CHARACTERIZATION OF TANZANIA SHORTHORN ZEBU CATTLE FOR TOLERANCE TO TICKS AND EAST COAST FEVER

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A THESIS SUBMITTED IN FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY OF SOKOINE UNIVERSITY OF AGRICULTURE. MOROGORO, TANZANIA.

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EXTENDED ABSTRACT

In Tanzania, livestock diseases, especially tick bone diseases (TBDs), impose loses to livestock in terms of mortality, decrease in production and increased costs of control measures. Among the TBDs, East Coast fever (ECF) is the most prevalent and fatal disease. The main control measure for TBDs is use of acaricides to control the vector ticks, chemotherapy of sick animals as well as immunization of cattle by the infection and treatment method (ITM). The applications of these are control measures are often limited by high costs of acaricides and veterinary drugs, development of resistance by vector ticks and the parasites as well as negative environmental impacts. This necessitated the search for alternative ways to control TBDs with minimum use of acaricides. Indigenous breeds of cattle are said to be tolerant to TBDs and adapted to harsh conditions prevailing in rural areas. Among the Tanzania Shorthorn Zebu (TSHZ) cattle, Tarime cattle are believed by livestock keepers to be tolerant to ticks and ECF. However, there are no scientific investigations which have been conducted to verify the farmers’ beliefs. Therefore, studies were conducted to validate the farmers’ belief that Tarime cattle are tolerant to ECF.

In the first study, a cross-sectional survey was conducted in four selected districts of Mara (Serengeti and Tarime) and Simiyu (Maswa and Meatu) regions to determine the perception of livestock farmers on ticks, tick-borne diseases and tolerance of their cattle to TBDs. In the second study, tick burden and prevalence of Theileria parva (T. parva) infection in Tarime cattle found in Serengeti and Tarime districts were assessed. In the third study, an on-station experiment was conducted to compare tick burden and induced immunity due to T. parva infection in Tarime and Sukuma zebu cattle under different tick control regimes. The fourth study was a review on tolerance of TSHZ cattle to ticks and ECF. In the first study, a well structured questionnaire and personal observation were
used to collect information. The aim of the study was to assess farmers’ knowledge and perceptions on tick species, tick-borne diseases, tick control measures, production constraints and tolerance of Tarime and Sukuma zebu cattle to TBDs. The study involved 60 agro-pastoralists from each of the districts of Serengeti, Tarime, Maswa and Meatu in Tanzania, making a total sample size of 240 livestock farmers. Data were collected through individual interview using a structured questionnaire. The results show that, livestock diseases were ranked as the first important problem affecting cattle production in Serengeti and Tarime districts while in Maswa and Meatu districts lack of livestock feeds during the dry season was ranked as the most important constraint, followed by livestock diseases. Among the diseases affecting cattle, the TBDs ranked first in Serengeti and Tarime districts while in Maswa and Meatu districts the TBDs ranked fourth and sixth, respectively. Most (74%) of the livestock farmers interviewed knew well the signs of the different TBDs. The majority of the respondents in all districts knew that ECF is caused by ticks, but did not associate the other TBDs with ticks. Most of the farmers interviewed were using acaricide to control ticks, and the most common method of application was hand spraying. All farmers used Oxytetracycline to treat TBDs, however, some farmers used local herbs. Although most farmers knew the signs of TBDs, they were not spraying/dipping their animals on regular basis due to economic reasons and the belief that their animals always carry ticks without being sick or dying and ECF affects only calves. About half of the farmers considered their breeds to be tolerant to ticks and ECF. It is concluded that, the livestock farmers in the Lake zone have substantial knowledge on tick species and TBDs symptoms and they consider ECF as less important disease compared to the other TBDs.

