Prevalence of Ticks and Tick-borne Diseases in Cattle on Communal Rangelands in the Highland Areas of the Eastern Cape Province, South Africa

By

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Declaration

Apart from the assistance received that has been reported in the Acknowledgements, References and in the appropriate places in the text, this Dissertation represents the original work of the author. No part of this dissertation has been presented for any other degree at any other University.

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Abstract

Surveys were conducted to compare the seasonal tick prevalence and loads, and sero-prevalence of tick-borne diseases (TBD) in Nguni and non-descript cattle on the sweet and sour communal rangelands of the Eastern Cape Province. The tick species observed on both rangeland types were *Rhipicephalus appendiculatus* (71.0 %), *Rhipicephalus* (*Boophilus*) species (29.2 %) and *Rhipicephalus evertsi evertsi* (40.2 %). *Hyalomma* species (19.0 %) occurred only on the sour rangeland. Tick loads were higher ($P < 0.05$) in the hot-wet season than in the cool-dry season. Cattle in the sweet rangeland had significantly lower ($P < 0.05$) tick loads than those in the sour rangeland. *Rhipicephalus appendiculatus* loads were lower ($P < 0.05$) in the indigenous Nguni than non-descript cattle in the hot-wet and post-rainy season. *Hyalomma* species were also significantly lower ($P < 0.05$) in the Nguni than non-descript cattle in all the seasons. Three TBDs were observed, namely *Babesia bovis* (44.6 %), *Babesia bigemina* (45.9 %) and *Anaplasma marginale* (25.6 %). All the animals were sero-negative for *Ehrlichia ruminantium*. Nguni cattle had lower ($P < 0.05$) sero-prevalence for *A. marginale* in the cool-dry season and *B. bigemina* in the cool-dry and hot-wet seasons. Cattle in the sweet rangeland had significantly lower sero-prevalence of *B. bovis* and *B. bigemina*. Infection with *B. bovis* and *A. marginale* decreased ($P < 0.05$) the packed cell volume. Nguni cattle were recommended for use in the integrated control of ticks and TBD in the communal areas of South Africa as they were better able to cope with tick and TBD infestations than non-descript breeds.

Key words: Nguni, *Rhipicephalus appendiculatus*, *Babesia bovis*, tick loads, season

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Dedication

To wisdom, understanding and upright judgement!

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Chapter 1: Introduction

1.1 Background

Of the 14.1 million cattle in South Africa, 3.1 million are in the Eastern Cape (National Livestock Statistics, 2006) and approximately half of these belong to communal farmers (Palmer and Ainslie, 2006). Cattle production contributes considerably to the livelihoods of communal farmers in the Eastern Cape Province of South Africa. Cattle are a source of food for household consumption (Sansoucy, 1995) and they provide draught power for crop production, hides, manure and cash through sales (Chimonyo *et al.*, 1999; Palmer and Ainslie, 2006).

Cattle owned by resource-poor farmers are kept on communal rangelands where they are grazed extensively (Masika and Mafu, 2004). Communal grazing is characterised by poor management of cattle and low productivity. Communal farmers rarely use drugs to treat their animals. Consequently, diseases and parasitism are rife and major threats to cattle production in communal areas (Kaewthamasorn and Wongsamee, 2006; Rajput *et al.*, 2006). Surveys have indicated that communal farmers perceive ticks as the most important health constraint to their cattle (Dreyer *et al.*, 1998; Dold and Cocks, 2001).

Ticks cause substantial losses in cattle production, in terms of diseases, reduced productivity and fertility and often death, and are economically the most important ecto-parasites of cattle (Rajput *et al.*, 2006). Ticks suck blood, damage hides and skins, introduce toxins and predispose cattle to myiasis and dermatophilosis (Gates and Wescott, 2000; Mtshali *et al.*, 2004). Furthermore, they reduce body weight gains and milk yield, in addition to creating sites for secondary invasion by

pathogenic organisms (Gates and Wescott, 2000; Turton, 2001; Kaufman *et al.*, 2006). More significantly, ticks transmit diseases from infected cattle to healthy ones. Ticks transmit a greater variety of pathogenic micro-organisms than any other arthropod vector group, and are among the most important vectors of diseases affecting animals (Jongejan, 2007).

The most economically important genera of tick-borne prokaryotic and eukaryotic haemoparasites infecting cattle in communal areas are the rickettsiae *Anaplasma* and *Ehrlichia* (*Cowdria*), and the protozoan parasites *Babesia* and *Theileria* (Bell-Sakyi *et al.*, 2004). Anaplasmosis, heartwater and babesiosis are the most important constraints to the health and improved productivity of cattle in South Africa (Coetzer *et al.*, 1994; Mtshali *et al.*, 2004). They cause high morbidity and mortality, decreased meat and milk production and loss of draught power, manure and financial resources through the institution of control measures (Makala *et al.*, 2003). Serological tests, such as the indirect fluorescent antibody test (IFAT) and enzyme-linked immuno-sorbent assay (ELISA) can be used to detect antibodies and give accurate estimates of TBD sero-prevalence in cattle (Minjauw and McLeod, 2004).

Most indigenous cattle in areas where tick-borne diseases (TBDs) occur possess a natural resistance to these diseases (d'Ieteren and Kimani, 2007). These cattle are exposed to the diseases early in life and thus do not usually develop the clinical disease and are subsequently immune (Latif, 1992). In South Africa, the Nguni has been reported as a hardy breed uniquely adapted to the local environment and possessing a high tolerance to ticks and TBDs (Spickett *et al.*, 1989; Mapiye *et al.*, 2007; Muchenje *et al.*, 2008). The indiscriminate crossing of Nguni cattle with exotic breeds to improve productivity (Dold and Cock, 2001) is likely to lead to loss of some of their desirable traits, such as resistance or tolerance to TBDs. The prevalence of ticks

and TBDs in the Nguni and its crosses (which can be described as non-descript) in the extensive system in which they are currently kept in South Africa has not been determined.

Muchenje *et al.* (2008), through conducting tick counts, observed that Nguni steers were less susceptible to ticks than Bonsmara and Angus steers raised on natural sweet rangeland. It is essential, however, to determine the impact of ticks and TBDs under communal grazing management and in the breeds of cattle that are kept in the communal areas. Estimates of the prevalence of ticks and TBDs in cattle in small scale farming areas in the Free State have been made (Dryer *et al.*, 1998; Mbati *et al.*, 2002). These studies however, did not compare the prevalence of the parasites in the different breeds kept under communal farmer management and across different rangelands types. Developing control strategies for ticks and TBDs based on extrapolation of studies conducted elsewhere are often inappropriate due to differences in ecological factors and management practices that exist between different areas. Climatic conditions of the sweet and sour rangelands differ (Ellery *et al.*, 1995), yet the interaction between veld type and season on tick counts and TBDs are often ignored.

1.2 Justification

The low-lying coastal areas of the Eastern Cape predominantly carry the commercial cattle producing farms. There is therefore a need to sample cattle from the highland sweet and sour rangelands which are more inland as they carry the bulk of communal cattle in the province. The prevalence and seasonal occurrence of ticks and TBDs in the different breeds of cattle under the communal farming system in the Eastern Cape has not been determined. Information on the prevalence and seasonal occurrence of ticks and TBDs in cattle facilitates the development of

sustainable control strategies to enable communal farmers to reduce the burden of these parasites on their stock. For the farmers to fully benefit from the research there is need for their active participation during data collection. During this time their ethical considerations and the welfare of their animals should not be ignored. The current efforts to restock communal areas with Nguni cattle require determination of the breed's performance and resistance to ticks and TBDs in communal areas. This information can also be useful in advising communal farmers on the selection and rearing of appropriate breeds that are tolerant to ticks and TBDs. Data on the prevalence and distribution of these parasites can be used for future research on development of drugs and other remedies to protect animals from ticks and TBDs in the province.

1.3 Objectives

The main objective of the current study was to compare the tick loads, prevalence of ticks and sero-prevalence of TBDs in cattle of different breeds in communal areas of the Eastern Cape.

The specific objectives were to:

i. Determine the prevalence of ticks in Nguni and non-descript breeds of cattle in communal areas of the sweet and sour rangelands of the Eastern Cape; and

ii. Determine the sero-prevalence of babesiosis, anaplasmosis and ehrlichiosis, and the associated changes in packed cell volume in Nguni and non-descript breeds of cattle in communal areas of the sweet and sour rangelands of the Eastern Cape.

1.4 Hypotheses

The hypotheses tested were that:

i. There were breed differences in the prevalence of ticks in Nguni and non-descript cattle in the sweet and sour rangelands of the Eastern Cape.

ii. There were breed differences in the sero-prevalence of babesiosis, anaplasmosis and ehrlichiosis in Nguni and non-descript cattle in the sweet and sour rangelands of the Eastern Cape.

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Chapter 2: Literature review

2.1 Introduction

Tick-borne diseases and the damage caused by tick bites are, arguably, the major constraints to cattle production in communal areas of South Africa (Dold and Cocks, 2001). The current chapter discusses cattle production systems, breeds, and highlights the major tick species and TBDs of cattle in South Africa. It also reviews tick control and current serological methods used for the diagnosis of TBDs.

2.2 Cattle production in communal areas

Cattle production in South Africa can broadly be divided into two: large-scale commercial farming and small-holder farming in the communal areas (Gilimani, 2005). Large-scale commercial farming is profit-driven, while resource-poor farmers form the bulk of small-holder farmers and rear their cattle on communal rangelands (Bryson *et al.*, 2002). The majority of small-holder farmers reside in communal areas and are categorised as 'subsistence farmers' since they produce mainly for household consumption (Van Averbeke and Mohamed, 2006). Cattle provide draught power, skins, dung, meat and milk, are used for home consumption (Chimonyo *et al.*, 1999) with a few instances of sales. Cattle ownership is fundamental to social status and self-esteem for the communal farmer (Minjauw and McLeod, 2003). Animals graze on natural pasture on common grazing grounds (Masika and Mafu, 2004). At night, the animals are protected from theft and/or predation by confinement in night enclosures which are constructed using thorn bushes, stones or wooden poles, depending on the availability of these materials (Campbell *et al.*, 2006).

There is limited livestock and rangeland management, resulting in rangeland degradation and poor cattle body condition, especially during winter (Bester *et al.*, 2003). Cattle in the sweet rangelands are, however, spared the ravages of winter as forages retain palatability and high nutritive value to maintain animal condition throughout the dry season (Sibanda, 1999). Communal farmers rarely practise controlled breeding, supplementary feeding and marketing of animals. The only human input into the communal farming system is the unpaid family labour used to look after the animals. The main management objective is to avoid risks of diseases, drought and to maintain herd numbers (Campbell *et al.*, 2006).

Livestock diseases, parasitism and deaths are the major threats to communal cattle production in the small-holder production system (Dold and Cocks, 2001; Rajput *et al.*, 2006; Hesterburg *et al.*, 2007). Communal farmers have limited access to veterinary care in terms of support services, information about the prevention and treatment of livestock diseases, and preventive and therapeutic veterinary medicines (Dold and Cocks, 2001). The cost of veterinary drugs is beyond the reach of the majority of communal farmers (Mbati *et al.*, 2002). Ticks and TBDs are ranked by communal farmers as the most important health constraint in their cattle (Dreyer *et al.*, 1998; Hesterburg *et al.*, 2007). Due to lack of funds and shortage of manpower, veterinary services are limited in most communal areas of South Africa (Dold and Cocks, 2001). Communal farmers presently rely on the use of traditional medicines to combat the constraint of ticks and TBDs in their stock (Hesterburg, *et al.*, 2007).

