DETERMINATION OF NORMAL TESTICULAR DEVELOPMENT IN TROPICAL ADAPTED SOUTH AFRICAN BEEF BULLS DURING PHASE C (CENTRALISED TESTING CENTRE)

by

SOPHIA MAKOPI MAMPURU

MINI-DISSERTATION

Submitted in fulfilment of the requirements for the degree of

MASTER OF SCIENCE IN **AGRICULTURE**

in

ANIMAL PRODUCTION

In the

FACULTY OF SCIENCE AND AGRICULTURE

(School of Agricultural and Environmental Sciences)

at the

UNIVERSITY OF LIMPOPO

SUPERVISOR: Dr T.J Mugwabana

CO-SUPERVISOR: Prof T.L Tyasi

2023

DECLARATION

I declare that the mini dissertation hereby submitted to the University of Limpopo for the degree of Master of Science in Agriculture (Animal Production) is my independent work and research, and that it has not previously been presented as a study at this university or elsewhere. I further declare that all materials contained herein have been duly acknowledged.

Name: Mampuru Ms

Signature: ……………………………….. Date…………………………….

Dr TJ Mugwabana ……………………… Date…………………………….

(Supervisor)

Prof TL Tyasi ……………………………. Date…………………………….

(Co-supervisor)

ACKNOWLEDGEMENTS

Firstly, I would like to thank God the Almighty for making all this work possible.

I would love to also express my sincere gratitude to both my supervisors, Dr T.J Mugwabana and Prof T.L Tyasi for their patience, constructive criticism, and the support they gave me throughout the whole study.

Acknowledgements and deepest gratitude to the AGRICULTURAL Research Council (ARC) for sharing the data used in the study. I would like to appreciate my fellow postgraduate students at the University of Limpopo (UL) for all their support and guidance tirelessly. My deepest gratitude to everyone who contributed, from registration up until the end of this document.

DEDICATION

This mini dissertation is dedicated to my mother Euphrates Matseke and my grandmother Makopi Mampuru, I hope they are proud.

ABSTRACT

Bull fertility is considered the most important consideration in the herd because bulls are responsible for conception across several females. There is a high correlation between scrotal circumference and fertility in bulls, and the bigger the better. However, there is a lack of information on the normal testicular development in tropical adapted beef bulls in South African. Therefore, a study was conducted to determine the normal testicular development by age and breed in tropical adapted South African beef bulls enrolled with the INTERGIS during Phase C testing. The recorded data was sourced from the ARC and analysed using the International Business Machines (IBM) Statistical Package for the Social Sciences (SPSS), version 25. The study used thirteen beef breeds namely; Angus SA, Afrikaner, Beefmaster, Bonsmara, Brahman, Charolais, Drakensberger, Hereford, Nguni, Limousin, Santa Gertrudis, Simmental, and Sussex. The bulls were between the ages of 150 and 250 days when admitted to Phase C with a testing length of 270 days. Analysis of variance (ANOVA) and Pearson's correlation were used for data analysis. ANOVA results indicated that the Simmental bulls had the highest SC (36.00 cm) while the Brahman bulls had the lowest SC (30.00 cm). The results also showed a significant difference ($P<0.05$) between breeds on SC with the exception of the Sussex, Brahman, Santa Gertrudis, Hereford and the Nguni. The results for Pearson's correlation showed a highly significant correlation ($P<0.01$) on the breed and SC for all the breed with the exception of the Simmental and the Hereford. The breed and age of tropically adapted beef bulls had an effect on their SC. There was a significant difference between different age groups ($P<0.01$) with the exception in Nguni, Sussex and Brahman ($P>0.05$) cattle breeds.

Keywords: Scrotal circumference, Age, Breeds

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CHAPTER 1

GENERAL INTRODUCTION

1.1. Background

Agriculture is a source of income for farmers with limited resources. In Sub-Saharan Africa, majority of these farmers live in communal settings and depend primarily on livestock and crop farming systems to make a living (Thornton and Herrero, 2015). Thierfelder *et al*. (2018) demonstrated that livestock in Southern Africa is an essential component of agriculture, which significantly boosts the local economy. Livestock farming also create employment for a lot of individuals in South Africa where unemployment is one of the major socio-economic issues. Effective bull management is crucial for optimizing production performance and overall herd profitability in cattle operations. Bulls play a pivotal role in breeding programs by transmitting desirable genetic traits to offspring, influencing reproductive efficiency, and ultimately impacting the productivity and profitability of the entire herd. To Maintain a healthy herd, one must regularly check/ assess the bulls. The mating performance of the bull must also be monitored by comparing the expected and actual in-calf rates (Webb *et al*., 2023). Communal farmers can draw lessons from their commercial counterpart especially on bull management practices and their selection for use in their herd. This will help them better understand their herd and improve production thus giving them the opportunity to be more marketable. Centralised performance test can either be conducted at ARC test centres (Phase C1) and/or private test centres (Phase C2) where bulls are tested under standardised conditions for a period of 84 days following an adaptation period (Niemand, 2013).

Some of the South African native breeds include Nguni, Boran, Afrikaner, and Tuli (Abin, 2014). Most of the European breeds were introduced in South Africa during the colonial periods and other breeds from various continents were eventually introduced resulting in various crossbreeds, some of which are adapted to the environmental conditions (Bjornlund *et al.*, 2020). Due to the development of distinctive adaptation features that allow them to live and produce despite the severe conditions, regionally adapted cattle of South Africa display considerably stronger levels of resilience to such situations (Mirkena *et al*., 2010). Today, there are more than 45 identified cow breeds in South Africa (Makina *et al*., 2015). Cattlemen in the nation have long been involved in the development of productive cattle, both in the dairy and meat industries, with breed associations assisting them in achieving the greatest outcomes (Agus and Widi, 2018). Some South African cow breeds have been influenced in some manner by direct importation of breeds from the United Kingdom or Central Europe, or by crossbreeding to generate more adaptable synthetic varieties (Felius *et al*., 2015).

Breeds known as "tropical adapted" were specifically developed to survive in the hot, humid conditions found in South Africa and other tropical areas. These breeds are renowned for their capacity to withstand heat stress, immunity to tropical-environment specific illnesses, and effective forage-to-meat conversion (Bhardwaj *et al*., 2021). However, it is essential to evaluate the reproductive performance of these bulls which impacts the productivity and efficiency of beef production to ensure successful breeding. The determination of normal testicular development involves examining various aspects of the testicles and other parameters that affect the testicular development such as age and body weight. The aim is to establish baseline data regarding the normal. By establishing the normal testicular development parameters, breeders and producers can identify bulls with optimal reproductive potential. Bulls with well- developed and functional testicles are more likely to produce abundant, high-quality semen, which enhances their ability to impregnate cows and sire healthy calves (Westfalewicz *et al*., 2021). This information assists breeders in selecting superior breeding bulls, improving the genetic traits, and overall productivity of the beef cattle population.

1.2 Problem statement

Bull testing in South Africa is a function of the state and is managed by the Agricultural Research Council on behalf of the breeders' association (ARC, 2014). Smallholder beef farmers do not participate in the bull testing process. However, they can benefit from information generated under the tests (McHale *et al*., 2013). In South Africa, smallholder beef is characterised by lower calving rate compared to that realised in commercial setups. According to Musemwa *et al.* (2010) bulls reared in communal farming are of poor quality. Therefore, to improve such conditions, farmers should practice selection and be well informed about some of the factors that compromise the quality of their bulls. Under smallholder farming systems, calving rate is about 40% compared to that of commercial farming system (between 65 and 85%) (Mapiye *et al*., 2018). The low calving rate is the result of various factors including poor fertility and poor or lack of bull selections (Barth, 2018). Every bull in the herd is available for breeding including the young ones (Matsumoto, 2016). The subfertility or infertility status of the bull, largely affected by testicular development is counted among the key causes of low reproductive efficiency in smallholder beef herds (Maime, 2015). However, there is a lack of information on the normal testicular development in tropical adapted South African beef bulls such as Angus SA, Afrikaner, Beefmaster, Bonsmara, Brahman, Charolais, Drakensberger, Hereford, Nguni, Limousin, Santa Gertrudis, Simmentaler and Sussex, which will be used in the study. The bull is the most important animal in the herd because it is responsible for impregnating several cows (Kutz, 2019). There is a high correlation between scrotal circumference and fertility in bulls (Permal, 2014). Therefore, an assessment of scrotal circumference can be used to evaluate fertility in bulls. However, the most accurate method of assessing the fertility status of the bull is through microscopic semen evaluation (Naib *et al*., 2011). Lack of infrastructure, expertise, and capital to pay for these exercise renders the use of microscopic semen evaluation beyond reach by many smallholder farmers (Mutero *et al*., 2016).