The second study was carried out to assess the distribution, abundance of different tick species and the prevalence of *T. parva* infection in Tarime zebu cattle kept in selected
wards of Serengeti and Tarime districts in Mara region. Adult ticks were sampled, identified and counted from 360 animals which were extensively managed in communal rangelands. Concurrently, blood samples were collected and thereafter DNA extracted and nested polymerase chain reaction (nPCR) carried out to determine the prevalence of *T. parva*. Two primers specific for p104 gene were used in PCR amplification to detect the presence of *T. parva* DNA. Four genera of ticks, namely, *Rhipicephalus, Amblyomma, Boophilus* and *Hyalomma* were identified. *Rhipicephalus spp* accounted for 71.8% of the total ticks whereas *Amblyomma, Boophilus* and *Hyalomma* constituted 14.1%, 14.0% and 0.1% of the ticks, respectively. There was significantly (p < 0.05) more animals infested with ticks in Tarime district (96.1%) than in Serengeti (61.7%). The average counts of ticks were higher in adult animals (p < 0.05) than in young animals. The overall prevalence of *T. parva* was 27.7% and was significantly (p < 0.05) higher in Serengeti (38.3%) than in Tarime district (16.7%). Interestingly, it was observed that, all animals which tested positive for *T. parva* did not show any clinical signs of ECF, suggesting existence of subclinical infection in Tarime zebu. These results suggest that Tarime cattle can tolerate ECF infection and are likely to save as potential carriers of *T. parva* to other less tolerant cattle breeds in mixed herds. Since Tarime cattle are preferred by most farmers with mixed herds, routine screening for *T. parva* is highly recommended to minimize introduction of infected cattle into an immunologically naive population.

The third study was conducted to determine tick burden and immunological parameters of resistance to ECF in Tarime and Sukuma zebu strains exposed to natural tick infestation. Tick load, Packed Cell Volume (PCV), *T. parva* specific antibody percent positivity (PP) and prevalence of *T. parva* parasites were studied in relation to dipping regime, breed, and season of the year. A total of 25 animals per strain were used in this experiment. Animals of each strain were divided into three groups. Animals in group I were dipped
every two weeks while those in group II were dipped every three weeks. Animals in group III were not dipped at all. Comparison of effect of dipping frequency on tick burden showed no significant (p > 0.05) differences when cattle which were dipped in each strain either once every 2 or 3 weeks in the two seasons. However, Tarime cattle had significantly higher (p < 0.05) tick count than Sukuma cattle and non-dipped groups maintained high tick infestation throughout the experimental period. The PCV values in all cattle were within the normal physiological range; although this parameter was significantly lower (p < 0.05) in Tarime cattle. All cattle regardless of breed were sero-positive (had antibody levels above ELISA cut-off point), but Tarime cattle maintained higher antibody percent positivity compared to Sukuma cattle by 15%. Conversely, the prevalence of *T. parva* parasites was lower in Tarime (36%) compared to Sukuma cattle (38%), although the difference was not significant. During the study period 20% (5/25) of Sukuma cattle contracted ECF, but none of the Tarime cattle showed any clinical signs for the disease. The differences between the two zebu strains shown in terms of antibody percent positivity and *T. parva* parasite prevalence indicate different ability of the two cattle strains to resist tick infestation and ECF infection under natural challenge. Thus, the higher antibody levels but lower parasite prevalence under high tick challenge observed in Tarime cattle, suggests a high proportion of individual animals in this strain, which are carriers of *T. parva*. Thus, these findings confirm the farmers’ perceptions on suitability of Tarime cattle under prevailing ecological conditions in the Lake zone of Tanzania.

The fourth study was a review on the prevalence and tolerance of Tanzania Shorthorn Zebu cattle to East Coast fever in Tanzania. A search of peer-reviewed publications on ECF, ticks and ECF tolerant cattle in Tanzania was conducted from comprehensive databases including PubMed, Science Direct, Swetswise and CAB direct. The search was extended to available theses, conference proceedings and project reports. The findings
show that ECF is not seen by most livestock keepers as the most serious disease compared to other TBDs. This is because the zebu cattle which are predominantly kept all over the country are thought to be tolerant to ticks and ECF. Some of the zebu strains which are thought to tolerate ECF are Sukuma, Tarime, Maasai and Fipa cattle. In all agro ecological zones the most prevalent tick species is *R. appendiculatus*, this implies that the TSHZ cattle are at risk of being infected with ECF if the animals are not resistant and proper tick control regimes are not observed. In Tanzania, ECF control has mainly relied predominantly on tick control using acaricides and chemotherapy while ECF immunization has been recently introduced. Some research results also showed that, immunization can boost immune status of indigenous cattle and, if ECF immunization can be sustained, large areas are expected to be endemically stable. However, the adoption of immunization on wider scale is questionable due to its cost and change in farmers’ attitudes, socio-demographic and environmental concerns. The study recommends interdisciplinary collaboration between veterinary epidemiologists, socio and agricultural economists and ecologists not only to find a single most suitable method of ticks and ECF control but to integrate the available methods with tolerant strains of cattle available to create endemic stability condition for the disease which can reduce the complexity of the whole process.