2.3 Cattle breeds found in communal areas

2.3.1 Indigenous cattle

The cattle breeds kept by communal farmers in the small-holder farming areas of South Africa are the indigenous, imported and non-descript (crossbred) breeds (Scholtz *et al.*, 2008).

The Nguni is the major indigenous cattle breed of South Africa. It is a small to medium sized breed that is reported to be adapted to the harsh environments (Collin-Luswet, 2000) of South Africa's communal areas where droughts are periodic, dry season nutrition is low and cattle diseases are endemic (Otto *et al.*, 2000). Nguni cattle possess various adaptation characteristics that make them suitable for rearing in communal areas. They have excellent reproductive performance (du Plessis *et al.*, 2006) good walking and foraging ability, low maintenance requirements and good meat quality (Schoeman, 1989; Strydom *et al.*, 2001; Muchenje *et al.*, 2008a). The Nguni has a great ability to maintain its condition in winter (Ndlovu, 2008; Ndlovu *et al.*, 2007). As selective grazers and browsers, Nguni cattle are able to obtain optimal nutritional value from the available natural vegetation, thus enabling them to survive under poor rangeland conditions (Bester *et al.*, 2003). The Nguni breed can walk long distances in search of grazing and water; it is also tolerant of extreme temperatures (Bester *et al.*, 2003).

Nguni cattle have been reported to be resistant to internal parasites (Ndlovu, 2007) and ticks (Muchenje *et al.*, 2008b), making them suitable for rearing in the disease-endemic communal rangelands of South Africa. The mechanism for tick resistance is not fully understood but could be related to a pre-immunity to ticks often established through a continuous contact with the infectious agents from early in life (Mattioli *et al.*, 2000). Avoidance behaviour, skin

hypersensitivity and increased grooming may also contribute to increased resistance of the Nguni breed to ticks (Meltzer, 1996). It is thought that by exploiting their innate and acquired resistance against ticks and TBDs (Minjauw and McLeod, 2003), indigenous Nguni cattle can be reared with minimal tick control. Tick control by frequent application of acaricides is regarded to be costly in indigenous cattle (Mattioli *et al.*, 1998). Moreover, frequent and prolonged use of acaricide compounds, might depress body weight gain in cattle (Fivaz and de Waal, 1993). Studies have been conducted to determine the tolerance of Nguni cattle to ticks although under controlled conditions (Scholtz *et al.*, 1991; Muchenje *et al.*, 2008b). It is imperative, however, to conduct tick counts in the Nguni on different rangeland types so as to determine its resistance to ticks under communal grazing management.

2.3.2 Imported and non-descript cattle

Imported breeds, such as the Brahman, Angus, Hereford, Simmental, Holstein, Jersey and Brown Swiss, were introduced into South Africa after early colonial farmers and scientists rejected indigenous cattle as unproductive and advocated their replacement with large-framed and fast growing cattle of European origin (Bayer *et al.*, 2004). Communal farmers accepted these high-input, highly productive imported breeds as more superior and adopted them into the communal farming system (Bester *et al.*, 2003). Exotic breeds however, are not adapted to the conditions, and perform poorly under the prevailing management practices of communal systems (Scholtz, 1988). Uncontrolled breeding and indiscriminate crossing of indigenous Nguni cattle with exotic breeds to improve productivity has led to the production of numerous non-descript crosses (Scholtz *et al.*, 2008).

Imported breeds are less tolerant to ticks than indigenous breeds in southern Africa (Rechav and Kostrzewski, 1991; Norval *et al.,* 1996; Muchenje *et al.,* 2008b). Indigenous-exotic crosses are known to endure tick infestation for longer periods without acaricide application, and have lower tick burdens than imported breeds (Fivaz and de Waal, 1993). However, their level of resistance to ticks is thought to be comparably lower than that of pure indigenous breeds (Fivaz *et al.*, 1992; Wambura *et al.*, 1998). Tick loads in non-descript and indigenous Nguni cattle on communal rangelands have not been determined. Information gained from such comparisons can be useful in advising farmers on the rearing of locally-adapted tick-resistant breeds on their rangelands.

2.4 Common ticks in South Africa

Ticks are blood-sucking obligate external parasites belonging to the phylum *Arthropoda* and make up the largest collection of creatures in the order *Acarina* (Rajput *et al.*, 2006). They have direct detrimental effects on cattle (de Castro, 1997) but more importantly, they transmit various of pathogenic micro-organisms from infected cattle to healthy ones (Jongejan, 2007). The most common tick species and the diseases that they transmit to cattle in South Africa are shown in Table 2.1.

Table 2.1: Ticks and the pathogens they transmit in cattle in South Africa

Tick species	Description	Pathogens transmitted
Rhipicephalus (*Boophilus*) spp.	bluish ticks with hexagonal basis capitulum, short compressed and ridged palps, faint/absent anal groove	*Babesia bigemina*, *Babesia bovis*, *Anaplasma marginale*
Rhipicephalus appendiculatus	brownish, reddish-brown or dark ticks with short palps and reddish-brown legs	*Theileria parva*
Rhipicephalus evertsi evertsi	medium sized, beady-eyed, dark brown ticks with reddish-orange legs	*Anaplasma marginale*
Hyalomma spp.	dark-brown- bodied ticks with numerous puntactions on the scutum and long, banded legs	*Anaplasma marginale*
Amblyomma hebraeum	brightly ornamented ticks, that have eyes and long, robust mouthparts	*Ehrlichia ruminantium*

Sources: Horak *et al.* (1991), Walker (1991) and Coetzer *et al.* (1994).

2.4.1 Distribution of ticks in cattle in South Africa

Several surveys have been carried out to determine the tick loads of cattle in South Africa. Studies in the eastern Free State have revealed that the principal ticks affecting cattle belonging to resource poor farmers are *Boophilus decoloratus* (53.1%), *Rhipicephalus evertsi evertsi* (44.7%), *Rhipicephalus follis* (1.0%), *Rhipicephalus gertrudae* (0.7%) and *Rhipicephalus warburtoni* (0.4%) (Hlatswayo *et al.*, 2002; Mbati *et al.*, 2004). In the south west region of the same province, Fourie and Horak (1991) observed that *Amblyomma marmoreum*, *Hyalomma marginatum rufipes* and *Hyalomma truncatum* were the predominant species. A second study by Fourie *et al.* (1996) in the south west region revealed that *Ixodes rubicundus* and *Hyalomma marginatum rufipes* were the most prevalent tick species.

In KwaZulu-Natal, Baker *et al.* (1989) observed that *Boophilus decoloratus*, *Rhipicephalus appendiculatus* and *Rhipicephalus evertsi evertsi* were the most prevalent species on cattle raised on commercial farms. *Hyalomma marginatum rufipes*, *Rhipicephalus appendiculatus* and *Rhipicephalus evertsi evertsi* were the most numerous species in the Limpopo Province. Also present in the Limpopo province were *Amblyomma hebraeum*, *Boophilus decoloratus*, *Hyalomma truncatum* and *Rhipicephalus simus* (Schroeder, 1980; Horak, 1982). Bryson *et al.* (2002) noted that the adults of *Amblyomma hebraeum*, *Rhipicephalus appendiculatus* and *Rhipicephalus evertsi evertsi* were the most numerous tick species in North West Province, while in Mpumalanga *Boophilus decoloratus* constituted more than 75% of the total tick population.

In a five year survey conducted in the Eastern Cape Province, *Rhipicephalus* (*Boophilus*) *decoloratus*, *Amblyomma hebraeum*, *Rhipicephalus appendiculatus* and *Rhipicephalus evertsi*

evertsi were found to be the most common tick species infesting cattle (Rechav, 1982). In contrast to these early findings, Horak (1999) observed *Amblyomma hebraeum*, *Haemaphysalis silacea*, *Rhipicephalus appendiculatus* and *Rhipicephalus glabroscutatum* to be most prevalent tick species on yearling commercial cattle on Valley Bushveld. Muchenje *et al.* (2008a) in an on-station study comparing tick loads on Nguni, Angus and Bonsmara steers on sweet rangeland revealed that *Boophilus decoloratus*, *Amblyomma habraeum*, *Rhipicephalus evertsi evertsi* and *Hyalomma* species were the most common tick infestations. With the exception of *Hyalomma* species, the same tick species composition observed by Muchenje *et al.* (2008a), was found to infest cattle and goats in the communal areas of the Eastern Cape (Nyangiwe and Horak, 2007).

Research on the tick species affecting cattle belonging to small-holder farmers in South Africa is limited. Many studies have focused on cattle in the commercial farming system and it is apparent that cattle management and tick control in this farming sector will differ considerably to that in communal farming areas (Bryson *et al.*, 2002). Few studies have focused on cattle kept by resource-poor farmers in communal areas. These studies however did not compare the prevalence of the parasites in different breeds kept under communal farmer management and across different rangelands types. Other studies have focused on comparing tick loads in indigenous and exotic beef breeds under controlled conditions (Norval *et al.*, 1996; Muchenje *et al.*, 2008a). No studies have focused on the comparison of tick loads in the indigenous and non-descript cattle under communal grazing management. Information on the tick loads of cattle can be used in conjunction with sero-diagnostic methods to estimate and compare the level of resistance of different cattle breeds to ticks (Wambura *et al.*, 1998; Mattioli *et al.*, 2000).

Tick distribution and occurrence differs with geographic distribution and vegetation type (Mtshali *et al.,* 2004). No efforts have been made to compare tick loads in cattle on sweet and sour rangelands. Sour rangeland occurs in areas with high water supply and denser vegetation cover (Ellery *et al.,* 1995) and it is more likely to have higher prevalence of ticks than the sweet rangeland which occurs in areas with low water supply and sparse vegetation cover. Comparing the prevalence of ticks in different rangeland types assists policy makers to design appropriate control programmes for each particular rangeland type.

2.5 Pathogenic effects of ticks

Direct effects of ticks on cattle are tick worry, blood loss, damage to hides and skins of animals and introduction of toxins (de Castro, 1997).

2.5.1 Tick worry

Tick worry is a generalized state of unease and irritability of cattle severely infested with ticks, often leading to serious loss of energy and weight. This negative effect on the growth of animals and their production is thought to be due to the effects of a toxin in the saliva of ticks (Hunter, 2004). Moderate to heavy tick infestations can impact negatively on the growth and production of cattle. Infestations with *Rhipicephalus* (*Boophilus*) *decoloratus* and *Rhipicephalus appendiculatus* were reported to cause weight losses of 1.5 g and 4.4 g, respectively (Norval *et al.*, 1988) while *Amblyomma* spp. resulted in losses of about 63 g (Stachurski *et al.*, 1993). Similarly, milk production was reduced by 9 g per each engorging *Rhipicephalus appendiculatus* female in indigenous Sanga cattle (Norval *et al.,* 1997)

2.5.2 Anaemia

Anaemia is an inevitable consequence of heavy infestation by any blood-feeding parasite, and cattle deaths attributable to anaemia as a result of tick infestation are common (Jonsson, 2006). Engorging ixodid females will increase their weight by 100–200 times but the actual amount of blood ingested is much greater than this, as blood meal is concentrated and fluid excreted in saliva (Kemp *et al.*, 1982). The anaemia caused by heavy tick infestation results in loss of condition in cattle causing a reduction in meat production and milk yield (Gates and Wescott, 2000).