1.3 Rationale

The scrotal circumference and the growth rate in the beef cattle industry are important in the selection of young bulls, therefore, affects the fertility of a bull (Šafus *et al*., 2011). In a study conducted by Permal (2014) on Tho Tho bulls, a high correlation between scrotal circumference, testicular parameters and body weight compared to age was reported. Larger scrotal circumference is characterised by more morphologically normalsperms than bulls with smaller scrotal circumference with a morphology of <70% whichcan cause infertilities (Moser *et al*., 2011). These findings are in line with those found by Coulter *et al.* (1981) that blonde d' Aquitaine breeds have a smaller scrotal circumference than Aberdeen Angus, therefore as breeding pressure increases, bullssuch as Aberdeen Angus with greater SC must be used to maintain acceptable fertility.

Fertility is the single most important trait that affects the productivity of cattle particularly under natural breeding practices (Yakubu *et al*., 2019). A fertile bull can impregnate 90% of 50 disease free normal cycling females within two months breeding period (Thundathi *et al*., 2016). The profitability of a beef enterprise and its sustainability is determined by calving percentage (Meissner *et al*., 2014). In the study conducted by Permal (2014) on Tho Tho bulls, a high correlation between scrotal circumference, testicular parameters and body weight was reported. There are various reports on the SC and its association with age, testicular growth and body weight in exotic bulls which are not adapted to South Africa such as Tho Tho bulls, Aberdeen Angus and blonde d' Aquitaine. However, studies associated with the determination of normal testicular development in tropical adapted South African beef bulls are limited. Hence this study mainly sought to determine the normal testicular development in tropical adapted South African beef bulls. This will help the farmers to better manage their herd and make thoughtful decision especially during bull selection.

1.4 Aim of the study

The aim of the study is to outline the normal testicular development in tropical adapted South African beef bulls.

1.5 Objectives of the study will be to:

i. Determine the minimum acceptable development of scrotal circumference by age, breed, and live weight in South African beef bulls during Phase C.
ii. Describe the relationship between scrotal circumference, age, and live weight in South African beef bulls during Phase C.
iii. Determine the effect of breed and age on scrotal circumference in tropical adapted South African beef bulls.

1.6 Hypotheses

i. There is no minimum acceptable development of scrotum circumference by age, breed, and live weight in South African beef bulls during Phase C.
ii. There is no relationship between scrotal circumference, age, and live weight in South African beef bulls during Phase C.
iii. The scrotum circumference in tropical adapted South African beef bulls is not affected by age and breed.

CHAPTER 2

LITERATURE REVIEW

2.1 Introduction

The cattle industry is responsible for the production of cattle for various purposes, including beef*,* hides, dairy, toilet paper (to make it soft) and nail polish remover. This industry is counted among the most important commodity in South Africa with beef industry considered as one of the fastest growing commodities in agricultural sector following the broiler sector (Meissner *et al*., 2013). The growth of the industry is attributed to income growth, adaption of new technology and structural change. The production of cattle in South Africa is predominantly practiced by communal and emerging farmers (National Livestock Statistics, 2006). However, the main challenge in communal and emerging sector is that bulls used have a scrotal circumference which is less than the acceptable threshold of 30 cm resulting in less productive/ infertile bulls (Musemwa *et al*., 2010). According to a study conducted by Maime (2015), A lot of emerging rural farmers never ask for breeding soundness evaluation or proof that the bull was negative for *B. abortus*, *T. fetus* or *C. fetus* when purchasing a bull. As a result, farmersend up purchasing bulls that are of poor quality thus subsequently affecting the herd in general. To improve the functionality of poor-quality bulls, farmers should follow andunderstand practices such as selection and breeding soundness evaluation. Animals with superior characteristics are selected and allowed to mate during selection and breeding process. Furthermore, those characteristics are then transmitted to their offspring. When this is done over a long period of time, it results in livestock improvement (Cánovas, 2016).

Fertility is the most economically important trait, and therefore a fertile bull selection makes a dramatic difference to the quality of the breeding herd, because his progeny is used for future breeding, passing on his traits (Hayes *et al*., 2013). For the bull to transmit the superior characteristics to the next generation it must first be fertile (Musemwa *et al*., 2010). Hence it is important that farmers should know and understand factors that affect fertility in bulls including testicular development. Low fertility in bulls caused by various factors not limited to abnormality in testicle and scrotum sac development of the bull as well as nutrition is known to be one of the causes of low reproductive efficiency in smallholder beef herds (Kastelic *et al.,* 2018). This study reviews the normal testicular development of tropically adapted South African beef bulls and its impact on production.

2.2 Significance of beef cattle production in South Africa

The production of beef cattle in South Africa has a rich heritage. Cattle were used in the past and even today for distinct reasons in various cultures throughout the country (Hocquette *et al*., 2018). Beef cattle are primarily reared for meat production. There are three main stages in beef production: cow-calf operations, backgrounding, and feedlot operations. The cow-calf operation is mainly designed to breed cows specifically for their offspring, which are backgrounded for a feedlot. When raised in a feedlot, cattle are known as feeder cattle. (Lusk *et al*., 2021). Tropically adapted cattle breeds, such as the Nguni breed, are predominantly found in communal and emerging farming systems. The Nguni cattle breed have been shown to have a greater capability to yield good quality meat. The Nguni cattle are well adapted to harsh conditions such as drought and they produce good quality meat that is comparable to imported breeds in a more convenient and efficient way (Köster *et al*., 2021). This attribute also suits the increased demand for organic meat by consumers worldwide (Torres *et al*., 2016). Some of the beef breeds commonly used in South Africa include Africander, Brahman, Bonsmara, Charolais, Drakensberger and etc., with different breeds being suited to different climatic conditions and areas (Buchanan and Lenstra, 2015).

Commercial farmers are estimated at 22 000 and employs 138 000 people, emerging farmers andcommunal farmers are at 3 million and employs 9 million people (Hall *et al*., 2013). There are approximately 100 commercial feedlots with 5 000 employees in South Africa and 332 abattoirs (Shirzad *et al*., 2019). Beef industry is a major contributor to livelihood with 125 000 people who are dependent on the livestock industry. This industry consists of a variety of farmers, ranging from subsistence to large commercial producers and breeders. In the large part of the country, stock farming is the only viable/ agricultural activity (Rantšo and Makhobotloane, 2020).

2.3 Testicular development in bulls

The testicles, also known as the testes or male gonads, lie behind the penis in a pouch of skin called the scrotum (Mohanty and Singh, 2017). The two testicles move freely within the scrotum, but each testicle is attached to the body wall by a thin cord called the spermatic cord, which runs through a cavity in the pelvis and into the abdomen. The testicles are outside the body and are kept at a temperature about 2°c below core body temperature. This is because sperm production and quality are optimal at this temperature (Shahat *et al*., 2020).