Generally, the findings of this study confirm farmers’ perception on the ability of Tarime cattle to tolerate ECF infection under prevailing ecological conditions in the Lake zone of Tanzania. The use of this strain, therefore, can be an alternative way of minimizing acaricide usage, whereby application can be done on monthly basis. However, when Tarime and other breeds are kept together, screening of the animals for presence of *T. parva* parasites is recommended. From these findings it is concluded that using Tarime cattle could be effective and economical in rural areas as it minimizes the use of synthetic
acaricide to control ticks and TBDs. Further research studies to determine the gene responsible for tolerance of Tarime cattle to ECF are recommended.
DECLARATION

I, EMMANUEL LEVILLAL KATAMBOI LAISSER, do hereby declare to the Senate of Sokoine University of Agriculture that this thesis is my own original work done within the period of registration and that it has neither been submitted nor being concurrently submitted for degree award in any other institution.

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(PhD Candidate)

The above declaration is confirmed;

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Prof. CHENYAMBUGA, S. W.  Date

(Supervisor)

______________________________  _______________
Prof. KARIMURIBO, E. D.  Date

(Supervisor)
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DEDICATION

This thesis is dedicated to my beloved wife; Martha Tesha, my children Loitarasakaki and William Melita for their tolerance of my long absence from the family for the entire study period, my lovely sister Lidya who handled extended family responsibilities during my absence and to my deceased parents Mr. Katamboi Ngoilenya Laisser and Mrs. Naisiligaki Lotubukokoki Lekashu who laid my educational foundation.
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<th>Description</th>
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<tr>
<td>°C</td>
<td>Degree Celsius</td>
</tr>
<tr>
<td>ANOVA</td>
<td>Analysis of Variance</td>
</tr>
<tr>
<td>BQ</td>
<td>Blackquarter</td>
</tr>
<tr>
<td>CBPP</td>
<td>Contagious Bovine Pleuropneumonia</td>
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<tr>
<td>DAARS</td>
<td>Department of Animal, Aquaculture and Range Science</td>
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<tr>
<td>DNA</td>
<td>Deoxyribonucleic Acid</td>
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<tr>
<td>ECF</td>
<td>East Coast fever</td>
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<tr>
<td>EDTA</td>
<td>Ethylenediaminetetraacetic Acid</td>
</tr>
<tr>
<td>ELISA</td>
<td>Enzyme-linked Immunosorbent Assay</td>
</tr>
<tr>
<td>FAO</td>
<td>Food and Agriculture Organization of the United Nations</td>
</tr>
<tr>
<td>FMD</td>
<td>Foot and Mouth Diseases</td>
</tr>
<tr>
<td>GLM</td>
<td>General Linear Model</td>
</tr>
<tr>
<td>ITM</td>
<td>Infection and treatment method</td>
</tr>
<tr>
<td>LSD</td>
<td>Lumpy skin diseases</td>
</tr>
<tr>
<td>MLFD</td>
<td>Ministry of Livestock and Fisheries Development</td>
</tr>
<tr>
<td>MOEV</td>
<td>Ministry of Education and Vocational Training</td>
</tr>
<tr>
<td>N or n</td>
<td>Number of observation</td>
</tr>
<tr>
<td>nPCR</td>
<td>Nested Polymerized Chain Reaction</td>
</tr>
<tr>
<td>NS</td>
<td>Not significant</td>
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<tr>
<td>OTC</td>
<td>Oxytetracycline</td>
</tr>
<tr>
<td>P-value</td>
<td>Probability value</td>
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<tr>
<td>PCR</td>
<td>Polymerized Chain Reaction</td>
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<tr>
<td>PCV</td>
<td>Packed cell volume</td>
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<tr>
<td>Acronym</td>
<td>Description</td>
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<td>-------------</td>
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<tr>
<td>RBC</td>
<td>Red blood cells</td>
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<tr>
<td>s.e</td>
<td>Standard error</td>
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<td>SAS</td>
<td>Statistical Analysis Systems</td>
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<td>SUA</td>
<td>Sokoine University of Agriculture</td>
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<td>TBDs</td>
<td>Tick Borne-Diseases</td>
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<tr>
<td>TVLA</td>
<td>Tanzania Veterinary Laboratory Agency</td>
</tr>
<tr>
<td>URT</td>
<td>United Republic of Tanzania</td>
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<tr>
<td>UV</td>
<td>Ultra Violet</td>
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CHAPTER ONE