2.5.3 Wounds and myiasis

The mouthparts of ticks puncture the skins during feeding, causing damage to the hide, the damage taking the form of small rounded areas of necrosis, which is often followed by secondary fly attack resulting in serious skin infection (Gracey *et al.*, 1999). Ticks with longer mouthparts such as *Amblyomma* and *Hyalomma* cause more extensive damage than those with shorter mouthparts such as *Boophilus* and *Rhipicephalus*. The involvement of host reactions leading to tissue damage may be dependent upon recruitment of inflammatory responses characterized by dermal cell infiltrates which form the lesions (Mattioli *et al.*, 2000). Tick wounds may become infested by screwworms or other agents of myiasis, and are also associated with the spread of bovine dermatophilosis caused by *Dermatophilus congolensis* (Kahn, 2006).

2.5.4 Toxicoses

Tick saliva contains toxins which have a specific pathogenic effect. The toxins affect not only the attachment site but also the entire organs of the host. Some ticks produce neurotropic toxins

which induce tick paralysis that is characterized by an acute ascending flaccid motor paralysis (Kahn, 2006). Examples are paralysis caused by the feeding of *Dermacentor andersoni*, Australian tick paralysis caused by *Ixodes holocylus*, and tick toxicosis caused by *Rhipicephalus* species (Drummond, 1983). Females of the species *Hyalomma truncatum* produce a dermotropic (epitheliotropic) toxin which causes sweating sickness in calves and some adult cattle (Kahn, 2006).

2.6 Use of tick-resistant breeds in tick control

In many sub-tropical and semi-arid environments in Africa indigenous dual purpose breeds are highly resistant to ticks, resulting in low infestation rates that cause significant reductions in direct losses (Norval *et al.*, 1991). Resistance to tick infestation in cattle varies both between breeds and between individuals. Within-breed variation in genetic resistance to ticks can potentially be used to breed for resistance (de Castro, 1997). The phenomena of host resistance to ticks and enzootic stability to tick born diseases are well documented (Latif and Pegram, 1992; Solomon and Kaaya, 1998; Mattioli *et al.*, 2000).

The ability to resist ticks is acquired, but it develops according to genetic factors. The host genes that may play a role in the manifestation of tick resistance have yet to be identified. However DNA markers that may be used for diagnosis of tick resistance/susceptibility have been identified (Olafson *et al.*, 2007). It may thus be possible to breed cattle for tick resistance and reduce the need for use of costly and potentially harmful acaricides. The simplest form of utilisation of host genetic resistance is cross-breeding susceptible exotic cattle with indigenous tick-resistant breeds (de Castro, 1997). It is imperative however, to compare the level of

resistance, by comparing tick counts, of the indigenous-exotic crosses with that of the indigenous breeds so as to assess the benefit of cross-breeding. It is also important to determine whether breeding for tick resistance can be compatible with breeding for particular production characteristics such as meat or milk yield.

2.7 Tick-borne diseases

Tick-borne diseases cause probably the most economically serious losses of ruminants in southern Africa. They cause direct losses, such as mortality, reduction in meat and milk yield, and indirectly through the institution of control measures (Makala *et al.*, 2003). The TBDs of economic significance in the communal areas of South Africa are babesiosis, anaplasmosis and heartwater (Dreyer *et al.*, 1998; Mbati *et al.*, 2002).

2.7.1 Babesiosis

Babesia bovis and *Babesia bigemina* are the main causal agents of bovine babesiosis in South Africa (Coetzer *et al.*, 1994). The symptoms of the acute form of the disease include anaemia, fever, haemoglobinuria, ataxia, high parasitaemia, and sometimes death (Bock *et al.*, 2004). Animals that recover from primary infection become carriers; in these animals, parasitaemia is virtually undetectable on microscopy. Subclinical infections may endure for long periods (Brown *et al.*, 2006) with infected animals acting as reservoirs. Babesiosis tends to be more important in non-resistant exotic animals, although *B. bovis* infections are very severe and even local breeds of cattle can be greatly affected by *B. bigemina* under conditions of poor health or nutrition (Minjauw and McLeod, 2004). Specific and sensitive diagnostic methods can be used to monitor the prevalence of these infections for efficient control strategies to be implemented. Serological

methods are useful for epidemiological studies of bovine babesiosis (Buling *et al.*, 2007). Although the distribution of babesiosis in the Eastern Cape can be determined by the vector distribution (Coetzer *et al.*, 1994), its prevalence in the different breeds of cattle on communal rangelands still needs to be determined.

2.7.2 Anaplasmosis

Anaplasma marginale (Rickettsiales: Anaplasmataceae) is the causative agent of bovine anaplasmosis worldwide (Kocan *et al.*, 2004). The clinical symptoms of bovine anaplasmosis may include fever, weight loss, abortion, lethargy, icterus, and often death in animals older than two years (de Waals, 2000). Cattle that survive acute infection develop persistent infections characterized by cyclic low-level rickettsaemia (French *et al.*, 1998; French *et al.*, 1999). Persistently infected or "carrier" cattle have lifelong immunity and are resistant to clinical disease on challenge exposure. *Bos taurus* breeds (Holstein, Brown Swiss, or Hereford) are more likely to develop acute anaplasmosis than Zebu cattle and their crossbreeds. Most of the cattle farming areas in South Africa, including the Eastern Cape, occur in the endemic and epidemic areas of anaplasmosis (de Waals, 2000). Serological tests have been developed for the evaluation of anaplasmosis and these are useful in the development of preventive measures (Barros *et al.*, 2005). Despite the availability of these tests, the prevalence of anaplasmosis in cattle reared on the communal rangelands in the Eastern Cape remains unknown.

2.7.3 Heartwater

Heartwater is caused by the rickettsial organism *Ehrlichia* (*Cowdria*) *ruminantium* and is transmitted by ticks of the genus *Amblyomma*. It is causes heavy losses in cattle in southern

Africa (Coetzer *et al.*, 1994; Makala *et al.*, 2003). Heartwater occurs in four different clinical forms, namely peracute, acute, subacute and subclinical, determined by variations in susceptibility of the hosts and the virulence of various strains of the heartwater agent (Uilenberg, 1983). Exotic and 'naïve' indigenous cattle are more severely affected by heartwater than cattle from endemic areas. The distribution of heartwater in South Africa is limited to the occurrence of the tick vector (Coetzer *et al.*, 1994). In the Eastern Cape researchers have shown that the *Amblyomma* tick vector and heartwater occur under the commercial production system, there is a need to determine the prevalence of heartwater in the inland communal areas of the province.

2.8 Serological techniques for the diagnoses of tick-borne diseases

Direct and indirect methods have been developed for the diagnosis of TBDs of livestock. The direct method involves identifying the parasite in Giemsa-stained blood smears or lymph-node biopsy samples. Direct methods are good for clinical diagnosis but less useful for determining the prevalence of TBDs, and, therefore, of little use for epidemiological surveys (Minjauw and McLeod, 2004). Indirect methods based on serology have been developed and give a more accurate diagnosis of TBDs. The indirect tests that can be used to detect and screen for TBD infections in cattle in South Africa include the immunofluorescent antibody test (IFAT), enzyme-linked immuno-sorbent assay (ELISA) and nucleic acid based tests.

The indirect immunofluorescent antibody test (IFAT) is the standard test that is used to detect antibodies to *Babesia* parasites of cattle. This test has a high sensitivity and good specificity and is reproducible (Krause *et al.*, 1994; Hunfeld *et al.*, 2002). Titres from 1:32 to 1:160 are diagnostic and specific, with positive predictive values of 69–100% and negative predictive

values of 96–99% (Hunfeld *et al.*, 2002). The IFAT is still used as the 'gold standard' to evaluate the sensitivity and specificity of other serological tests in the diagnosis of babesiosis (Ravindran *et al.*, 2007). Its major disadvantages are low sample throughput and subjectivity.

In the last decades, enzyme linked immuno-sorbent assay (ELISA) has replaced IFAT. The ELISA technique has advantages such as possibility of analysis of a large number of tests in a shorter time and the discrimination of positive from negative sera without subjectivity (Madruga *et al.*, 2000). The complement ELISA which uses major surface protein-5 (MSP-5) as recombinant antigen (Molloy *et al.*, 1998), has been developed for the sero-diagnosis of *Anaplasma marginale*. This test has a specificity of 94 % and sensitivity of 99 % (Ndungu *et al.*, 1995). It is thus useful for serological surveys of epidemiological studies and as an evaluation tool in deciding the preventive measures to be used (Barros *et al.*, 2005). It has been suggested that application of ELISA for *B. bigemina* is still unreliable until a more purified *Babesia*-specific antigen or specific monoclonal antibodies are available (El-Ghaysh *et al.*, 1996).

Other tests involving nucleic acid probes or the polymerase chain reaction (PCR) can also be used in the diagnosis of TBD (Calder *et al.*, 1996; Ravindran *et al.*, 2006). The PCR technique is a sensitive method and is also very specific. However, great care has to be taken to prevent contamination by extraneous DNA. Since most nucleic acid probes are radioactively labelled, the technique requires laboratories of the highest standard, equipped for handling radioactive materials. To overcome the limitation of low sample throughput caused by gel electrophoresis detection of the PCR product, a relatively new method that couples the PCR with ELISA has been developed (Thammasirirak *et al.*, 2003). The PCR-ELISA has been used for the detection

of Babesia bovis and has been shown to be highly sensitive and has a high sample throughput (Thammasirirak *et al.*, 2003).

The IFAT will be used to test for antibodies to *Babesia* while the ELISA will be used to test for antibodies to *Anaplasma* in this study. These are presently the most reliable and available tests for the diagnosis of TBD in South Africa.

2.9 Summary

Ticks and TBDs are important constraints to communal cattle production. Information regarding the tick species, tick loads and TBDs affecting cattle in communal areas of the Eastern Cape of South Africa is limited. There is a need, therefore, to determine the tick species composition and prevalence across rangeland types, seasons and cattle breeds that occur in the communal areas. The main objective of the present study was to determine the prevalence of ticks and TBDs in cattle in the communal areas of the Eastern Cape Province of South Africa.

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Chapter 3: Tick loads and prevalence in Nguni and non-descript cattle on communal rangelands of the Eastern Cape

Abstract

The objective of this study was to compare tick loads and prevalence in Nguni and non-descript cattle in the sweet and sour communal rangelands of the Eastern Cape Province, South Africa. Engorged adult female ixodid ticks were collected and identified seasonally from 144 cattle raised on sweet and sour rangelands from August 2007 to April 2008. Three tick species were identified in the sweet and sour rangelands, namely *Rhipicephalus appendiculatus*, *Rhipicephalus* (*Boophilus*) species and *Rhipicephalus evertsi evertsi* with prevalences of 71.1, 29.2 and 40.2 %, respectively. *Hyalomma* species (19.0 %) occurred only in the sour rangeland. Higher tick counts were recorded in the hot-wet season than in the cool-dry season ($P < 0.05$). Cattle in the sweet rangeland had significantly lower tick loads than those in the sour rangeland in all the seasons except the hot-dry season. The Nguni breed had lower ($P < 0.05$) tick loads of *Rhipicephalus appendiculatus* in the hot-wet and post-rainy season and *Hyalomma* species in all seasons than the non-descript cattle. The use of a tick resistant Nguni breed in the integrated control of ticks on cattle in the communal areas of South Africa is recommended.

Key words: indigenous cattle, *Rhipicephalus appendiculatus*, hot-dry season, sour rangeland

3.1 Introduction

Ticks and tick-borne diseases are ranked as the most important cattle health constraints by farmers in the communal areas of South Africa (Dreyer *et al.*, 1999; Dold and Cocks, 2001).