Testicular development starts before birth during the embryonic and fetal phases. The testes will progressively migrate to reach the scrotum. Testicular migration in bulls is completed before birth and it is divided into three stages. In the first stage namely nephric displacement, the testis stays closer to the caudal end of the abdominal cavity while the rest of the animal grows (Marani and Koch, 2014) In the second stage namely transabdominal passage the gubernaculum is pulled by some obscure forces (Miller and Torday, 2019). According to Hutson *et al.* (2013) gubernaculum doesn't move in proportion with the rest of the body and therefore the testis changes its position. The processes vaginalis develop around the posterior connection of the gubernaculum, which enlarges to create the inguinal canal, marking the end of the process, known as the "inguinal passage"(Newcomer *et al*., 2014). The testis is relocated to the inguinal hole and the gubernaculum stops growing when it is 11 weeks old. The gubernaculum expands between weeks 14 and 15, and by week 22 the testis enters the scrotum (Loof, 2020).

2.3.1 Function of the testicles

The scrotum carries testicles, provides physical protection for the testicles, and helps regulate temperature for optimum sperm production. Testes are responsible for the production of sperm and a hormone called testosterone (Rostom *et al.,* 2022). The testicles have two functions: producing sperm and producing hormones. The main hormone secreted by the testicles is testosterone (Rostom *et al.,* 2022). Testosterone is secreted by cells that lie between the testicular tubules called Leydig cells. The testicles also produce Inhibin B and Anti-Mllerian hormone, Insulin-like factor 3 and Estradiol (Tharakan *et al*., 2020).

- Testosterone: This hormone is produced in the testes, it is responsible for the development and maintenance of the reproductive tracts in males (Swelum *et al*., 2017). It plays a role in maintaining the secondary sex characteristics associated with masculinity (increases muscular and skeletal growth) and it is essential for normal sperm formation (Rajak *et al*., 2014). The rapid physical development and acceleration of weight gain during the puberty of male specimens of beef cattle breeds stems from the increase in testosterone.

Testosterone also has an influence on the behaviour of a bull and libido. However, a proper balance of all hormones is vital for a successful reproductivefunction (Syarifuddin *et al*., 2017).

- Inhibin B: This hormone Controls FSH secretion via a negative feedback mechanism. In the adult, inhibin B production depends both on FSH and on spermatogenic status, but it is not known in which way germ cells contribute to inhibin B production.
- Anti-Mullerian hormone: The growth of the internal male reproductive organs depends on this hormone.
- Insulin-like factor 3: This hormone aids in the testicles' descent from the abdomen into the scrotum and their growth there.
- Estradiol: This hormone is crucial for the production of sperm.

2.4 Factors affecting testicular development.

2.4.1 Effects of age on scrotal circumference

Age in bulls can be determined through records of birth date or estimated from dentition. Menegassi *et al.* (2019) indicated that scrotal circumference increases with 2 to 3 centimetres between one and two years of age for most breeds. Bull age has the greatest effect on testicular development in young bulls especially those that are 6 to 36 months of age (Perumal, 2014). These findings are similar to those of Holroyd *et al.* (2000) where age influenced the scrotal circumference in Santa Gertrudis and Brahman bulls. The Scrotal circumference of 2- and 3-years old Brahmans ranged from 26.0-42.25 cm with an average of 33.7cm and 34.3cm respectively. The repeatability measurements where moderate to very highly repeatable. In 2- and 3-years old Santa Gertrudis the scrotal circumference ranged from 29.0-48.0 cm with an average of 36.4 cm and 37.2 cm respectively. It was observed that testicular growth is slow for the first six months of a bovine. After this period, rapid testicular growth occurs, which is due to increase in the quantity of testicular parenchyma especially the diameter and length of seminiferous tubules, causing an increase in volume, size, and weight of the testis (Perumal *et al*., 2017).

2.4.2 Effect of breed on scrotal circumference in bulls

Taurus (European breed), and *Bos-Indicus* (zebu breed) and *Indicus* crosses on different nutritional levels. The table indicates minimum standards

Table 2.1. Guidelines for minimal scrotal circumference by breed and age (Entwistle and Fordyce, 2003)

Age (months)	**Bos-Taurus and *Bos-Indicus* cross breeds on moderate to good nutrition**	***Bos- indicus* breeds on moderate to good nutrition**	**Bulls on poor to marginal nutrition**
12 -15	30cm	24 cm	2 cm less
18	32 cm	28 cm	2 cm less
≥ 24	34 cm	30 cm	2 cm less

In a study by Fordyce *et al.* (2014) it has been indicated that breeds of *Bos-taurus* have a higher scrotal circumference than those of *Bos-indicus*. However, in a study conducted by Entwistle *et al.* (1992) Brahman (*Bos indicus*) had a higher scrotal circumference than most of the British breeds. Results showed by Entwistle and Fordyce (2003) in table 1 are similar to the results of Coulter *et al*. (1981) and Coulter *et al.* (1987) where the SC was measured on 3063 and 7918 *Bos-taurus* beef breeds, respectively. Scrotal circumference ranged from 32.1 cm (Simmental) to 37.7 cm (Limousin). Bulls aged 1 year had a Scrotal circumference that ranged from 32.1 (Simmental) to 37.7 cm (Limousin). When Coulter *et al.* (1987) compared 7918 bulls aged 2, the SC ranged from 36.3 cm (Angus,) and 32.2 cm (Limousin). These results were comparable to those noted by Menegassi *et al.* (2011).

2.4.3 Effect of nutrition on scrotal circumference in bulls

Nutrition is also one of a widely studied factor that can adversely affect seminal quality and scientists often find it difficult to quantify its effect. Kenny and Byrne (2018) stated that diets adequate in nutrients such as protein, vitamins, minerals, and energy appear to hasten the onset of puberty in beef bulls. However, Chenoweth (2021), cautions that the feeding of high-energy diets to post-pubertal beef bulls may be of no benefit to reproductive capability including seminal quality and may in fact reduce the reproductive potential. Bulls can lose a considerable amount of weight due to mating and reduced feed intake/ poor nutrition level during the breeding season (Barth, 2018). Poor nutrition can adversely affect male fertility through poor testicular development, diminished libido, reduced progressively motile spermatozoa, and increased morphological defects (Mwansa and Makarechian, 1991; Singh *et al*., 2018). Prolonged nutritional restriction can also influence the interstitial, Sertoli and Leydig cell population which changes testicular steroidogenesis and spermatogenesis (Bollwein *et al*., 2017).

2.4.4 Abnormalities and diseases of the scrotal circumference in bulls.

a. Testicular necrosis

This is usually seen in cases where regions of the testicle die off. It usually affects the entire testicle, but in some types, it can be a segmental condition (Łopuszyński *et al*., 2011). Blood flow to the testicles occurs through a tortuous testicular artery that is part of the pampiniform plexus. When the blood reaches the testicles, it is barely pulsating and has a lower pressure than normal arterial pressure. A slight change in flow leads to ischemia. Luckily this doesn't happen very often (Junior *et al*., 2018). Testicular necrosis is particularly observed in retained (or cryptorchid) testicles (Kutzler, 2022).

b. Cryptorchidism

Cryptorchidism, also known as retained testicles, is rarely reported in bulls, but the prevalence is much higher (Amann and Veeramachaneni, 2018). Testicular and epididymal retention is mainly found in the subcutaneous region and is twice as common on the left (Amann and Veeramachaneni, 2018). Polled Hereford and Shorthorn cattle are more at risk than other breeds (Young *et al*., 2020). Little is known about the pathogenesis of cryptorchidism in bulls, although it is believed to be hereditary. The histological appearance of cryptorchid testicles is similar that of hypoplastic testicles and epididymis. Neoplasia in cryptorchid testicles is very rare, with an interstitial cell tumor and a fibrolipoma reported (Osawa *et a*l., 2011).