1.0 INTRODUCTION

1.1 Background Information

Animal diseases are one of the main challenges which undermine productivity of indigenous cattle. The most critical diseases are Tick-borne diseases (TBDs), particularly East Coast fever (ECF). In Tanzania, TBDs pose a more serious, fatal and economic threat to cattle production compared to other diseases (Chenyambuga et al., 2008). These TBDs include anaplasmosis, babesiosis, ECF and heartwater. Among these diseases, ECF or theileriosis is reported to be the most prevalent and fatal disease among TBDs in East Africa, particularly in Tanzania (Okuthe and Buyu, 2006; Swai et al., 2007).

In Tanzania about 80% of the national cattle herds are at risk of being infected with TBDs annually and the direct economic losses are estimated at US$ 248 million and mortality rate of 920,000 animals per year (Kivaria, 2006). The Government of Tanzania has decided to subsidize the price of acaricides to enable many farmers to dip their animals so as to reduce mortality rates due to TBDs. However, many livestock keepers are unable to purchase acaricides and, therefore, continue to lose animals from TBDs (Mugisha et al., 2005). Due to this fact, there is a need to look for other means of controlling the disease rather than relying on application of acaricides which seems to be unsustainable.

Keeping genetically resistant breeds is one of the sustainable and cheap means of controlling diseases (de Castro, 1991). In other countries breeds of cattle which are resistant to ticks infestation and TBDs have been developed (Latif et al., 1991). Thus, it is possible to have a breed of cattle in Tanzania which is tolerant to TBDs like those found in other African countries. Some farmers around Lake Victoria do not spray or dip their
cattle against ticks, the vectors of *Theileria parva*, the parasite which causes ECF, due to their perception that their cattle are resistant (Chenyambuga *et al.*, 2008). Tarime cattle strain which is found in the Lake Zone is believed to be tolerant to TBDs by their owners. This belief has led to the spread of Tarime cattle to many parts of the country, particularly in Rukwa and Mbeya regions as a coping strategy to TBDs. Therefore, there is a need to conduct a scientific investigation to ascertain the farmers’ belief and determine the mechanism responsible for tolerance of Tarime zebu strain to TBDs.

1.2 Problem Statement and Justification

Tolerance of Tarime zebu strain to ECF has not been scientifically proved, but it is based merely on farmers’ perceptions. In addition, there is no documented information which indicates that Tarime cattle do not succumb to clinical disease when infected with *T. parva*. Thus, tolerance of Tarime zebu strain to TBDs is based only on perceptions and attitudes of livestock farmers. Thus, there is need for conducting scientific investigations to verify the tolerance of Tarime zebu strain to ticks and ECF. This study therefore, intended to assess the ability of Tarime cattle to tolerate tick infestation and ECF infection. It was expected that the findings from this study will assist policy makers, farmers and breeders to make rational decision for genetic improvement and conservation of Tarime zebu strain.

1.3 Objectives

1.3.1 Overall objective

To establish whether Tarime cattle are tolerant to tick infestation and ECF infection.
1.3.2 Specific objectives

The specific objectives were:

i. To determine the perceptions of livestock farmers on tolerance of Tarime cattle to ticks and ECF.

ii. To determine tick burdens and prevalence of ECF in Tarime cattle kept in Tarime and Serengeti districts.

iii. To compare the tolerance/susceptibility of Tarime and Sukuma zebu cattle to ECF and tick infestation.

iv. To review tolerance and susceptibility of TSHZ cattle to ticks and *Theileria parva* infection.