Several studies have been conducted on the ticks of cattle in South Africa and many of these have concentrated on the commercial production system (e.g. Rechav, 1982; Horak *et al.*, 1991; Horak, 1999). Tick control programmes in the commercial farming sector differ considerably to those in the communal farming areas (Bryson *et al.*, 2002). Commercial farmers rely on intensive tick control using acaricides while resource poor farmers cannot afford commercial acaricides and resort to using traditional medicines to control ticks (Hesterburg *et al.*, 2007). There is a need to identify common ticks and determine their prevalence and loads in cattle on communal rangelands in South Africa to formulate and implement appropriate tick control strategies.

Some studies have compared tick loads in indigenous and exotic beef breeds on controlled on-farm conditions (Norval *et al.*, 1996; Muchenje *et al.*, 2008). No studies, however, have been carried out to compare the tick loads of the Nguni breed and indigenous-exotic crosses (non-descript cattle), raised under communal grazing management. It is important to identify and recommend breeds that are resistant to ticks and can be used by farmers on communal rangelands. Information on the tick loads of cattle can be used to estimate and compare the level of resistance of different cattle breeds to ticks (Wambura *et al.*, 1998; Mattioli *et al.*, 2000).

Tick occurrence and tick loads vary with seasons, geographic location, vegetation type, breed and age of the animal (Mtshali *et al.*, 2004). There are little, if any efforts that have been made to compare seasonal dynamics of ticks in different cattle breeds on sweet and sour rangelands, which are likely to vary due to differences in rainfall distribution and vegetation densities (Ellery, 1995). Comparing the prevalence and loads of ticks in different rangeland types assists

policy makers to design appropriate control programmes for each rangeland type. The objective of the current study was to compare tick loads of Nguni and non-descript cattle kept on the communal sweet and sour rangelands of the Eastern Cape Province of South Africa.

3.2 Materials and Methods

3.2.1 Description of study sites

Tick collection was conducted in Magwiji, Ukhahlamba district, representing the sweet rangeland and Cala, Chris Hani district, representing the sour rangeland. Both sites are found in the Eastern Cape Province, South Africa.

Magwiji is located on 30°37′ S and 27°22′ E and lies at an altitude of 1507 m above sea level. The climate varies from hot-wet to extreme cold with heavy frost and snowfall along the mountain area. Average annual rainfall is less than 500 mm in the hot-wet season and less than 200 mm with frost and snow in the cool-dry season. Highest mean temperature is recorded in January (22°C) and lowest in July (9°C). The most common grass species are *Themeda triandra*, *Setaria sphacelata*, *Microchloa caffra*, *Elionurus muticus* and *Heteropogon contortus* (Acocks, 1988). This rangeland type is referred to as sweet rangeland because forages retain palatability and high nutrient content throughout the year (Ellery et al., 1995). The slope and soil depth ranges between 3.1 and 5.0 % and 501-700 mm, respectively. Soils are generally sandy with the clay content ranging from 15 to 24.9 % and silt content from 15 to 20 %, soil organic content ranges between 0.6 and 2 %. The soil pH is within the range of 6.5 and 7.5.

Cala is located on 31°33′ S and 27°36′ E with an altitude of 1441 m above sea level. It receives moderate rainfall of 600 – 800 mm in the hot-wet season (November to April) and low rainfall of 200 mm in the cool-dry season (mid-May to October). Average monthly temperature is highest in January (20°C) and lowest in July (11°C). The most common grass species are *Themeda triandra*, *Heteropogon contortus*, *Sporobolus africanus* and *Microchloa ciliate*. *Euryops pyroides*, *Chrysocoma ciliate* and *Dyspyrose scrabrida* are the common bush species in the areas (Lesoli, 2008). This rangeland type is referred to as sour because the forages lose palatability and nutrient content during the dry season. Soil clay content ranges between 15 to 24.9 %, silt content from 20.1 to 30 %, and soil organic content between 1.0 and 2 %. The soil pH is within the range of 5.6 and 6.5.

3.2.2 The study animals

A total of 144 (72 from each rangeland type) cattle of different ages based on dentition, both sexes and two breeds, Nguni and non-descript crosses produced by indiscriminate crossing of indigenous Nguni cattle with exotic breeds, were initially selected at the beginning of the study as shown in Table 3.1. It was intended to monitor these cattle for four seasons (1 year). However, the numbers decreased in the hot-wet and post-rainy season due to sales and slaughtering. The animals were selected on the basis of the owners' willingness to participate in the study and assurance of the availability of the cattle throughout the study period. All the selected animals were ear tagged at the beginning of the study for easy identification. The cattle were grazed on communal rangelands and not dipped throughout the study period.

Table 3.1: Composition of the study animals

		Sweet rangeland							
		Cold-dry		Hot-dry		Hot-wet		Post-rainy	
	Breed[a]	NG	ND	NG	ND	NG	ND	NG	ND
Age (years)									
1-2		6	7	6	7	6	7	6	6
>2-3		7	7	6	7	5	6	5	5
>3-4		8	8	7	7	6	6	6	5
>4-5		7	8	7	7	7	7	6	7
>5		7	7	7	7	6	6	6	6
		Sour rangeland							
		Cold-dry		Hot-dry		Hot-wet		Post-rainy	
	Breed	NG	ND	NG	ND	NG	ND	NG	ND
Age (years)									
1-2		7	7	7	7	6	6	5	5
>2-3		6	7	6	7	6	6	5	6
>3-4		7	8	6	7	5	6	5	6
>4-5		8	8	7	8	7	7	6	6
>5		7	7	7	7	6	6	6	6

[a] NG = Nguni breed and ND = non-descript breed

3.2.3 Tick collection and identification

Engorged adult ixodid ticks were collected once in the cool-dry (August 2007), hot-dry (October 2008), hot-wet (January 2008) and post-rainy (April 2008) seasons. The samples were collected between about 08h00 and 11h00. The ticks were collected from the head, ears, neck, belly, back, legs, perineum and tail of each animal. Collected samples were placed in sample vials containing 6 % formalin mixed with 3 % glycerine and labelled. The label contained the name of the community, owner's name, animal identification code, date and month of collection. The ticks were identified based on morphological and structural differences of the adult ticks of each species by a qualified veterinarian at the Animal Science Laboratory, University of Fort Hare. The grouping to their genus and species was made according to the methods developed by Hoogstraal (1956) and Horak *et al.* (2002). Prevalence for each tick species was calculated as:

$$P = \frac{d}{n} \times 100$$

where P represents the prevalence;

d represents the number of animals that tested positive for a particular tick species; and

n represents the total number of animals sampled (Thrusfield, 1995).

3.2.4 Statistical analyses

The tick counts were transformed according to the following formula $y = \log 10 (x+1)$ to confer normality. The data was analysed using SAS (2003). Specifically the chi-square test was used to determine associations between tick prevalence and rangeland type, breed, season, sex, age and their interactions. Frequencies were determined using PROC FREQ of SAS (2003). The effect of rangeland type, breed, season, sex, age, position and their interactions on tick counts was

determined using the generalised linear model procedures for repeated measures (SAS, 2003). Pair wise comparisons of means were performed using the PDIFF option.

3.3 Results

3.3.1 Tick prevalence

A total of 1034 ticks were collected from cattle in both the sweet and sour rangelands. Three tick species were identified on both sweet and sour rangeland, viz *Rhipicephalus appendiculatus* (77.1 %), *Rhipicephalus evertsi evertsi* (40.2 %) and *Rhipicephalus* (*Boophilus*) *decoloratus* (29.2). A fourth, *Hyalomma* species (19.0 %), occurred only on the sour rangeland.

Table 3.2 shows the seasonal prevalence of ticks in cattle herds in the sweet and sour rangelands. Season was significantly associated with the prevalence of all the tick species identified. The highest ($P < 0.05$) prevalence was observed in the hot-wet season for *R. appendiculatus*, *R.* (*Boophilus*) *decoloratus* and *Hyalomma* species, and in the hot-dry season for *R. evertsi*, while the lowest ($P < 0.05$) prevalence occurred in the cool-dry season for all the tick species. There was a significant association between rangeland type and season on the prevalence of all tick species observed. The sweet rangeland had lower ($P < 0.05$) tick prevalence than sour rangeland for all the tick species in all the seasons.

The association between age of animal and tick prevalence across both the sweet and sour rangelands is shown in Table 3.3. Cattle greater than 4 to 5 years old had higher ($P < 0.05$) tick prevalence than all the other age groups, while cattle younger than two years had the lowest ($P <$

0.05) tick prevalence. Breed and sex of cattle were not significantly associated with prevalence of all the tick species identified.

Table 3.2: Seasonal prevalence (%) of ticks in cattle herds in the sweet and sour rangelands of the Eastern Cape Province across the two breed types

	Sweet rangeland				Sour rangeland				P value
Identified tick species	Cool-dry	Hot-dry	Hot-wet	Post-rainy	Cool-dry	Hot-dry	Hot-wet	Post-rainy	
R. appendiculatus	44.6	80.4	89.0	71.1	49.0	86.5	95.3	88.9	*
Boophilus	7.7	14.9	42.7	56.6	11.8	17.5	42.9	72.1	*
R. evertsi	47.1	55.4	23.8	25.6	73.8	55.7	28.9	48.8	*
Hyalomma	0.0	0.0	0.0	0.0	6.0	37.8	52.4	9.3	*
Overall	72.5	89.7	92.7	80.2	81.5	91.9	95.3	92.1	*

*$P < 0.05$

Table 3.3: Tick prevalence in cattle of different age groups on communal grazing in the Eastern Cape Province across breed types

	Prevalence (%)					Significance
Age (years)	1-2	>2-3	>3-4	>4-5	>5	
R. appendiculatus	9.0	9.0	15.0	26.6	11.3	*
Boophilus	4.1	3.2	6.9	11.3	4.3	NS
R. evertsi	4.3	6.1	6.9	54.5	36.5	*
Hyalomma	0.9	2.9	5.5	6.1	2.9	*

*$P < 0.05$.

NS: not significant.

3.3.2 Effect of rangeland type, season, breed, age, sex and position on tick loads

Table 3.4 shows the seasonal changes in the overall tick load and tick loads of the four ticks species identified in the sweet and sour rangelands. Rangeland type, season, position and the interaction of rangeland type and season significantly ($P < 0.05$) affected the overall tick load. The sweet rangeland had significantly lower ($P < 0.05$) overall tick load than sour rangeland in all the seasons. The lowest ($P < 0.05$) overall tick loads were observed in the sweet rangeland during the cool-dry season (0.44 ± 0.081) while the highest occurred in the sour rangeland during the hot-wet season (1.23 ± 0.082). Breed, age, sex, and the interactions of season and breed, rangeland type and breed, age and sex, and rangeland type, season and breed did not affect ($P > 0.05$) the overall tick load. As shown in Figure 3.1 the most common sites of attachment of ticks on the study animals were, in descending order, the perineum, ears and belly.

3.3.2.1 Rhipicephalus appendiculatus

The tick loads of *R. appendiculatus* were significantly affected ($P < 0.05$) by rangeland type, season, position and the interactions of rangeland type and season, and season and breed. Cattle in the sweet rangeland had significantly lower ($P < 0.05$) tick loads of *R. appendiculatus* in all the seasons except the cool-dry season in which the tick loads were similar ($P > 0.05$). As shown in Table 3.4, the Nguni breed had lower ($P < 0.05$) tick loads for *R. appendiculatus* in the hot-wet and post-rainy seasons than the non-descript breed. The perineum had the highest ($P < 0.05$) tick loads of *R. appendiculatus* followed by the ears and the neck (Figure 3.1). Breed, age, sex and the interactions of rangeland type and breed, sex and age, and rangeland type, season and age did not affect ($P > 0.05$) the tick loads of *R. appendiculatus*.