c. Testicular hypoplasia

Testicular hypoplasia is when the testicle does not develop to its normal size. It is always accompanied by a failure of the epididymis to grow to its normal size (Neves *et al*., 2019). Most cases of hypoplasia are due to cryptorchidism, However, in this section, primary hypoplasia is limited to situations where the testicle descended normally (Fuke *et al*., 2020). Hypoplasia is almost always thought of as a lack of enlargement of the prepubertal testicle, but there can be cases where even the prepubertal testicle is smaller than normal (Ulbright *and* Young, 2014). Hypoplasia is best diagnosed clinically by noting that the testicles have not increased in size since puberty (Aycan *et al.,* 2013). Testicular hypoplasia in bulls, and in some bull breeds, is very common. Breeding fitness testing and culling of bulls with a scrotal circumference below the normal range is common. The causes are probably multifactorial. Studies suggest it is hereditary in the Swedish highland breed, where it has recessive inheritance with incomplete penetrance (Brinks *et al*., 2021). The majority of unilaterally hypoplastic testicles are on the left side. In a *Bos indicus* breed, the hereditary hypoplasia is associated with a branching of the testicular artery near the aorta on the same side, suggesting that reduced blood flow may be responsible (Rousseaux and Bolon, 2018). The most severe cases of hypoplasia have a pure Sertoli cell pattern, or they have some normal and abnormal tubules mixed, often with affected tubules being in the more dorsal part of the testes (Ghanami *et al*., 2021).

d. Testicular degeneration

The contraction and expansion of the scrotum and its muscular attachments raise and lower the testicles depending on the ambient temperature and body temperature of the bull (Sarfraz *et al*., 2019). Testicular degeneration is a process that results in the testis' structural degeneration and ultimate failure of testicular function (Barth, 2014). Normal sperm production requires maintaining the scrotal temperature at that level, which is typically lower than body temperature. The testes of mammals function normally when testis temperature is maintained at 2–6 °C less than body temperature. Anything that significantly affects this will have an impact on normal sperm production and may eventually cause testicular degeneration (Kastelic *et al*., 2018).

Severe scrotal frostbite that results in loss of testis mobility within the scrotum, incomplete descent of a testis, heat and swelling from orchitis, trauma, chronic illness that may involve prolonged fever and endotoxins that are strong inhibitors of gonadotropin secretion, are other less common causes of chronic abnormal thermoregulation that may cause temporary or permanent testicular degeneration (Barth, 2014). Obesity is a leading cause of testis degeneration in bulls. Many bulls become obese, and fat accumulates in the scrotum leading to chronic abnormal thermoregulation with increased testicular temperatures (Liu *et al*., 2014)

e. Scrotal/testicle shape

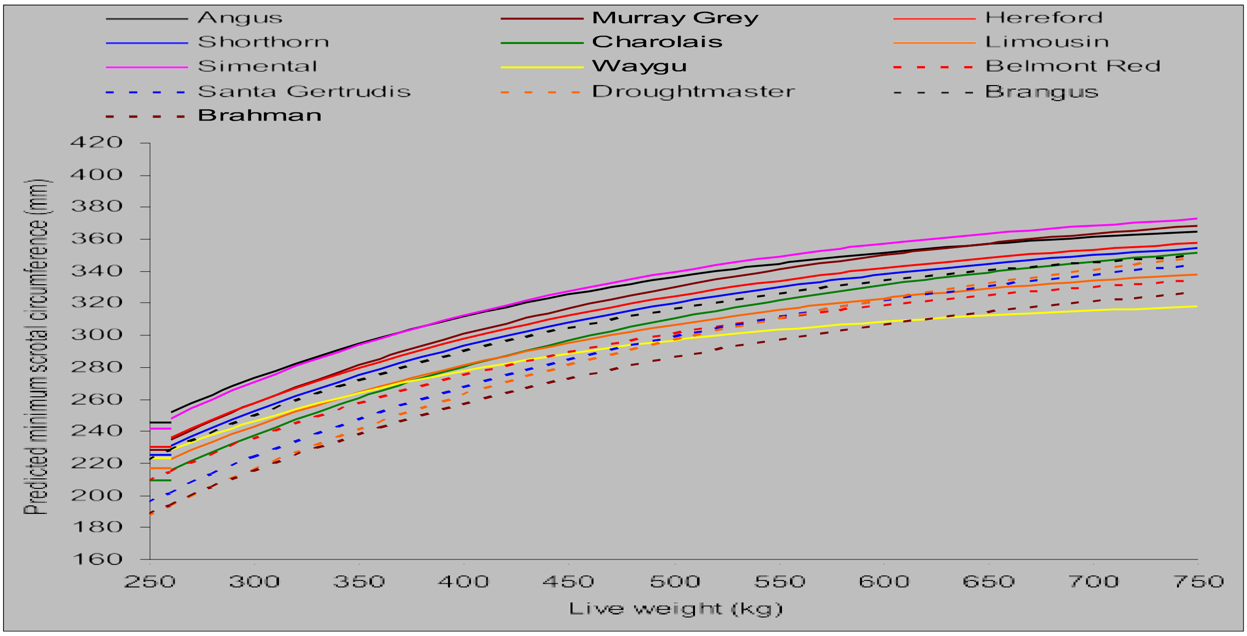
Abnormal testicle and scrotal sac development are also considered as some of the common cause of low fertility in bulls. The testicles should be symmetrical, nearly the same size, and freely movable in the scrotum. There are three basic shapes in beef bulls. These are the "normal" or "bottle-shaped" scrotum "straight-sided" scrotum, and "wedge-shaped" scrotum. Bulls having a normal scrotum with a distinct neck generally have the best testicular development (Gressler *et al.,* 2000). The normal scrotum offers the best opportunity for temperature control of the testicles. Often bulls with straight-sided scrotums are only moderate in testicle size. The straight-sided neck of the scrotum is generally the result of fat deposits that may impair proper thermoregulation. Wedge-shaped scrotums are pointed toward the bottom and hold the testicles close to the body wall. Bulls with this scrotal configuration have undersized testicles and seldom produce semen of adequate quality (Thundathil *et al*.,2016).

2.5 The relationship between body weight, age, serving capacity, motile spermatozoa, and scrotal circumstance.

Body weight can be determined using a weighing scale. According to a study conducted by Perumal (2014) it has been indicated that in a scenario where the bulls are exposed to the same environmental conditions and given the same feeds, a positive correlation between testicular weight and age was found and the estimated average testicular weight ranging from 18,50-51,25% greater in bulls that range from the age of 25 to >49 months compared to bulls aged 18-24 months old. As a general principle, it is recommended that the minimum acceptable scrotal circumference is the bottom 5% value at any weight (Bonilha *et al.*, 2008).

In a study conducted by Holroyd *et al.* (2000) 2 years old Santa Gertrudis bulls hada body weight which was positively correlated with the number of service and the percentage of motile spermatozoa. The number of services achieved in the serving capacity test showed a very weak positive correlation(r=0.13). The correlation between the percentage of normal motile spermatozoa and body weight was weak(r=0.27). These results were similar to those of Fordyce (2013) where a weak positive correlation between scrotal circumference and percentage of normal spermatozoa was found in 2 years old Santa Gertrudis. However, a study conducted by Perumal (2014) indicated that body weight was highly correlated withthe various testicular measurements than age. Most previous reports have suggested age as the reference for assessing whether bull scrotal circumference is within normal range (Menegassi *et al*., 2019). However, Fordyce *et al.* (2014) showed that the preferred reference point is liveweight because a wide range in nutritional experiences of young bulls cause marked variation in growth. Fordyce *et al.* (2014) showed normal ranges for scrotal circumference where the breeds arevalid in the 250- to 750-kg range.

Figure 2.1: Liveweight effects on average scrotal circumference (mm) (90%



confidence interval) as predicted by an asymptotic model (Fordyce *et al*., 2014).