1.4 Research Hypothesis of the Study

i. Livestock farmers in the Lake zone of Tanzania perceive that their cattle are tolerant to ticks and ECF and thus, there is no need for dipping to control ticks.

ii. There is no difference in tick burden and prevalence of ECF in Tarime cattle kept in Tarime and Serengeti districts.

iii. There is no difference in tolerance/susceptibility between Tarime and Sukuma zebu cattle to ECF and tick infestation.

1.5 General Methodology

Study Area

This study was conducted in four districts located in two regions of Tanzania and on station at Misungwi district in Mwanza region as shown in the map in Figure 1. The districts include Serengeti and Tarime in Mara region, Maswa and Meatu in Simiyu region. The regions are home to two common strains of Tanzania Shorthorn Zebu (Plates 1 and 2) populations used in this study, namely Sukuma and Tarime cattle. Both, Mara
and Simiyu regions are located in the Lake zone of Tanzania. These regions are geographically separated and have different agro-climatic conditions. Mara region lies between latitudes 1° 0’ and 2° 31’ south of Equator and between longitudes 33° 10’ and 35° 15’ east of Greenwich while Simiyu region lies between latitude 2°1’ and 4° south of Equator and between longitudes 33°3’ and 35°1’ east of Greenwich. Tarime Zebu cattle (Plate 1) are kept in Serengeti and Tarime districts while Sukuma cattle (Plate 2) are kept in Maswa and Meatu districts. Both Tarime and Sukuma cattle belong to the Tanzania Shorthorn Zebu strains. The dominant farming system in all districts is agro-pastoral production system in which livestock farmers practice herded grazing in communal and fallow lands.

Figure 1: Map of Tanzania showing the study areas

To achieve specific objective 1, a cross-sectional survey was carried out in four districts; two in Mara region and two districts in Simiyu region (Figure 1). The districts in Mara region were Tarime and Serengeti while in Simiyu the districts were Maswa and Meatu.
The districts in Mara region have higher number of Tarime cattle while those in Simiyu have Sukuma cattle. In each of the four districts, three wards and two villages per ward were selected, forming a total of 12 wards and 24 villages. Within a village the list of households keeping Tarime or Sukuma zebu cattle were used as a sampling frame from which respondents were picked randomly using a table of random numbers. Ten households per village were sampled, giving a sample size of 60 households in each district (240 respondents in total). The heads of the households were the main respondents. However, other members of the household, whenever necessary, had an opportunity to provide supplementary information. Information collected were: household socio-economic characteristics, livestock production constraints, most important disease in the area, cattle mortality rate due to TBDs in the area, ability of cattle to tolerate ticks and ECF, tick species and their control. This information was collected using a structured questionnaire.

Plate 1: A and B Mature Tarime cattle strain
The information collected were coded and recorded into the spreadsheets and the data were analyzed using the Statistical Package for Social Sciences (SPSS), statistical software Release 16.0 (SPSS, 2008). The following descriptive statistics were generated: means, standard deviations, frequencies and percentages. The percentage of farmers’ response for a particular variable such as age and education of the farmers, experience in keeping livestock, important diseases, common tick species, tick control method used, frequency of dipping/spraying, tolerance of animals to ticks and ECF were compared among the districts using a chi–square test to test whether the proportions in the four districts were different from each other. The differences among the districts were considered significant at $P \leq 0.05$. One paper (Paper I) was generated from this study. This paper focused on the perception of livestock farmer’s regarding tolerance of their animals to ticks and TBDs. The results from this study gave comparative and general information on the tolerance of the two strains kept by these communities on ticks and TBDs.

To achieve specific objective 2, a study was designed and carried out in the two districts of Mara region, namely Tarime and Serengeti districts. In each district three wards were
selected making a total of six wards. Two villages were purposively selected (based on having large number of Tarime cattle) from each ward making a total of 12 villages for both districts (Figure 1). In each village, 10 households (120 households in total) were randomly selected using random numbers and a total of 360 animals (three per household i.e. one calf, one weaner and one adult animal) were selected for ticks identification and counting as well as blood sampling for *T. parva* detection. The desired sample size for the study was calculated according to the procedure described by Thrusfield (1995).