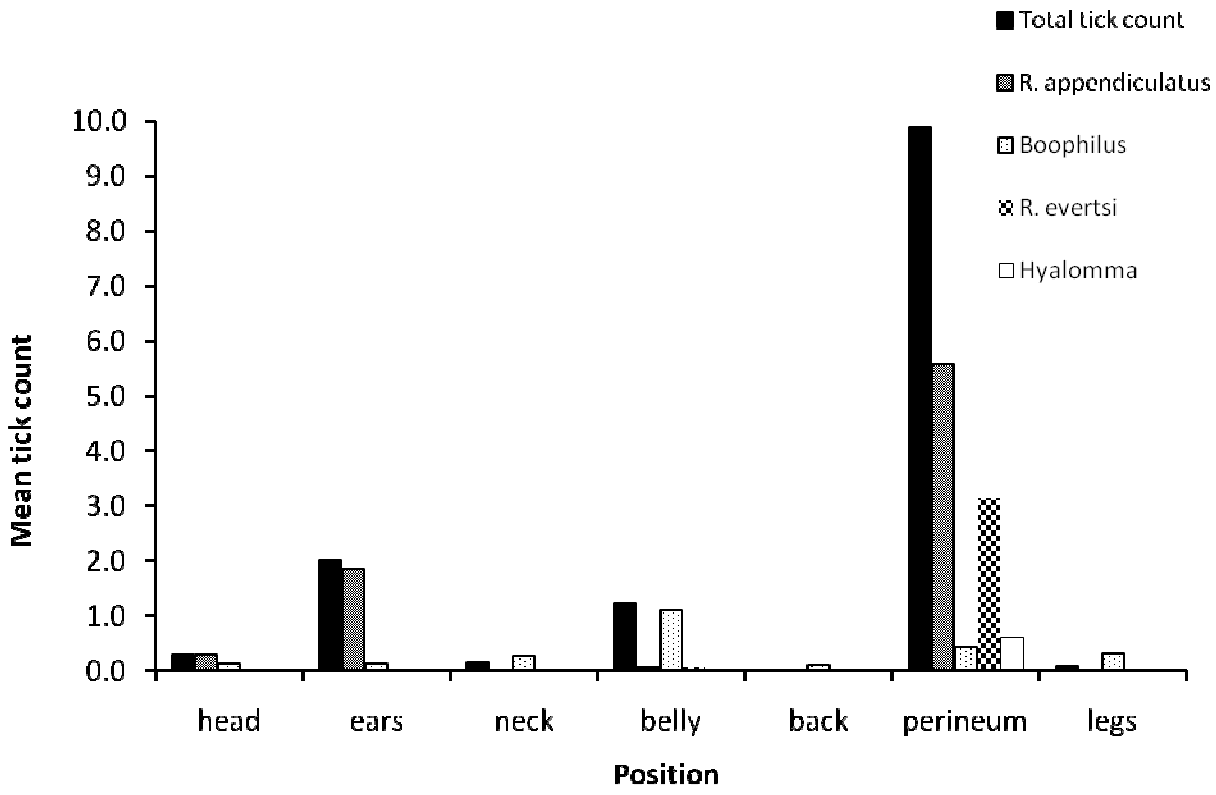

Figure 3.1: Tick loads per position in Nguni and non-descript cattle on the communal rangelands

Table 3.4: Seasonal changes in mean tick loads in Nguni and non-descript cattle

		Seasons			
		Cool-dry	Hot-dry	Hot-wet	Post-rainy
Tick species	**Breed**				
R. appendiculatus	Nguni	0.48 ± 0.074[a]	0.58 ± 0.082[b]	0.72 ± 0.086[c]	0.48 ± 0.091[a]
	Non-descript	0.43 ± 0.063 [a]	0.59 ± 0.066 [b]	0.99 ± 0.081 [d]	0.66 ± 0.069[bc]
Boophilus	Nguni	0.12 ± 0.052[a]	0.10 ± 0.058[a]	0.29 ± 0.06[b]	0.37 ± 0.065[b]
	Non-descript	0.07 ± 0.045[a]	0.11 ± 0.047[a]	0.30 ± 0.057[b]	0.52 ± 0.049[c]
R. evertsi	Nguni	0.29 ± 0.065[b]	0.39 ± 0.072[c]	0.19 ± 0.076[a]	0.15 ± 0.080[a]
	Non-descript	0.30 ± 0.056[b]	0.38 ± 0.058[c]	0.17 ± 0.071[a]	0.25 ± 0.060[b]
Hyalomma	Nguni	0.00 ± 0.002[a]	0.00 ± 0.003[a]	0.00 ± 0.004[a]	0.00 ± 0.002[a]
	Non-descript	0.01 ± 0.003 [a]	0.16 ± 0.033 [c]	0.27 ± 0.040 [c]	0.11 ± 0.030[ab]

[abcd] Values with different superscripts for each tick species are different ($P < 0.05$).

3.3.2.2 Rhipicephalus (*Boophilus*) species

Rangeland type, season, position and the interaction of rangeland type and season significantly affected ($P < 0.05$) the tick loads of *R.* (*Boophilus*) species. Cattle in the sweet rangeland had significantly lower ($P < 0.05$) tick loads of *R.* (*Boophilus*) species in all the seasons except the cool-dry season in which the tick loads were similar ($P > 0.05$). The belly had the highest ($P < 0.05$) tick infestation followed by the perineum, neck and legs (Figure 3.1). Breed, age, sex and the interactions of rangeland type and breed, sex and age, and rangeland type, season and age did not affect ($P > 0.05$) the tick loads of *R. appendiculatus*.

3.3.2.3 Rhipicephalus evertsi evertsi

Rangeland type, season, position and the interaction of rangeland type and season significantly affected ($P < 0.05$) the tick loads of *R. evertsi*. Significantly lower ($P < 0.05$) tick loads of *R. evertsi* were observed in the sweet rangeland in the cool-dry and post rainy season. Only two positions were infested by *R. evertsi*, the perineum which had the highest ($P < 0.05$) tick loads and the belly (Figure 3.1). Breed, age, sex and the interactions of rangeland type and breed, sex and age, and rangeland type, season and age did not affect ($P > 0.05$) the tick loads of *R. evertsi*.

3.3.2.4 Hyalomma species

Tick loads of *Hyalomma* species were significantly affected by rangeland type, season, position and the interaction of rangeland type and season. *Hyalomma* species only occurred on cattle on the sour rangeland where the highest ($P < 0.05$) loads were observed in the hot-wet season and the lowest ($P < 0.05$) in the cool-dry season. The perineum was the only position of attachment

of *Hyalomma* species (Figure 3.1). The tick loads of *Hyalomma* species were not affected ($P > 0.05$) by breed, age and sex.

3.4 Discussion

Four tick species, *R. appendiculatus*, *R.* (*Boophilus*) species, *R. evertsi evertsi* and *Hyalomma* species were identified in the study. This is in agreement with Muchenje *et al.* (2008) who, except for *Amblyomma hebraeum*, found similar species composition infesting cattle on a sweet rangeland in the Eastern Cape Province. The absence of *Amblyomma hebraeum* in the present study may be attributed to the fact that this tick occurs in the warm, moist coastal areas of the Eastern Cape (Coetzer *et al.*, 1994) and so was not observed in the study sites which are located inland.

The differences in tick prevalence and species distribution observed in the sweet and sour rangelands were most likely influenced by differences in the vegetation composition and cover, humidity and annual rainfall (Randolph, 1997; Wesonga *et al.*, 2006). Sour rangeland occurs in areas with high rainfall and denser and tall vegetation cover (Ellery *et al.*, 1995) and thus had higher prevalence of ticks than the sweet rangeland which occurs in areas with low rainfall and sparse and short vegetation cover. The distribution of *Hyalomma* species is limited by winter frost (Walker, 1994). Thus, the extremely cold conditions that occur during the cool-dry season and in the early morning and late evening in other seasons in the sweet rangeland may have caused the absence of this species from cattle in sweet rangeland.

The higher tick prevalence and tick loads observed in the hot-wet season than in the cool-dry season could be attributed to the more conducive conditions for tick proliferation and survival during this season. Accelerated tick proliferation occurs when environmental temperatures and humidity are high (Chilton and Bull, 1994; Chilton *et al.*, 2000; Zeleke and Bekele, 2004). The present study's findings agree with Webb and David (2002), Wesonga *et al.* (2006) and Muchenje *et al.* (2008) who observed high tick counts during the hot-wet season.

Nguni cattle carried markedly lower tick loads of *R. appendiculatus* and *Hyalomma* species than the non-descript cattle during the hot-wet and post-rainy seasons, suggesting that indigenous Nguni cattle could have a higher innate and/or acquired resistance. This agrees with Scholtz *et al.* (1991), Norval *et al.* (1996) and Muchenje *et al.* (2008) who observed the Nguni breed to have lower tick counts compared to exotic and synthetic breeds. Even though the mechanism of tick resistance is not fully understood, it could be related to a pre-immunity to ticks often established through continuous contact with the infectious agents from early in life (Mattioli *et al.*, 2000). Avoidance behaviour, skin hypersensitivity and increased grooming (Meltzer, 1996) may also contribute to increased resistance of the Nguni breed to ticks.

The lower tick loads observed in the younger stock could be attributed to some form of innate protection that declines with age (Wickel and Bergman, 1997). It is possible that continuous selective grooming of the younger animals by their respective dams (Fivaz and de Waal, 1993) may have resulted in the lower tick loads in the younger stock. In addition, older animals have a larger surface area predisposing them to higher tick infestations than younger animals. The large

surface area in older animals agrees with Swai *et al.* (2005) who observed that mature animals have higher odds of carrying ticks than young stock due to their larger body surface area.

The differences in the common attachment sites among the four tick species in the study suggest preferential feeding behaviour in the different tick species. Three tick species, *R. evertsi, R. appendiculatus* and *Hyalomma* species most commonly infested the perineum while *R.* (*Boophilus*) species infested the belly. The feeding site of ticks may have been influenced by attractant odours from the various predilection sites especially the perineum (Wanzala *et al.*, 2004). The higher tick infestations on the perineum could also be ascribed to the fact that ticks prefer warm, moist and hidden sites with a good vascular supply and thin skin (Muchenje *et al.*, 2008). All these current study's findings are in agreement with those of Spicket *et al.* (1989) and Muchenje *et al.* (2008), who reported high tick infestations in secluded sites with less hair.

Tick prevalence was observed to be generally high and this is an indication of the tick problem in the Eastern Cape. The high tick loads in the hot-wet and post-rainy seasons across rangeland types and breeds warrants more frequent use of efficacious acaricides during these periods to avert major cattle losses through deaths and loss of productivity. There is a need to develop strategic tick control practices to abate heavy tick loads while encouraging the development of endemic stability in communal cattle herds. Indigenous Nguni cattle are recommended for use in the integrated control of ticks in the communal areas of South Africa as they are more resistant to tick infestations than non-descript cattle. The use of good grazing management practices, such as rotational grazing, rather than the current uncontrolled extensive grazing, is also recommended to

reduce rangeland infestivity to cattle. Indications from the current study suggest a need for more intensive tick control in the sour than the sweet rangelands.

3.5 Conclusions

Rhipicephalus appendiculatus, *Rhipicephalus* (*Boophilus*) *decoloratus*, *Rhipicephalus evertsi evertsi* were the most prevalent tick species. *Hyalomma* species was only found in the sour rangeland. Tick prevalence and loads showed a definite seasonal pattern peaking during the hot-wet season and dropping in the cool-dry season. The Nguni breed had lower tick loads of *R. appendiculatus* and *Hyalomma* species in the hot-wet and post-rainy season than the non-descript cattle. Strategic tick control using acaricides during the hot-wet and post-rainy seasons to avert major losses caused by high tick loads, especially in the sour rangeland is recommended. It is recommended that the Nguni breed be used in the integrated control of ticks on cattle in the communal areas of the Eastern Cape. It is important to determine whether the tick loads and prevalences are related to prevalences of tick-borne diseases in the communal areas.