2.6 Importance of measuring scrotal circumference

Measuring scrotal circumference can indicate weather testicular development is within the recommended range (Loaiza *et al*., 2013). In order for a bull to impregnate a cow it must first reach puberty, which occurs when the bull can produce viable sperm (Thundathil *et al*., 2016). Therefore, measuring the scrotal circumference can make it easy for one to tell if a bull has reached puberty. According to Kastelic (2014), there isa correlation between SC and earlier age at puberty in heifers sired by that particular bull. However, puberty can also be influenced by nutrition, breed, and weight. Heifersthat conceive early during their first breeding season are more efficient and can have increased lifetime production due to the number of estrus cycles that the heifer will experience (Day and Nogueira, 2013). Increasing efficiency to calve at the targeted age range minimizes excessive days on feed thus improving the profit. SC was foundto be genetically correlated with earlier return to cycling in female relatives within tropically adapted cattle (Bonamy *et al*., 2018). According to Chacón (2014), it was indicated that SC is correlated with sperm motility and morphology, these are independently assessed as part of the BBSE.

2.7 Bull breeding boundness evaluation (BBSE)

This is a method of assessing the bull's likelihood of impregnating 25-30 open, healthy, cycling cows or heifers in a defined breeding season (Lamb *et al*., 2016). It involves four components used to assess the fertility in bulls namely: physical exam, scrotal circumference, sperm quality and serving capacity (Susilawati *et al*., 2020). The physical examination of the reproductive tract includes evaluation of the testes, spermatic cords, and epididymis. Scrotal circumference is directly related to the sperm producing tissue, sperm cell normality and the onset of puberty in the bull and his female offspring (Kastelic, 2014). Increased scrotal circumference is associated with earlier age at puberty, increased semen production and improved semen quality (Brinks, 2021). As a result, its assessment is an important part of a Veterinary Bull Breeding Soundness Evaluation (VBBSE), it is used to assess whether bulls' reproductive function is normal before deciding on the selection and management process (Paul, 2012). There is also evidence that increased scrotal circumference is associated with improved female fertility and earlier age at puberty in a bull's daughters (Boerner *et al*., 2015). Examination of the external factors before and during semen collection will detect any inflammation, foreskin adhesions, warts, abscesses, and penile deviations.

The internal examination is conducted to detect any abnormalities in the internal reproductive organs. The evaluation of scrotalcircumference involves examining the scrotal circumference, as well as estimating thebull's sperm-producing capacity (Russell *et al*., 2021). However, according to Lone *etal*. (2017) bulls must meet minimum scrotal circumference measurements in order to pass a BBSE.

Table 2.2. Minimum requirements of the scrotal circumference of bulls by age (adopted by Gosey, 2019)

Age in months	≤ 15	15 − 18	18 − 21	21 −24	≥ 24
SC (cm)	30	31	32	33	34

Table 2.3. Minimum requirements of the scrotal circumference of by body weight (adopted by Byrne, 2013)

Live weight (Kg)	300	400	500	600	700	800	900
SC (cm)	27.5	31.0	33.5	35.0	36.0	36.5	37.0

2.8 Scrotal circumference and bull fertility

A deficiency in a bull has a larger impact on herd productivity than fertility problems in a single female, hence a bull is considered to be half the herd. When AI is used, each ejaculate can produce 300 inseminations, representing at least 60,000 doses per bull per year (Hossain *et al*., 2020). Therefore, it is extremely important in selection of breeding bulls to consider fertility. Fertility can be determined through evaluation of sperm motility and sperm morphology and measuring the scrotal circumference. A fertile bull usually constitutes at least 30 per cent progressively motile spermatozoa with 70 percent properly shaped spermatozoa (Hammond *et al.*, 1996). In yearling bulls, researchers observed that as scrotal circumference increase, motility, sperm concentration, and sperm output increase (Kushwaha *et al*., 2018). Young bulls with higher scrotal circumference produce greater semen volume with more spermatozoa during their first ejaculation (Henry and Torres-Júnior, 2018). However, SC is a trait that may be affected by various factors such as genetic, environmental, or individual bull differences (Polizel *et al*., 2021).

2.9 The relationship between scrotal circumference/testicular development and testosterone.

A strong positive correlation between testosterone concentration and number of sperm (r=0.703), SC (r=0,799) as well as sperm motility (r=0.857) was observed in a study conducted by Dasrul *et al*. (2020). Furthermore, there was a strong negative correlation (r=-0,877) between testosterone concentrations and sperm abnormalities. Similarly, a positive correlation between testosterone level and scrotal circumference (r=0.414) and ejaculatory volume (r=0.348) were observed in a study conducted by Ammar *et al.* (2021). However, no correlation of testosterone level with sperm motility (r=0.145), sperm concentration (r=0.264), semen pH (r=-0.208) and total sperm abnormalities (r=-0.242) was found.

2.10 Scrotal circumference and female fertility traits

Scrotal circumference is highly correlated with age at puberty in half-sibling heifers (Kassetas *et al*., 2021). Heritability estimates for female reproductive traits are generally low, while heritability estimates of testicular traits are moderate to high (Boerne *et al*., 2019). It has been estimated that for every 1-centimetre increase of a sire's scrotal circumference over the population average, one can expect a 4-day decrease in the age at onset of puberty in heifer offspring (Klein *et al*., 2022). The variation of one centimetre increase in testicular circumference on reduction in age at puberty has been from 0.75 to 10 days. Therefore, it is well accepted that sire with above average scrotal circumference should produce female offspring that reach puberty sooner and have greater lifetime reproductive potential.

2.11 Heritability components of the bull fertility

Heritability measures the number of observed differences in a trait resulting from genes passed from one generation to another (Berry *et a*l., 2014). According to Ogunniyan and Olakojo (2014), the values of heritability ranges from 0 (low) to 1 (high). Low heritable traits are usually under less genetic control and are affected by the environment (non-genetic factors) to a greater extent than traits that are highly heritable (Mangino *et al*., 2017). Testicular size as measured by SC would appear to be moderately heritable in most breeds of beef bulls. Therefore, increased precocity based on the selection of bulls for larger scrotal circumference may enhance the precocity of their progeny (Gressler *et al.*, 2000).

Components of fertility such as SC, sperm motility and sperm morphology vary in terms of heritability. Kastelic (2014) found a heritability estimate of 41% for age at puberty in daughters and SC in half-brothers of a bull with a larger testicular development. However, Van Melis *et al.* (2010) Found 53% and 42% for these characteristics. According to a study conducted by Coulter (1982), Semen production traits were found to have moderate heritability (from 0.15 to 0.30) while some of the semen quality traits such as motility had high heritability of close to 0.60. Gressler *et al.* (2000) found heritability traits from sperm motility, sperm number and sperm concentration to be generally low, they ranged from 0.03 to 0.24. The generally high heritability for scrotal circumference and low heritability for semen quality traits and the consistently high desirable genetic correlations between scrotal circumference and those semen quality traits suggest that direct selection for increased scrotal circumference size would be most effective in bringing about improvement in semen quality traits (Viu *et al*., 2015).

2.12 Cattle adaptation to the tropical environment

The evolution of cattle results from the domestication of animals, environmental pressure, and human migration subsequently, resulting in over 1000 cattle breed world-wide, with more than 30 beef breeds registered in South Africa (Zwane *et al*., 2019). Over the past years, research has been conducted to assess adaptability, efficiency, meat quality and production of various cattle. Dutta *et al*. (2020) indicated that, according to fossil evidence and mitochondrial DNA, the Indian zebu (*B. indicus*) which represent today's modern Indian zebu breed, diverged from the Eurasian auroch (*B. primigenius* primigenius) 300,000 years ago. The Eurasian auroch eventually diverged into the African zebu (*B. primigenius indicus*), European taurus (*B. taurus taurus*), and African taurus (*B. taurus africanus*). As mankind began to populate the planet, the cattle that accompanied them on their journey had to adapt to the environmental conditions they were exposed to (Georges *et al*., 2019). Those which survived the associated environmental conditions were subsequently selected for domestication. Environmental pressures (e.g. local predators, climate, biome) dictated which individual cattle were successful with regards to survival rates (ability to find resources and avoid predators), thriftiness (efficiency in current biome forage availability), adaptability (capacity to change with the environment), and productivity. Consequently, cattle diverged into subspecies that differed in physiology, nutritional requirements, social behaviour, and body composition (Cooke *et al*., 2020).