Tick counts were done according to Londt *et al.* (1979), counting ticks at predilection sites. The predilection sites were head, sternum and hind quarters (Baker and Ducasse, 1967). Visible adult ticks were counted from one side of the animal body and identified using the keys of Mathysse and Colbo (1987). The half body counts were doubled to estimate the number of ticks for the whole body. Animal category, tick type, tick attachment sites, household head, village, ward and district were recorded.

Whole blood samples were collected by jugular vein puncture using vacutainer tubes with EDTA. The samples were labeled and stored in a cool box with ice packs while in the field and later put into a refrigerator until when they were transferred to the laboratory at the Department of Microbiology and Parasitology at SUA for further analyses. Genomic DNA was extracted from 354 whole blood samples using the Pure Gene Blood Core (QIAGEN) Kit (Minnesota, USA) according to the manufacturer’s instructions (six samples clotted before DNA extraction and they were omitted). *Theileria parva* genomic DNA was detected using a nested polymerase chain reaction (nPCR). Primers used in the amplification were for a *T. parva*-specific 104-kDa antigen (p104) gene obtained from GenBank (Accession No. M29954) using procedure described by Odongo *et al.* (2010).
The nPCR products were separated on 1.5% agarose gel and visualized in ultra violet (UV) trans-illuminator (Plate 3).

![Plate 3: A representation of amplicons obtained from amplification of the *T. parva* p104 gene using nested PCR. An agarose gel of 1.5% was used for analysis of PCR products. M is a 1kb Marker (From fermentas – South Africa). 1 - 11 and 14 – 24 are tested samples while 12 and 25 are Negative controls, 13 and 26 are Positive control](image)

The data obtained from tick count and PCR were coded and analysed using SPSS version 16 (SPSS, 2008). The percentage of animals infested with different tick species and the prevalence of *T. parva* in animals were compared using a chi-square to test the significance of the differences in tick infestation and prevalence of *T. parva* between villages, wards and districts. ANOVA was used to assess the statistical significance differences in tick counts between wards and between districts. The fixed effects assessed were district, ward within district, animal category and attachment site. All results were considered significant at *p* ≤ 0.05.

Paper II was generated from this study. This paper looked on the tick burden and prevalence of *T. parva* in Tarime zebu cattle kept in Serengeti and Tarime districts (Figure 1).
The results from this study gave comparative information on tick burden and *T. parva* prevalence in this breed when managed in two different geographical locations with different tick management practices.

To achieve specific objective 3, an on-station experiment was conducted at TALIRI-Mabuki Research Center (Figure 1) to observe how Tarime and Sukuma cattle behave on natural condition for tick infestation and *T. parva* infection. *T. parva* antibodies were determined under different tick control regimes and seasons. A total of fifty cattle (25 Tarime and 25 Sukuma) were randomly selected for this study. Animals of each breed were divided into three groups, comprising eight, eight and nine cattle. Animals in groups 1 and 2 were dipped once after two and three weeks, respectively, while those in group three (control group) were not dipped throughout the study period. Data on tick count were taken on weekly basis while blood sample for PCV, *T. parva* parasite and *T. parva* antibody detection were taken at an interval of three weeks. This experiment covered both dry and rainy seasons. Data were analyzed and the results are presented in paper three of this Thesis.

To achieve specific objective 4, a search of peer-reviewed publications on ECF, ticks and ECF tolerant cattle in Tanzania (Plates 1, 2, 4, 5, 6 and 7) was conducted from comprehensive databases, including PubMed, Science Direct, Swetswise, and CAB direct. The search was extended to available theses, conference proceedings and project reports. Keywords were standardized across the databases to produce comparable searches and these were: East Coast fever, *Theileria parva*, *Rhipicephalus appendiculatus*, epidemiology, prevalence, incidence, cattle, tolerance and Tanzania. References of all relevant articles were also searched to identify articles that could have been missed in the search. The search was conducted for all available years in each
database. The keyword search produced several articles. We screened all the articles and then identified the most relevant ones for Tanzania.