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Chapter 4: Sero-prevalence of tick-borne diseases in Nguni and non-descript cattle on communal rangelands of the Eastern Cape

Abstract

A survey was conducted to determine the sero-prevalence of tick-borne diseases in Nguni and non-descript cattle on the sweet and sour communal rangelands across seasons. Body condition scores, body weights, packed cell volume and antibodies to *Babesia bovis*, *Babesia bigemina*, *Ehrlichia ruminatium* and *Anaplasma marginale* were determined seasonally in 144 cattle raised on communal rangelands from August 2007 to April 2008. Of the 379 samples collected, 44.6% were sero-positive for *B. bovis*, 45.9% for *B. bigemina* and 25.6% for *A. marginale*. All the animals were sero-negative for *Ehrlichia ruminantium*. Nguni cattle had lower ($P < 0.05$) sero-prevalence for *A. marginale* in the cool-dry season and *B. bigemina* in the cool-dry and hot-wet seasons. Cattle in the sweet rangeland had significantly lower sero-prevalence of *B. bovis* and *B. bigemina* in all the seasons. Infection with *B. bovis* and *B. bigemina* negatively affected ($P < 0.05$) body weight and body condition scores while *B. bovis* and *A. marginale* infections significantly affected the packed cell volume. The sero-prevalence of TBD was lower in the Nguni than non-descript breed. Use of the adapted and TBD-resistant Nguni breed on communal rangelands is recommended.

4.1 Introduction

Little is known about the occurrence and prevalence of TBDs in the communal areas of the Eastern Cape, despite their importance. Most data on TBD occurrence is obtained from inferences on the distribution of the tick vector (De Vos, 1979; Regassa *et al.*, 2003) and this

may be erroneous at times as the presence of the vector does not necessarily mean the presence of the disease (Fivaz *et al.*, 1992). The distribution of tick species in a particular area is not static (Tonnensen *et al.*, 2004) thus confounding the extrapolation of TBD occurrence from tick distribution data.

In chapter 3, tick prevalence and loads were shown to be lower in the indigenous Nguni than the non-descript cattle on communal rangelands. Breed differences can be expected in the sero-prevalence of TBDs in the communal cattle based on the tick prevalence results. It is essential to determine the sero-prevalence of TBDs to estimate and compare the level of TBD-resistance in different cattle breeds (Mattioli *et al.*, 2000). The determination of TBD sero-prevalence enables the recommendation of TBD-resistant cattle breeds that can be used for sustainable and profitable production by farmers on communal rangelands. The objective of the current study was to determine the sero-prevalence of TBD in Nguni and non-descript cattle on communal rangelands in the Eastern Cape Province of South Africa.

4.2 Materials and Methods

4.2.1 Description of study sites

Tick collection was carried out in Magwiji and Cala. The description of the sites is detailed in Section 3.2.1.

4.2.2 Study animals

The same animals selected in Chapter 3 were used in this study. The cattle were grazed on communal rangelands and no animal was treated for TBDs throughout the study period. The study was conducted from August 2007 to April 2008.

4.2.3 Determination of body weights and body condition scores

For each animal, the body weight and body condition score (BCS) was determined before blood collection was carried out in each season. Weights were estimated using a cattle weigh-band while visual assessment of the body condition was made using the five-point European system (Edmonson, 1989).

4.2.4 Blood collection

Cattle were held in a race during blood sample collection. Blood samples were collected between 08h00 and 11h00, once in the cool-dry (August 2007), hot-dry (October 2007), hot-wet (January 2008) and post-rainy (April 2008) season. Tail venipuncture was performed using an 18-gauge needle into labelled Vacutainer® blood tubes.

4.2.5 Determination of packed cell volume

For the determination of packed cell volume (PCV) blood was collected in Vacutainer® blood tubes containing EDTA anti-coagulant. The blood was transferred into micro-haematocrit tubes and centrifuged in a micro-haematocrit centrifuge at a relative centrifuge force of 0.169g for three minutes. Reading of the PCV was performed on the Micro-haematocrit Reader Scale.

4.2.6 Serological testing

For serological testing, the blood was collected in empty Vacutainer® tubes and allowed to clot at room temperature for four hours and then centrifuged at a relative centrifuge force of 1.006g for 10 minutes at 25°C. The sera were decanted and stored in clean cryotubes at -10°C till serological testing.

Antibodies to *Babesia bovis*, *Babesia bigemina* and *Ehrlichia ruminatium* were detected using the indirect fluorescent antibody test (IFAT), as described by Joyner *et al.* (1972) and Goldman *et al.* (1972). The method involved making two-fold dilutions of the test and control sera of 1/80, 1/160, using phosphate buffered saline (PBS). Following this, the antigen slides were fixed in cold acetone (kept in freezer 26/FZ/1a) for 1 minute and allowed to air dry on the work bench. Diluted serum samples were loaded on the slide in serial dilutions starting from the marked end of the slide. The slides were incubated in a humid chamber – cotton wool soaked with tap water to prevent the diluted serum sample on the antigen slide from drying, at approximately 37^{o}C for 1 hour. After incubation, the slides were washed with PBS on a magnetic stirrer set at 70 r.p.m for 10 minutes then in distilled water for 5 minutes. A conjugate was made up by diluting Sigma FITC conjugated anti-bovine in PBS at approximately 1/80. A 25 µl drop of diluted conjugate was placed on each well on the antigen slides to completely cover the well. The slides were incubated in a humid chamber – cotton wool soaked with tap water to prevent the diluted serum sample on the antigen slide from drying, at approximately 37^{o}C for 1 hour. After incubation, the conjugate was flicked off from the slide and the slides were washed in PBS for 10 minutes on a magnetic stirrer as described above. The slides were left to air dry on the work bench. A drop of 50 % glycerine/PBS was then placed on each well and covered with a 24 x 50 mm cover-slip.

The slides were examined under a fluorescent microscope using a 50 x water objective and the results were recorded. The results were interpreted as follows:

Negative Reaction: No fluorescence was observed.

Positive Reaction: A bright fluorescence was observed.

Interpretation of test: A titre of ≥1:80 was considered positive.

A competition inhibition enzyme-linked immunosorbent assay (CI-ELISA), as described by Ndung'u *et al.* (1995) and De Waal *et al.* (1995) was employed to detect *A. marginale* antibodies. In this method, antigen was diluted with PBS at 5 µl of antigen into 11ml of PBS. The diluted antigen was used to coat labelled ELISA plates by adding 100 µl of diluted antigen to each well of the ELISA plate before incubation for 2 hours, at 37 °C. The ELISA plate was washed on the EL X50 Washer set to rinse 3 times/well using 750 ml PBS and 500µl Tween 20. The plates were dried by tapping them on a dry towel cloth. Blocking buffer A was prepared by dispensing 5g of Elite skim milk powder into 100ml of PBS to make 5% solution. Each well of the ELISA plate was blocked with 250µl Blocking buffer A and the plate was incubated at 37°C for 2 hours in a humid chamber set at low revolutions. The Blocking buffer was discarded and the plates were dried on a dry cloth towel. Test and control sera were removed from storage and allowed to thaw.

Dilutions of 1:100 were made for the test and the control sera with PBS on the ELISA plates following which the plates were incubated at 37 °C for 30 minutes in a humid chamber on an automatic shaker set at very low revolutions. Monoclonal antibody was prepared by dilution with 1% serum dilution buffer as follows: 3µl of Monoclonal antibody into 9 ml of serum dilution

buffer. The diluted Monoclonal antibody was added into the test and control sera after incubation and subsequently the plates were re-incubated at 37°C for 1 hour in a humid chamber set at low revolutions. After incubation the ELISA plate was rinsed once using washing buffer then placed into a plate shaker for 10 min. The contents of the plate were discarded and it was dried on a dry towel cloth. HRP-Goat Anti Mouse IgG (11µl) was then diluted in 11ml of the 1% serum dilution buffer and 100µl of the diluted HRP-Goat Anti-mouse antibodies were added into all the wells of the plate and incubated at 37°C for 30 minutes. After incubation, the ELISA plate was rinsed once with washing buffer and then placed on the Plate shaker for 10 minutes. After 10 minutes the contents of the Plate were discarded into a wash basin and the Plate was dried on a dry towel cloth. The substrate, 50µl ready to use TMB Single solution, was then added to all the wells of the Plate following which the reaction was read with an ELISA reader at 405 nm, after every 5 minutes for 30 minutes.

The ELISA reader was connected to a computer. After reading the optical density (OD) using the ELISA Multiskan reader, results were automatically transferred into ELISA Multiskan software programme in the computer. These results were then transferred to Microsoft Excel File and saved in an Excel spread sheet. Calculations of Percent negativity (PN), Percent Inhibition (PI), Coefficient of Variation (CV) and whether sample was positive or negative were automatically done in the Excel programme using formulae written and protected against deleting or editing in the computer.

Sero-prevalence of the three haemo-parasites was calculated according to the formula developed by Thrusfield (1995) as follows:

$$P = \frac{d}{n} \times 100;$$

where P represents the sero-prevalence of the haemo-parasite;

d represents the number of animals that tested positive for antibodies to the parasite; and

n represents the total number of animals sampled.

4.2.7 Statistical analyses

Frequencies were determined using PROC FREQ (SAS, 2003). Data for body weight, BCS and PCV was square root transformed to confer normality. The effect of rangeland type, breed, season, sex, age and their interactions on body weight and BCS was determined using PROC GLM for repeated measures (SAS, 2003). The chi-square test was used to determine the association of rangeland type, season, breed, sex, age and their interactions with TBD prevalence. PROC CORR (SAS, 2003) was used to determine the correlations among body weight, BCS, PCV and haemo-parasite infection. The effect of TBD infection on body weight, BCS and PCV was determined using PROC GLM for repeated measures (SAS, 2003).

4.3 Results

4.3.1 Body weights

Figure 4.1 shows the seasonal changes in body weights of Nguni and non-descript cattle in the sweet and sour rangelands. Rangeland type, season, breed, age, sex and the interaction of sex and age significantly affected the body weights of the cattle. Cattle in the sour rangeland (387.6 kg ± 6.67) had higher ($P < 0.05$) body weight than those in the sweet rangeland (339.9 kg ± 8.90) across seasons. Significantly higher ($P < 0.05$) weights were observed in the post-rainy season

(401.9 kg ± 11.58) and the lowest ($P < 0.05$) in the hot-dry season (324.7 kg ± 9.72). Males had higher ($P < 0.05$) body weights than females in all age-groups except in the 1 to 2 year olds which had similar ($P > 0.05$) body weights.

4.3.2 Body condition scores

The seasonal change in BCS of Nguni and non-descript cattle in the two rangeland types is shown in Figure 4.2. Rangeland type, season, breed, age, sex, and the interaction of, age and sex, and rangeland type, season and breed affected ($P < 0.05$) the BCS of the communal cattle. Nguni cattle in the sour rangeland had significantly higher BCS in all the seasons except the hot-dry season in which they were similar to those in the sweet rangeland. Males had higher ($P < 0.05$) BCS than females in all age-groups except in the 1 to 2 year old category where the BCS were similar ($P > 0.05$) between the sexes.