According to Sartori *et al*. (2016) *Bos indicus* cattle exhibit increased resistance to many environmental stressors relative to *Bos taurus*, but tend to have slower growth, and meat quality as they have not been as intensively selected for these traits as specialised *Bos taurus* breeds. The Bos indicus breeds originate from the tropical parts of the world such as Asia and Sub-saharan Africa which is characterised by hot climate relative to the B*os taurus* breeds originating from the temperate parts of the world such as Europe and north Asia characterised by a cooler climate (Mwai *et al*., 2020). Breeding strategies which can be implemented are crossbreeding and pure breeding in the subtropics and tropics, to manipulate the adaptation of the animal (Mwai *et al*., 2015). The Sanga breeds are some of the hybrid breeds that originated from both *Bos- taurus* and *Bos-indicus* e.g., Bonsmara which was created after many cross matings and back-crosses consisting of five-eighths Afrikaner (Sanga-type), three-sixteenths Hereford, and three-sixteenths Shorthorn (Jang *et al*., 2021).

2.13 Measuring of the scrotal circumference

Measuring scrotal circumference can indicate whether testicular development is within the normal range (Menegassi *et al*., 2019). The scrotal circumference of a bull provides an important indication of his genetic merit for several important fertility traits (Russell *et al*., 2021). Tape metre or Coultier scrotal circumference measuring tape can be used to measure a bull’s SC. Bulls are strong and big, they can be unpredictable at times hence it is important that there should be a proper handling facility. Put the bull safely in a compression chute and fasten a bar behind him so that he can't back up within the chute. Then, either from behind the bull, which is usually preferable, or through opening the bottom side panels of the chute, one may access the scrotum. It will be safer to have someone "tailing" the bull. This may be accomplished by having a helper who will grab the bull's tail between 8 and 10 inches from the tail head, and then pushing the tail straight up. The risk of the bull kicking will be much reduced by forcing the tail upward rather than to the side, which will have minimal to no impact. The testicles of the bull may be brought up close towards his body wall, depending on the ambient environmental temperature and disposition of the bull. The testicles must first be softly placed into the bottom of the scrotum in order to measure the scrotal circumference properly (in cm or mm). Put your fingers and thumbs on either side of the scrotum above the testicles to hold them in this position. Avoid putting fingers between the testicles since doing so will unintentionally increase the scrotal circumference measurement and make it.

2.14 Summary of the review

The production of cattle in rural communities of South Africa remains the most important livestock sub-sector, with the communal and emerging farmers dominating the beef industry. Methods related with testicular parameters and scrotal circumference used to predict the potential of sperm production and particularly to identify the bulls with high sperm output potential at an early age are important. Scrotal circumference is easily measured, highly heritable and favourably related to semen quality. Hence it is one of the most considered methods of predicting the fertility status of the bull. This moderate to high repeatability physical traits such as scrotal circumference demonstrate that bulls can be selected based on an annual pre- breading examination. The bull is considered to be half of its herd, which implies that a bull can service 25 to 30 cows. Therefore, the selection of fertile bulls can be used as the most powerful method to improve a herd and the production of a farming enterprise since the bull has a high genetic influence on its progeny. Practices such as physical examination of a bull can provide the foundation of a breeding soundness examination.

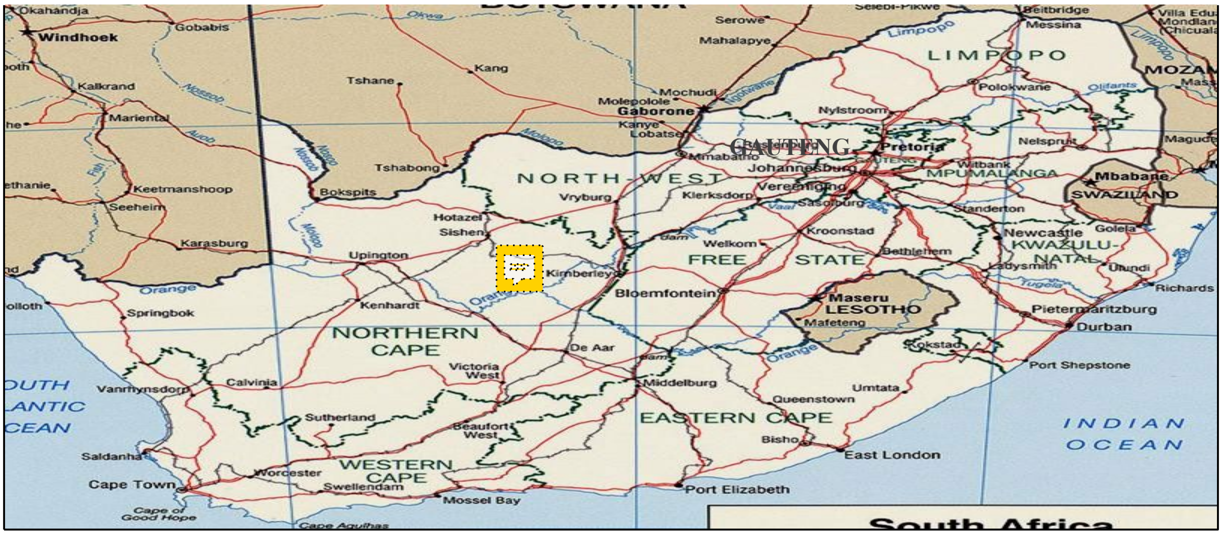
CHAPTER 3

MATERIALS AND METHODS

3.1 Study site

The study used previously recorded Phase C (on-station) data at a Bull Testing Centre either privately owned or owned by the Agricultural Research Council (ARC) and enrolled with the Integrated Registration and Genetic Information System (INTERGIS) program. Phase C Bull Testing Centres are located in Gauteng, Northwest, Free State, and Western Cape (Hendricks, 2021).

Figure 3.1. Bull Testing Centres located in Gauteng, Northwest, Free State and Western Cape.



The temperature at the Agricultural Research Council in Pretoria ranges from 6 to 21℃ in winter and 16 to 27.31℃ in summer, while rainfall ranges from 0 to 1.4 mm in winter and 84.4 to 106.6 mm in summer (Ramukhithi *et al*., 2019).

3.2 Experimental design

Cross sectional experimental design was used; measurements were taken ones. Cross sectional experimental design is a research design in which you collect data from many different animals at a single point in time (Musemwa *et al.,* 2010). Various provinces were used as the source of variation. All the animals accepted into phase C went through an adaptation period of 28 days and a test length of 84 days. Breeds with a minimum of 5000 data entry were used for data collection. The breeds that were used include Drakensberger, Hereford, Nguni, Limousing, Santa Gertrudis, Simmentaler, Angus SA, Afrikaner, Beefmaster, Bonsmara, Brahman, Charolais and Sussex

3.3 Data collection

Secondary Phase C data from the INTERGIS program was used for the study. All Testing Centres comply with the requirements for participating in the programme as spelt out by the ARC such as the availability of a functional weighing scale, accurate, complete, and up-to-date record keeping. Bulls were admitted into Phase C between 151 and 250 days of age at the beginning of adaptation. The maximum age possible of the bulls at the end of Phase C was 362 days. The bulls were adapted for between 21 and 90 days, and the test length lasted for 270 days. The data used in this study was on breed, age, SC, and live weight. The age was measured as the number of days from birth to the date of SC measurement (days) and also through examining the teeth, live weight was measured by subjecting the animal to cattle weighing scale (kg) (cattlescale.com) on the date of SC measurement and the SC was measured using a testes circumference meter(cm) developed by the ARC in South Africa.