Plate 4: A and B Mature Maasai cattle strain

Plate 5: A and B Mature Iringa red cattle strain

Plate 6: Mature Singida white cattle strain

Plate 7: Mature Ufipa cattle strain
1.6 Organisation of the Thesis

This thesis is arranged in six chapters. The first chapter contains an extended abstract and introduction. The abstract gives brief summary of the results from all presented papers. The introductory part gives highlights of background information on TBDs in Tanzania. It also shows the measures which have been taken by both Government and livestock farmers to alleviate the situation as well as their limitations. It also looks on the potential and limitation of using some tolerant indigenous zebu cattle. The chapter also contains the overall theme of the thesis and descriptions of the general methodology of the study. This chapter ends with a list of references to support issues cited in the study. Chapter two to five of the thesis contain four published papers. Each paper is a chapter in this thesis. Chapter six draws research output of the thesis in which the results obtained from each specific objective are synthesized. This chapter also draws general conclusions and recommendations of the study.

REFERENCES


CHAPTER TWO

2.0 PAPER I

Knowledge and perception on ticks, tick-borne diseases and indigenous cattle tolerance to East Coast fever in agro-pastoral communities of Lake Zone in Tanzania

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Knowledge and perception on ticks, tick-borne diseases and indigenous cattle tolerance to East Coast fever in agro-pastoral communities of Lake Zone in Tanzania

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Abstract
This study was carried out to assess farmers’ knowledge and perceptions on tick species, tick-borne diseases (TBDs), tick control measures, production constraints and tolerance of Tarime and Sukuma zebu cattle to TBDs. The study involved 240 agro-pastoralists from Serengeti, Tarime, Maswa and Meatu districts in Tanzania. Data were collected through individual interview using a structured questionnaire.

Livestock diseases were ranked as the first important problem affecting cattle production in Serengeti and Tarime districts while in Maswa and Meatu districts lack of livestock feeds during the dry season was ranked as the most important constraints, followed by livestock diseases. Among the diseases affecting cattle, the TBDs ranked first in Serengeti and Tarime while in Maswa and Meatu the TBDs ranked fourth and sixth, respectively. Most of the livestock farmers interviewed (74%) knew well the signs of the TBDs. Circling/high stepping, red urine, hard dung and swollen lymph nodes were mentioned as clinical symptoms for heart water, babesiosis, anaplasmosis and East Coast fever (ECF), respectively. The majority of the respondents in all districts knew that ECF is caused by ticks, but did not associate the other TBDs with ticks. The most prevalent ticks were bont ticks (Ambyomma spp), blue ticks (Boophilus spp) and brown ear ticks (Rhipicephalus appendiculatus). Most of the farmers interviewed were using acaricide to control ticks, and the most common method of application was hand spraying. All farmers used Oxytetracycline to treat TBDs; however, some farmers used local herbs. Although most farmers knew the signs of TBDs, they were not spraying/dipping their animals on regular basis because of economic reasons and the belief that their animals always carry ticks without being sick or dying and ECF affects only calves. About half of the farmers considered their breeds to be tolerant to ticks and ECF. In conclusion, the livestock
farmers in the Lake zone have substantial knowledge on tick species and TBD symptoms and they have a perception that ECF is not the most important disease compared to the other TBDs.

**Keywords:** Acaricide application method, tick species, Zebu cattle

### Introduction

Livestock keeping is one of the major economic activities in agro-pastoral and pastoral communities of Tanzania and makes a significant contribution to food security and income (MLFD 2011). Tanzania has 22.8 million cattle, 15.6 million goats, 7.0 million sheep, 35.5 million indigenous chickens, 24.5 million commercial chickens, 2.01 million pigs and 291,960 donkeys (MLFD 2014). Among the livestock species kept in Tanzania, cattle contribute significantly to income, food and nutrition security of livestock keepers. Indigenous breeds of cattle make 95% of the national herd and the remaining are improved dairy (3%) and beef (2%) breeds, with the former saving as the main source of livestock products in the country (Msechu 2001). Tanzania Shorthorn Zebu (TSHZ) is the predominant breed and is comprised by a number of strains i.e. Singida White, Mbulu, Gogo, Chaga, Iringa Red, Sukuma, Maasai, Mkalama Dun, Tarime and Pare (Das and Mkonyi 2003).

