and the new bull will either be excluded, or have to fight resident bulls in order to establish a territory. All bulls to be mated should be introduced to the breeding paddock simultaneously to provide better opportunities for them all to establish territories.

# 7.4. Composition of bull groups

More equal sexual activity and paternity from individual bulls in multiple-sire mating herds is likely to be achieved if bull groups are young (less than 3 years), of similar age, genotype and have been reared together. There is also likely to be less aggressive interactions between such bulls and, as a result, less injuries.

### 8. Areas requiring further research

One of the most obvious features of the literature on cattle sexual behaviour is its age; the majority of the foundation work was conducted in the 1970s and 1980s, and much even pre-dates this. This is not to say that the quality of those studies is necessarily suspect, but certainly in the field of ethology, dramatic improvements have taken place in the methods, technology and conduct of studies in the last decade. These improvements are producing higher quality, and more robust data and results.

It is also noticeable in reviewing the literature that references to the same few studies persist in past reviews and when the original documents are investigated there are frequently a lack of experimental details and a paucity of robust data. There is the potential, therefore, that many "accepted facts" about aspects of cattle sexual behaviour are far from proven. It is probably timely to re-examine many aspects of libido to ensure that previous findings from a limited number of studies are, indeed, sound. In addition there are a number of areas that are, apparently, either unexplored or have given rise to equivocal results to date. The following are some examples and the elucidation of these areas has the potential to have significant benefits on the efficiency of bull use and the speed at which herd genetic improvement occurs.

### 8.1. Mate choice

The apparent preference for 'like breeds' requires more work. If it is, indeed, the case that males and females prefer to mate with their own genotype then bull genotype needs to be carefully considered in light of the female genotype. This is unlikely to be an issue where there is a single bull, but may play a role where there are multiple males of differing genotypes. We do not know what other factors are involved in mate selection and this needs to be researched further. There is evidence in other species that choice is made on gene quality and compatibility between genetics, being revealed through features such as symmetry and secondary sexual characteristics (see section 3.3). We have no idea if such systems operate in cattle. Given that producers attempt to introduce 'superior genetics' and hybrid vigour into their herds by using bulls of genotypes very different to that of the females, there could be significant consequences of mate choice on herd improvement.

## 8.2. Sperm selection and competition

Even if a bull mates with a cow, paternity is not assured due to sperm selection. Furthermore, in many species where there is mating by multiple males, sperm competition occurs in the female's reproductive tract. As we do not know if sperm selection and competition operate in cattle, these are areas that warrant further research, particularly as this would provide an indication of the relative importance to fertility of mate selection and sperm competition.

## 8.3. Courtship and the timing of mating

It has been suggested that bull courtship behaviour may actually induce oestrus in females, but there is,

to date, little evidence to support this hypothesis. Also, as timing of insemination in relation to oestrus appears to be a crucial factor in males achieving conception, we need to know more about the effects of courtship and guarding by individual bulls in multiple-sire groups and whether these behaviours are related to the number of offspring produced by individual bulls. Furthermore, it appears that older, more experienced bulls may be much more adept at determining the 'optimum' timing of mating. It would be very useful to determine the mechanism by which they do this in order to improve the efficiency of bull use.

### 8.4. Multiple matings and termination of oestrus

There are equivocal data relating to the effects on fertility of multiple matings with the same and different bulls. Some studies report that females will accept multiple serves from the same bull, others state that females will accept serves from different males, but not repeated serves from the same male. Furthermore, there are differing opinions as to whether multiple matings improve fertility, and there are interesting observations that indicate that multiple matings reduce the length of oestrus or even terminate it. This whole area warrants further investigation given that it has the potential to have very significant effects on fertility.

### 8.5. Pre-coital stimulation and sperm quality

There appears to be some evidence that pre-coital stimulation of bulls may result in improvements in sperm quality and fertility. However, as this hypothesis and the data on which it is based are rather tenuous, it is an area that warrants further investigation.

### 8.6. Dispersion

It has been suggested that cattle dispersion (and the factors that contribute to it) will affect the ease with which bulls locate females, and, thus, influence herd fertility. The emphasis in work on libido has been on that of the bull, and the role of the female in seeking the male has been given little attention. Further research is needed in the area of mate-seeking and the effects that cattle dispersion has on this.

### 8.7. Relocation of bulls

Given the high probability that bulls will continue to be purchased at considerable distances from where they will be used, there is a need to determine the effects of relocation on fertility and the relative contributions of the multiple stressors to any fertility problems.

### 8.8. Serving capacity tests and sexual competitiveness

Bulls assessed as having high serving capacity scores (in yard tests) appear to demonstrate the same levels of sexual activity, in single-sire mating systems, as those scored as having low serving capacity scores. However, in multiple-sire mating systems, high serving capacity bulls perform greater levels of sexual activity than do low serving capacity bulls. Furthermore, high serving capacity bulls, also, may have a greater tendency to serve females previously served by another bull, compared to lower serving capacity bulls, but this requires confirmation. These various behaviours may be related to the social relationships between bulls, or, alternatively, serving capacity tests may actually be measuring an aspect of sexual competitiveness, rather than libido per se. If serving capacity tests are to be continued to be used as a predictor of bull libido then these issues need to be resolved so that tests can be refined to give more meaningful results.

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