4.3.3 Packed cell volume

Figure 4.3 shows the seasonal changes in PCV in the communal cattle of the sweet and sour rangelands. Rangeland type, season, sex and the interactions between rangeland type and season, and rangeland type and breed significantly affected the PCV of the study animals. Cattle in the sweet rangeland had significantly higher PCV values in all the seasons than those in the sour rangeland except in the post rainy season in which the PCV values were similar ($P > 0.05$). Male cattle had higher PCV values than females in both rangeland types. Nguni cattle had higher ($P < 0.05$) PCV values than non-descript cattle in both rangeland types.

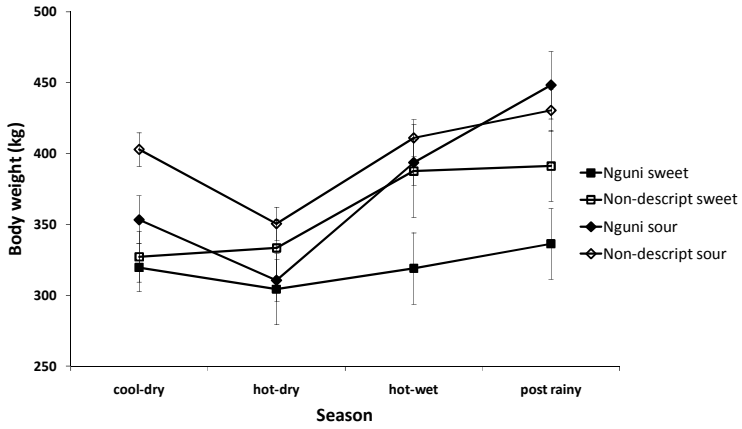

Figure 4.1: Seasonal change in body weight in Nguni and non-descript cattle in the sweet and sour rangelands

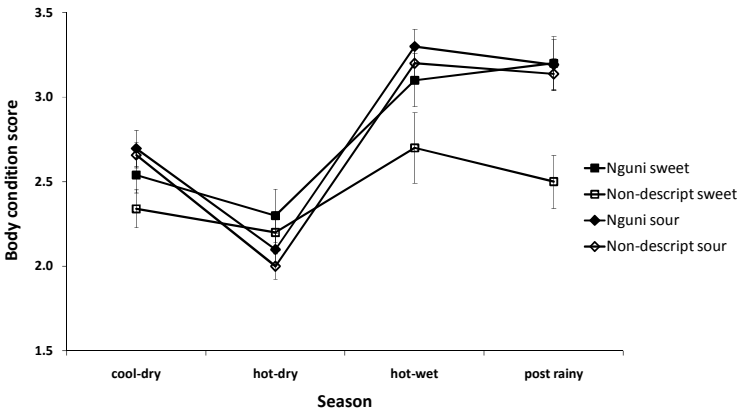

Figure 4.2: Seasonal change in body condition score of Nguni and non-descript cattle in the sweet and sour rangelands

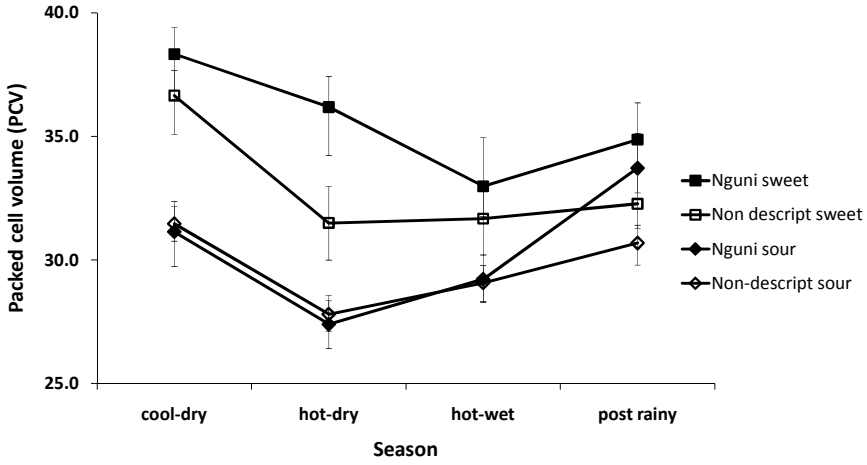

Figure 4.3: Seasonal change in packed cell volume in Nguni and non-descript cattle in the sweet and sour rangelands

4.3.4 Sero-prevalence of *Babesia bovis*

The IFAT revealed that 44.6 % of the cattle were positive for antibodies to *Babesia bovis*. There was a significant association ($P < 0.05$) between sero-prevalence of *B. bovis* and rangeland type, season, sex, the interaction of rangeland type and season and the interaction of season and breed. Cattle in the sweet rangeland had significantly lower sero-prevalence of *B. bovis* than those in the sour rangeland in all the seasons. Nguni cattle had lower ($P < 0.05$) sero-prevalence of *B. bovis* than non-descript cattle during the cool-dry and hot-wet seasons (Figure 4.4A). Females (49.1 %) had higher ($P < 0.05$) sero-prevalence of *B. bovis* than males (37.4 %). Breed and age were not significantly associated with the sero-prevalence of *B. bovis*.

4.3.5 Sero-prevalence of *Babesia bigemina*

Of the 144 cattle sampled, 45.9 % were positive for antibodies to *Babesia bigemina*. There was a significant association ($P < 0.05$) between the sero-prevalence of *B. bigemina* and season, age and the interaction of rangeland type and season. Cattle in the sweet rangeland had significantly lower sero-prevalence of *B. bigemina* than those in the sour rangeland in all seasons, except the post rainy season when they had similar ($P < 0.05$) sero-prevalence. The highest ($P < 0.05$) sero-prevalence of *B. bigemina* was observed in the cattle aged 1 to 2 years (81.5 %) while the lowest ($P < 0.05$) was observed in those greater than 5 years old (49.5 %). Rangeland type, breed and sex were not associated ($P > 0.05$) with the sero-prevalence of *B. bigemina*. The interaction of season and breed was not significantly associated with the sero-prevalence of *B. bigemina* (Figure 4.4B).

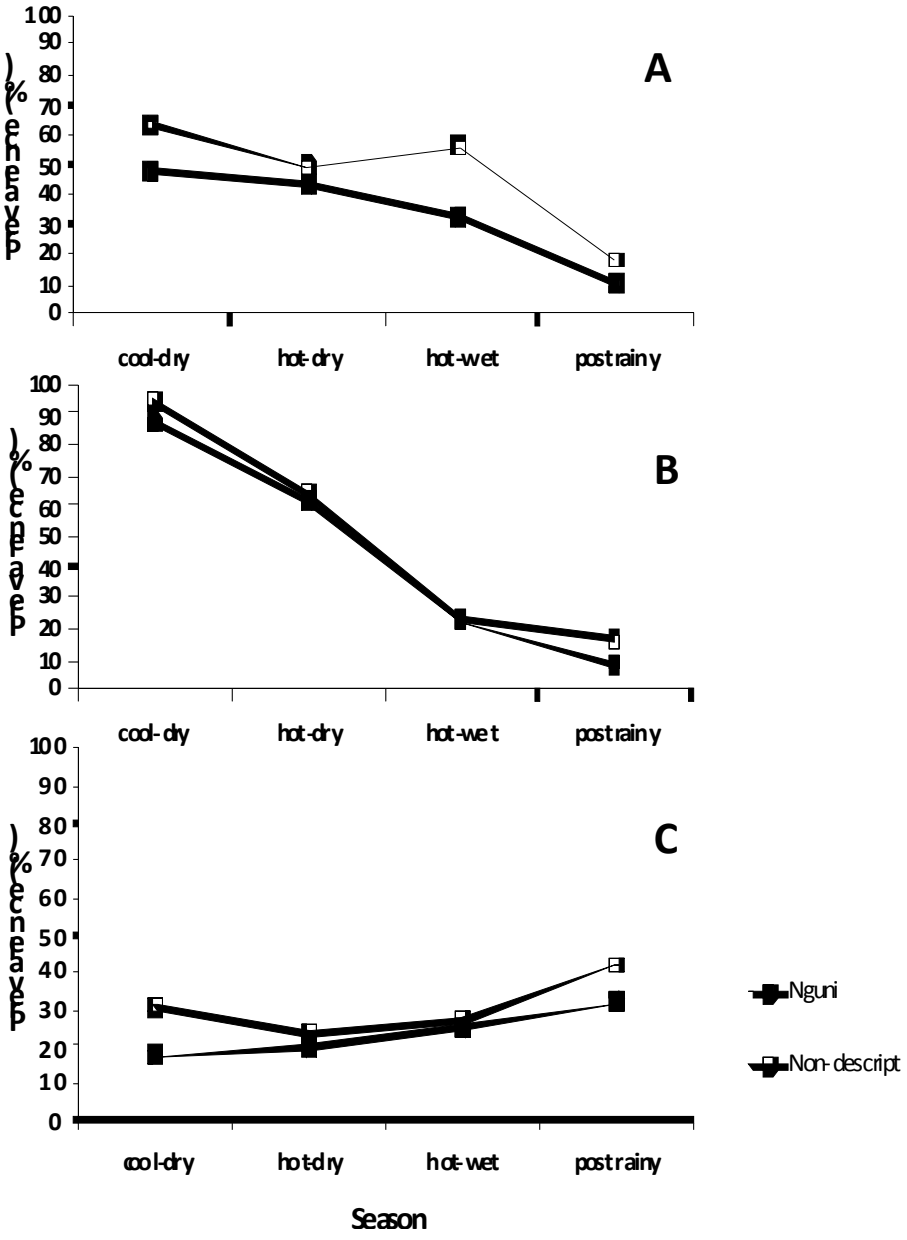

Figure 4.4: Seasonal change in sero-prevalence of *Babesia bovis* (A), *Babesia bigemina* (B) and *Anaplasma marginale* (C) in the communal cattle

4.3.6 Sero-prevalence of *Anaplasma marginale*

The CI-ELISA revealed that 25.6 % of the cattle were positive for antibodies to *Anaplasma marginale*. There was a significant association between the sero-prevalence of *A. marginale* and sex, age and the interactions of rangeland type and breed, and season and breed. Females had higher (30.4 %); sero-prevalence of *A. marginale* than males (18.1 %) ($P < 0.05$). The highest ($P < 0.05$) sero-prevalence of *A. marginale* was observed in the cattle aged 1 to 2 years (84.0 %) while the lowest ($P < 0.05$) was observed in those more than 5 years old (12.5 %). Nguni cattle in the sweet and sour rangeland had significantly lower prevalence of *A. marginale* than non-descript cattle in both rangeland types. Nguni cattle had lower ($P < 0.05$) sero-prevalence of *A. marginale* than non-descript cattle during the cool-dry season (Figure 4.4C). Rangeland type, season and breed were not significantly associated with the sero-prevalence of *A. marginale*.

4.3.7 Effect of tick-borne disease infection on body weight, body condition score and packed cell volume

Babesia bovis and *B. bigemina* significantly affected ($P < 0.05$) body weight in the communal cattle. The highest ($P < 0.05$) body weights were observed in cattle that tested negative for *B. bovis* and *B. bigemina* while the lowest ($P < 0.05$) occurred in cattle with high antibody titres for *B. bovis* and *B. bigemina*. Furthermore, *B. bovis* and *B. bigemina* significantly ($P < 0.05$) affected BCS in the communal cattle. Significantly higher ($P < 0.05$) BCS were observed in cattle which were not infected with *B. bovis* and *B. bigemina* while lower ($P < 0.05$) BCS occurred in cattle with low and high antibody titres of both haemo-parasites. The PCV values ranged from 26 to 35% in the sweet rangeland and 22 to 30% in the sour rangeland. *B. bovis* and *A. marginale* significantly affected PCV in the communal cattle. Lower ($P < 0.05$) PCV were observed in cattle which had high antibody titres of *B. bovis* and *A. marginale* while the highest ($P < 0.05$) occurred in cattle with high antibody titres of both haemo-parasites.