The testes circumference meter was scaled in millimetres and centimetres and its circular end was attached to a rod in its manufactured state (Figure1A). The rod can be folded inwards or outwards while adjusting the tape during measurement (Figure 1B). During measurement, animals were moved into a crush pen, restrained and the measuring tape was thrown around the scrotum. The tape can only measure SC from 200 mm and cannot measure anything below that. However, according to Hendricks (2018), bulls with SC smaller or equal to 200 mm should be recorded as 200 mm. Bulls were fed a standard, complete grower diet comprising at least 20 percent roughage and individually *fed ad lib* during the test period. On arrival bull calves were treated against internal and external parasites and, if necessary, skin diseases. Bull calves were spray-dipped regularly and fitted with neck belts to which their transponder and pen number was attached. A veterinarian examined the bulls regularly and was available in all cases of diseases and emergencies.

A

B

Figure 3.2: Measurement of the scrotal circumference. (A) A testis circumference meter applied around the scrotum (B) An ARC technician is adjusting the testis circumference meter during SC measurement.

3.4 Data analysis

In this study, data was analysed using the Statistical Package for the Social Sciences (SPSS IBM 2019) version 26.0 software. Histograms was constructed for age and SC to examine data for normal distribution. The change in SC per day (SC/d) was calculated for each individual bull by dividing the difference in SC1 and SC2 by the number of days on feed (Love *et al.,*1991). On the assumption that change was linear, SC/d was used to correct actual SC measurements to 240 and 365 d of age. Correlations between SC/d and age at entry was calculated for each breed by Pearson's correlation coefficient (r), to assess the degree of linearity of change in SC over the age range of bulls. The statistically significant level is determined using 5% for significant and 1% for highly significant. GLM model used:

$$Y_{ij} = \mu + T_i + B_j + e_{ij}$$

Where:

Y_{ij} = the observation of the scrotal circumference,

μ = the overall mean,

T_i= the treatment effect (i= age)

B_j= the treatment effect(j=breed) and

e_{ij}= the residual error

CHAPTER 4

RESULTS

4.1 The minimum acceptable development of scrotal circumference

The minimum acceptable development of scrotal circumference in Brahman, Nguni, Limousin, Santa Gertrudis, Afrikaner, Drakensberger, Charolais, Hereford, Sussex, Beefmaster, Bonsmara, South African Angus, and Simmental is presented in Table 4.1. All the breeds were above the minimum acceptable scrotal circumference of 20 cm.

Table 4.1: The minimum acceptable development of scrotal circumference

Breed	Number of bulls	Mean	Standard deviation
Nguni	137	30.201	3.053
Bonsmara	19515	34.351	2.515
Afrikaner	2361	31.637	2.849
Simmental	2263	35.927	3.157
Angus SA	2757	34.509	2.750
Hereford	1301	33.315	2.456
Charolais	907	32.269	3.178
Sussex	1560	32.623	2.708
Brahman	965	27.825	3.147
Drakensberger	3012	32.412	3.066
Beefmaster	2074	32.983	3.389
Limousin	1194	30.531	2.715
Standard minimum acceptable scrotal circumference: 20 cm (Menegassi *et al*., 2011)			

4.2 Correlation between scrotal circumference, age, and live weight of beef bulls during Phase C

Table 4.2 shows the correlation between scrotal circumference (SC), age, and body weight (BW) of beef bulls during Phase C. In Nguni, the results showed that SC had a highly positive significant correlation ($P<0.01$) with age and BW. However, age had a non-significant correlation with BW (P>0.05). The results for Brahman, Limousin, Afrikaner, Drakensberger, Charolais, Sussex, Beefmaster, Bonsmara, and Simmental showed a highly significant positive correlation (P<0.01) between SC, BW and age. In Hereford age had a significant positive correlation (P<0.05) with BW. Furthermore, a highly significant positive correlation (P<0.01) between SC and age as well as SC and BW was also observed. The results for Angus SA showed that SC had a highly positive significant correlation (P<0.01) with age and BW. However, a highly negative significant correlation (P<0.01) was observed between age and BW.

Table 4.2: Correlation between scrotal circumference, age, and live body weight of beef bulls during Phase C

Breeds	Variables	Age	Sc	BW
Nguni	Age	1.00	0.971**	0.096^{ns}
	Sc		1.00	0.263**
	BW			1.00
Bonsmara	Age	1.00	0.105**	0.119**
	Sc		1.00	0.288**
	BW			1.00
Afrikaner	Age	1.00	0.973**	0.182**
	Sc		1.00	0.187**
	BW			1.00
Simmental	Age	1.00	0.237**	0.223**
	SC		1.00	0.576**
	BW			1.00
Angus SA	Age	1.00	0.130**	-0.061**
	Sc		1.00	0.348 **
	BW			1.00

Hereford	Age Sc BW	1.00	0.124 ** 1.00	0.058* 0.855** 1.00
Charolais	Age Sc BW	1.00	0.259** 1.00	0.182** 0.422** 1.00
Sussex	Age SC BW	1.00	0.183** 1.00	0.143** 0.334** 1.00
Brahman	Age Sc BW	1.00	0.106 ** 1.00	0.162** 0.343** 1.00
Drakensberger	Age Sc BW	1.00	0.104** 1.00	0.102** 0.507** 1.00
Beef master	Age Sc BW	1.00	0.198** 1.00	0.263** 0.525** 1.00
Limousin	Age Sc BW	1.00	0.247** 1.00	0.235** 0.497** 1.00

SC: Scrotal circumference; BW: Body weight; ns: non-significant; **: significant at P<0.01

4.3 The effect of breed on scrotal circumference

Table 4.3 below shows the effect of breed on the scrotal circumference. The results showed a significant difference between breeds ($P<0.05$) on SC except between Bonsmara and Angus SA ($P>0.05$), Charolais and Drakensberger ($P>0.05$) as well as Sussex and Drakensberger ($P>0.05$).

Table 4.3: The effect of breed on scrotal circumference

Breeds	Number of bulls	Mean ± Standard deviation
Nguni	137	30.201± 3.053^{i}
Bonsmara	19515	34.351± 2.515^{b}
Afrikaner	2361	31.637± 2.849^{g}
Simmental	2263	35.927± 3.157^{a}
Angus SA	2757	34.509± 2.750^{b}
Hereford	1301	33.315± 2.456^{c}
Charolais	907	32.269± 3.178^{f}
Sussex	1560	32.623± 2.708^{e}
Brahman	965	27.825± 3.147^{j}
Drakensberger	3012	32.412± 3.066^{ef}
Beefmaster	2074	32.983± 3.389^{d}
Limousin	1194	30.531± 2.715^{h}

4.4 The effect of age on scrotal circumference

Table 4.4 below shows the effect of age on scrotal circumference. A highly significant difference on Sc between different age groups (P<0.01) was observed on breeds except between Nguni, Sussex, and Brahman (P>0.05).