4.3.8 Correlations among body weight, condition score, packed cell volume and tick-borne disease infestation

Tables 4.1 and 4.2 show the correlations among body weight, BCS, PCV and TBD infestation in Nguni and non-descript cattle respectively. Infestations by *B. bovis* and *B. bigemina* had significant negative correlations with body weight and BCS. There were negative correlations ($P < 0.05$) between *B. bovis* and *A. marginale* infestations and PCV.

Table 4.1: Correlations among body weight, body condition score, packed cell volume, *B. bovis, B. bigemina* and *A. marginale* in Nguni cattle

	B. bovis	*B. bigemina*	*A. marginale*
Weight	-0.10	-0.34**	0.03
BCS	-0.12	-0.42**	-0.01
PCV	-0.24**	-0.02	-0.11*

* Indicates significance at $P < 0.05$.

** Indicates significance at $P < 0.001$.

Table 4.2: Correlations among body weight, body condition score, packed cell volume, *B. bovis, B. bigemina* and *A. marginale* in non-descript cattle

	B. bovis	*B. bigemina*	*A. marginale*
Weight	-0.10	-0.33**	0.04
BCS	-0.23**	-0.38**	-0.02
PCV	-0.36**	-0.02	-0.11*

* Indicates significance at $P < 0.05$.

** Indicates significance at $P < 0.001$.

4.4 Discussion

Cattle on the sweet rangeland were observed to have lower weight losses, and higher BCS than those on the sour rangeland. This was attributed to the fact that forages in the sweet rangeland retain palatability and high nutritive value to maintain animal weight and condition throughout the dry season (Sibanda, 1999). Non-descript cattle had the highest body weights due to their large frame sizes. Although the Nguni cattle had lower body weights, they managed to maintain higher BCS than their non-descript counterparts. The Nguni cattle also had higher PCV values throughout the year evidence that the Nguni breed may be more adapted to the local production conditions. That Nguni cattle maintain higher BCS and PCV values agrees with Ndlovu (2007) who observed Nguni steers to have higher BCS and PCV values than Bonsmara and Angus steers.

Cattle reared on the communal sweet and sour rangelands in the study areas were sero-positive for three tick-borne haemo-parasites, *B. bovis*, *B. bigemina* and *A. marginale*. This agrees with the species of vector ticks, *R.* (*Boophilus*) species and *R. evertsi evertsi*, which were observed to occur in the study sites (Chapter 3). All the cattle were sero-negative to *E. ruminantium*, probably because the tick vector, *Amblyomma habraeum*, was not present in the study sites (Chapter 3).

The sero-prevalence of each of the three TBDs observed in the study was low (< 50 %), suggesting an endemically unstable situation. Endemic stability is more likely to exist where the prevalence of serum antibodies to infection is high (70%), while the antibody prevalence is usually low (30%) in the endemically unstable state (Perry and Young, 1995; Peter *et al.*, 1997).

However, the inherent resistance of cattle to ticks and TBDs also influences these thresholds (Moll *et al.* 1986; Swai *et al.*, 2007).

The lower sero-prevalence of *B. bovis* and *A. marginale* in Nguni than non-descript cattle in the cool-dry and hot wet seasons suggest a high degree of tolerance to these haemo-parasites in the Nguni cattle, especially in these seasons. The Nguni managed to maintain higher BCS than non-descript cattle during the cool dry season, an indication of a superior nutritional status (Ndlovu, 2007), which most likely contributed to their ability to resist TBD infections. Tolerance to ticks may contribute to modulate the transmission rates of TBDs to a sub-pathological level (Mattioloi *et al.*, 2000). Indigenous Nguni cattle have been shown to carry lower tick loads during the hot-wet season (Chapter 3) than non-descript cattle and this may have also resulted in the lower prevalence of TBDs in Nguni cattle in this season.

Cattle in the sweet rangeland had lower sero-prevalence of *B. bovis* and *B. bigemina*, which could be attributed to the lower tick infestations that occur in the sweet rangeland (Chapter 3). The sweet rangeland receives lower rainfall and has sparse vegetation cover compared to the sour rangeland (Ellery *et al.*, 1995). This may have resulted in the reduction of the tick vector *Rhipicephalus* (*Boophilus*) species, on the rangeland and thus lower transmission rate of *B. bovis* and *B. bigemina*.

A decreased trend of sero-positivity of *B. bigemina* and *A. marginale* associated with age was evident in this study. This is in agreement with Regassa *et al.* (2003) who observed a similar trend on a South African ranch where non-intensive tick control was applied. The greatest TBD

infection rate in cattle occurs at 6 to 20 months of age, and is uncommon in animals more than five years old, but the severity of the disease increases with age (Radostits *et al.*, 1995). This may explain the age-related decreased trend of sero-positivity observed in the study.

One of the most important indicators of TBD infection is anaemia (Mbati *et al.*, 2002) which can be accurately and practically evaluated by determining PCV (Jain, 1993). The negative correlations between PCV and infection by *B. bovis* and *A. marginale* could be explained by the fact that the two tick-borne haemo-parasites replicate inside erythrocytes leading to increased haemolysis and anaemia (Riond *et al.*, 2007). Although anaemia can be caused by factors other than TBDs, animals with elevated antibody responses may have possibly been more severely infected and thus had lower PCV values than sero-negative animals.

Infection with TBDs causes anorexia and, consequently, loss of body weight and condition (Kahn *et al.*, 2006). Two cases of anaplasmosis were observed and treated during the cool-dry season on the sour rangeland. In order to reduce production losses caused by high TBD infection during the cool-dry season, communal farmers should vaccinate their animals, especially adult stock, at the end of the post rainy season to ensure adequate protection throughout the cool-dry season. The poor body condition scores that were observed during the dry seasons require the use of feed supplements, such as the widely available *Acacia karoo* leaf meal, to maintain a high nutritional status in cattle throughout the year. Improved nutrition results in improved immunity to TBDs (Bock *et al.*, 2006) and may cause a decrease in the clinical cases of TBD during the dry seasons. The rearing of the Nguni breed on communal rangelands should be encouraged as

this breed maintains body condition and resists TBD infections better than the non-descript breed.

4.5 Conclusions

The prevalence of babesiosis and anaplasmosis in Nguni and non-descript cattle kept on communal rangelands was low. Cattle on the communal sweet and sour rangelands were sero-negative to heartwater. The Nguni breed maintained better BCS and was more resistant to *B. bovis* and *A. marginale* than non-descript cattle in the cool-dry and hot wet seasons. The prevalence of babesiosis was lower in the sweet rangeland probably because of reduced presence of the tick vector. The sero-prevalence of *A. marginale* and *B. bigemina* showed an age-related trend with the 1 to 2 year olds having the highest prevalence and the 5 year olds the lowest. Use of the adapted and TBD-resistant Nguni breed on communal rangelands is recommended.

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Chapter 5: General discussion, conclusion and recommendations

5.1 General discussion

Earlier research reported that ticks and tick-borne diseases (TBDs) are the major constraints to communal cattle production. For sustainable and profitable cattle production in the communal areas, it is crucial to identify tick and TBD-resistant cattle breeds that perform well on communal rangelands. In this study tick prevalence and loads, and sero-prevalence of tick-borne diseases were compared in Nguni and non-descript cattle on the communal sweet and sour rangeland of the Eastern Cape.

Seasonal tick loads and prevalence were determined in Chapter 3. The Nguni breed had lower tick loads of *Rhipicephalus appendiculatus* in the hot-wet and post-rainy season and *Hyalomma* species in all seasons than the non-descript cattle. This indicates a higher innate and/or acquired resistance of this breed to the parasites. Tick species composition and loads varied between the two rangeland types. *Hyalomma* species occurred only on the sour rangeland and cattle on the sweet rangeland had lower loads for all the tick species identified. Differences in the vegetation composition and cover, humidity and annual rainfall between the sweet and sour rangeland most likely influenced the distribution of ticks as observed in the study. In Chapter 3, tick prevalence and loads were observed to vary seasonally, being high in the hot-wet season when environmental temperatures and humidity are high, providing conducive conditions for tick proliferation and survival. Younger cattle carried lower tick loads, and this was most likely due to their protection by some form of age-related innate defence.

The low sero-prevalence of TBDs observed in the study suggests an endemically unstable situation. It is however, thought that the relatively high resistance of cattle to ticks and TBD may have influenced the observed sero-prevalence values. The Nguni breed had a higher degree of resistance to *B. bovis* and *A. marginale* in the hot wet season. Indigenous Nguni cattle were shown to carry lower tick loads during the hot-wet season (Chapter 3) than non-descript cattle and this may have resulted in the lower prevalence of TBDs in Nguni cattle in this season. Nguni cattle managed to maintain higher body condition scores than non-descript cattle throughout the period of study. This indicates a superior nutritional status, which could have also contributed to their improved ability to resist TBD infections. The lower sero-prevalence of *B. bovis* and *B. bigemina* in the sweet rangeland was probably related to low infestation rates due to the low tick loads in the sweet rangeland. The highest sero-prevalence of *A. marginale* and *B. bigemina* were observed in the cattle aged 1 to 2 years while the lowest was observed in those greater than five years old. This could be explained by the fact that in cattle, TBD infection rate is greatest at 6 to 20 months of age and uncommon in animals more than 5 years old.

5.2 Conclusions

Rhipicephalus appendiculatus, *Rhipicephalus* (*Boophilus*) *decoloratus*, *Rhipicephalus evertsi evertsi* were the most prevalent tick species on the communal rangelands of the Eastern Cape. *Hyalomma* species was only found in the sour rangeland. The Nguni breed had lower tick loads of *R. appendiculatus* in the hot-wet and post-rainy season and *Hyalomma* species than the non-descript cattle. Cattle on the sweet rangeland had lower loads of ticks than those on the sour rangeland. The prevalence of babesiosis and anaplasmosis in communal cattle on the sweet and sour rangelands was low. Cattle on the communal sweet and sour rangelands were sero-negative

to heartwater. The Nguni breed maintained better BCS and was more resistant to *B. bovis* and *A. marginale* than non-descript cattle in the hot-wet season. Cattle in the sweet rangeland had lower sero-prevalence of babesiosis than those in the sour rangeland. The current study's findings suggest greater adaptation and higher resistance to ticks and TBDs in the Nguni breed.

5.3 Recommendations

In the current study, Nguni cattle were shown to be more resistant to ticks and TBDs than non-descript cattle on communal sweet and sour rangelands. For sustainable and profitable cattle production on communal rangelands, it is recommended that communal farmers rear Nguni cattle which do not require regular treatment with costly veterinary medicines for tick and TBD control.

Strategic tick control methods such as good grazing management practices are recommended during the hot-wet season. Such strategies avert major losses caused by high tick loads, especially in the sour rangeland. Communal farmers should vaccinate their animals, especially adult non-descript cattle, at the end of the post rainy season to ensure adequate protection throughout the cool-dry season. This would help to reduce production losses caused by high TBD infection during the cool-dry season.

The poor body condition scores that were observed during the dry seasons require the use of feed supplements, such as the widely available *Acacia karoo* leaf meal, to maintain a high nutritional status in cattle throughout the year. Improved nutrition results in improved immunity to TBDs and may cause a decrease in the clinical cases of TBD during the dry seasons.

Further research, is, however required on the following aspects:

1. Further quantification of the losses due to ticks and TBD in low input cattle production systems.
2. Investigation of the immunological mechanisms of tick and TBD tolerance in Nguni cattle.
3. Heritability of ticks and TBD tolerance of Nguni cattle.