Table 4.4: The effect of age on scrotal circumference

	5-10 Months	**10-17 Months**
Breed	**Mean± Standard deviation**	**Mean± Standard deviation**
Nguni	27.000± 3.546[a]	30.399± 2.922[a]
Bonsmara	33.442± 2.418[b]	34.385± 2.512[a]
Afrikaner	30.170± 3.102[b]	31.755± 2.795[a]
Simmental	33.095± 3.282[b]	36.023± 3.108[a]
Angus SA	33.760± 2.606[b]	34.579± 2.753[a]
Hereford	32.412± 3.115[b]	33.391± 2.379[a]
Charolais	30.611± 3.415[b]	32.393± 3.126[a]
Sussex	32.653± 3.241[a]	32.623± 2.690[a]
Brahman	27.816± 3.513[a]	27.825± 3.133[a]
Drakensberger	31.585± 3.490[b]	32.488± 3.014[a]
Beefmaster	31.115± 2.808[b]	33.056± 3.389[a]
Limousin	29.124± 2.781[b]	30.703± 2.657[a]

CHAPTER 5

DISCUSSION, CONCLUSION AND RECOMMENDATION

5.1 Discussion

The results of the current study indicated that tropical adapted South African beef bulls during phase c have a normal testicular development. According to Menegassi *et al*. (2011) the minimum acceptable SC is 20 cm, and all the studied breeds were above the minimum acceptable scrotal circumference of 20 cm. In a situation where there is lack of equipment/ Computer assisted sperm analysis (CASA), especially in the rural communities, method such as measuring the scrotal circumference can be used to determine fertility in bulls (Permal, 2014). The first part of the study focused on the minimum acceptable development of scrotal circumference in 13 tropically adapted South African beef bulls. The study indicated that a breed with the highest SC is Simmental (35.927 cm), and breed with lowest SCis Brahman (27.825 cm). The results of Fordyce *et al.* (2014) are consistent with those of the current study where Simmental bulls had the highest scrotal circumference compared to all the studied breeds. These results are also in line with those of Coulter *et al.* (1979) where other breeds similar to those found in this study such as Angus bulls (35.282 cm), Hereford (33.992 cm), and Limousin (31.310 cm) were also explored. Furthermore, SC of Simmental bulls ranged from 33-37 cm and that in Charolais and Angus ranged from 32-36 cm, Hereford ranged from 31-35 cm and, Limousin ranged from 30-34 cm. In a study conducted by Menegassi *et al. (2011)* the results of Charolais (36-37cm), Limousine (33-37cm), and Hereford (36-37cm) where not in line with those found in the present study. This can be due to different environmental conditions, diets and age.

The second part of the study focused on the relationship between scrotal circumference, age, and body weight in South African beef bulls during Phase C. The results indicated that there was a significant correlation amongst the breeds except in Nguni, were age had a non-significant correlation with BW ($P>0.05$). These results suggest that there was no association between age and body weight in Nguni. The results can also be due to the Nguni attributes such as resilience on fragile and marginal land; and in drought and stress conditions for longer periods. According to Utsunomiya *et al.* (2022) *Bos indicus* breeds are better suited to tropical climates and typically more resilient to heat stress compared to *Bos-Taurus* (Gujar *et al*., 2022).

The results for Brahman, Limousin, Afrikaner, Drakensberger, Charolais, Sussex, Beefmaster, Bonsmara, and Simmental showed a highly significant positive correlation (P<0.01) between SC, BW and age. The results for Brahman are in line with those of Holroyd *et al.* (2000) whereby brahman showed a significant correlation (P<0.01) between SC, BW and age. Brito *et al.* (2012) reported different results from the current study where a highly negative correlation (P<0.01) between BW and age was observed in Charolais bulls. These results can be due to different diets and environments. In Hereford age had a significant positive correlation (P<0.05) with Sc and BW. The results for Menegassi *et al.* (2011) also indicated that in Hereford age had a significant positive correlation (P<0.05) with Sc. In Angus SA the results showed that SC had a highly positive significant correlation (P<0.01) with age and BW. However, a highly negative significant correlation (P<0.01) was observed between ageand BW. These results are consistent with those of Brito *et al.* (2012) whereby a negative significant correlation (P<0.05) in Angus SA between age and BW was observed.

The last objective showed the effect of breed and age on scrotal circumference (cm). The results of the current study showed a significant difference between breeds (P<0.05) on SC except between Bonsmara and Hereford (P>0.05), Charolais and Drakensberger (P>0.05) as well as Sussex and Drakensberger (P>0.05) where a potential of a type 2 error increases, which indicates a low statistical power thus increasing the chances of missing a difference between the breeds. According to a study conducted by Judge *et al.* (2017) early maturing genotypes present higher relative growth of some body structures and organs at the beginning of puberty. This may explain the higher SC values obtained in Hereford and Angus, which are early- maturing breeds, compared to other breeds. A study conducted by Menegassi *et al.* (2019) showed that Angus had SC of 33.5cm and Hereford had SC of 34.1cm. The results of the current study showed a SC of 34.509 cm in Angus and 33.315 cm in Hereford, which contrast with those found by Menegassi *et al.* (2019). Groenewald (2017) reported that Beefmaster (33.51 cm) bulls had larger (P < 0.05) SC than the Bonsmara bulls (32.79 cm) bulls.

These results contradicted with those of the current study were Bonsmara bulls (34.351 cm) had a SC higher than that of Beefmaster (32.983 cm). This can be due to the huge difference in sample size and nutrition. It was further reported that both Bonsmara and Beefmaster had a SC higher than that of Nguni bulls (30.30 cm ± 0.170). These results are consistent with those of the current study.

A highly significant difference on SC between different age groups (P<0.01) was also observed on breeds except between Nguni, Sussex, and Brahman (P>0.05). Menegassi *et al.* (2019) determined that measure of SC is performed during puberty, between 510 and 540 days old, allowing to identify bulls presenting fast testicular growth rate which is considered a characteristic of early sexual maturity. Neves *et al.* 2011 obtained values of 18.2 cm at 5-7 months, 18.8 cm at 7-12 months, 22.9 cm at 12-18 months, and 31.9 cm at 18-24 months of age in all the breeds. These results are consistent with those of the current study, where an increase in age showed an elevation in SC. Weerakoon *et al.* (2018) reported different results, where Sc was negatively correlated with Age in Japanese Black beef bulls. This difference can be due to different environmental conditions and genetic make-up.

5.2 Conclusion

Normal testicular development in tropical adapted South African beef bulls during Phase C (centralised testing centre) was assessed using measurements of scrotal circumference. Scrotal circumference has been found to be affected by age and body weight. The minimum acceptable development of scrotum circumference, relationship between scrotal circumference, age, as well as live weight and, the effect of breed and age on scrotal circumference in tropical adapted South African beef bulls were determined using procedure of means, procedure of correlation and procedure for General Linear Model, respectively. The results of this study showed that all the breeds where above the minimum scrotum circumference, therefore all the studied breeds can be suitable for breeding purposes especially in stud farms. Furthermore, all the breeds showed a positive correlation between SC, BW and age. Nguni, AngusSA, Brahman, Limousin, Afrikaner, Drakensberger, Charolais, Sussex, Beefmaster, Bonsmara, and Simmental showed a highly significant positive correlation between Sc, BW and age. In Hereford the relationship between SC, BW and age was not as strong as that of the other breeds. This information can be mostly used by stud farmers to prioritize breeds that show a highly significant positive correlation between SC, BW and age, so that the genetic potential of their herd can be improved. In this case breeds such as Hereford won't be prioritized for breeding purposes due to its compromised correlation between SC, BW and age compared to the other breeds. A highly negative significant correlation ($P<0.01$) was observed between age and BW. These can be due to many factors such as diet, which can be easily manipulated through balancing feed ration. Breeds with a highly significant difference on SC between different age groups observed in the current study indicate a favourable growth curve.

The findings of this study can therefore be used by farmers especially Stud farmers to make the right management decisions like selecting superior bulls that will improve their herd and production, formulating the correct/relevant feed rations for their bulls and setting price when selling. These findings may aid extension officers and researchers in better understanding the normal testicular development of tropical adapted South African beef bulls during phase C. This study can also help in early detection of potential issues, enabling timely intervention or exclusion of bulls with suboptimal reproductive proficiencies from breeding programs.

5.3 Recommendations

(i) Farmers should pay attention to these production parameters when selecting bulls in order to improve the genetic potential of their herd.

(ii) More research on determination of normal testicular development in tropical adapted South African beef bulls and other breeds as well as other species should be conducted.

(iii) Workshops where farmers will be taught about the importance of normal testicular development should be conducted particularly by extension agents and researchers.

(iv) Farmers should also be taught how to measure the Sc so that they can independently determine the normal testicular development.

CHAPTER 6

REFERENCES

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