In many countries, Tanzania inclusive, Ticks and Tick borne Diseases (TBDs) are the major health impediments to improved livestock production and cause considerable economic losses to livestock keepers as they negatively affect growth, milk production, draft power, fertility, quality of hides and survival of all classes of farm animals. However, control of ticks by using acaricides is increasingly becoming too expensive for the average livestock farmer (Mugisha et al 2005). Currently most livestock keepers do not use appropriate rate of acaricide recommended by the manufacturer due to high prices (Okello-Onen and Rutagwenda 1998).

Studies have shown that the intensive application (on weekly basis) of acaricide is uneconomical and unsustainable in the traditional livestock production system where indigenous cattle are kept (Pegram et al 1993). This is because the conventional tick and TBDs control programmes in many tropical countries were developed for tick eradication aiming to protect introduced exotic cattle breeds. The weekly application of acaricides has been adopted despite the fact that resistance to tick infestation differs among cattle breeds. Zebu cattle have much higher resistance to ticks and TBDs compared to Boran and European breeds (Glass and Jensen 2007).

Little research efforts have been done to put into consideration the knowledge and perception of indigenous/local farmers on disease management of their indigenous animals. The indigenous knowledge of local livestock keepers on tick and TBDs control is of paramount importance in the design of disease control programmes and adds value to modern animal health care system. Hence, the whole concept of tick and TBDs control programme need to be revised to incorporate indigenous knowledge and perceptions of farmers on diseases and available diseases control options (Geerlings 2001). The information on the use of indigenous knowledge in tick and TBDs control is scarce, especially in the Lake Zone of Tanzania where the majority of the livestock are kept. Therefore, epidemiological information based on livestock keepers’ knowledge and experiences is required in order to design effective control measures. This study was
conducted to assess the farmers’ knowledge and perceptions on ticks, TBD and their control measures, and perceptions of farmers with regard to tolerance of their breeds to TBDs as well as livestock production constraints.

Materials and Methods

Study area
The study was carried out in Tarime and Serengeti districts (Mara region) and Maswa and Meatu districts (Simiyu region) in the Lake Zone of Tanzania between March and June 2013. Mara region lies between latitudes 1° 0’ and 2° 31’ south of Equator and between longitudes 33° 10’ and 35° 15’ east of Greenwich while Simiyu region lies between latitude 2° 1” and 4° south of Equator and between longitudes 33°3” and 35°1” east of Greenwich. In Tarime and Serengeti districts Tarime Zebu cattle are kept while in Maswa and Meatu districts the Sukuma cattle are kept. Both Tarime and Sukuma cattle belong to the Tanzania Shorthorn zebu breed. The dominant farming system in all districts is agro-pastoral production system in which livestock farmers practice herded grazing in communal and fallow lands.

Sampling procedure
Purposive sampling technique was employed and the sampling frame was districts, villages and finally households. The purposive sampling was used to select the districts and villages with large numbers of the zebu strains which were of interest for the study. In each of the four districts, three wards and two villages per ward were selected, forming a total of 12 wards and 24 villages. Within a village the list of households keeping Tarime or Sukuma zebu cattle were used as a sampling frame from which respondents were picked randomly using a table of random numbers. Ten households per village were sampled, giving a sample size of 60 households in each district (240 respondents in total). The heads of the households were the main respondents. However, other members of the household, whenever necessary, had an opportunity to provide supplementary information. Information collected was; household socio-economic characteristics, livestock production constraints, most important disease in the area, cattle mortality rate due to TBDs in the area, ticks species and their control. This information was collected using structured questionnaires.

Data analysis
The information collected were coded and recorded into the spreadsheets and the data were analysed using the Statistical Package for Social Sciences (SPSS), statistical software Release 16.0 (SPSS 2008). The following descriptive statistics were generated: means, standard deviations, frequencies and percentages. The percentage of farmers’ response for a particular variable such as age and education of the farmers, experience in keeping livestock, important diseases, common tick species, tick control method used, frequency of dipping/spraying, tolerance of animals to ticks and ECF were compared among the districts using a chi–square test to test whether the proportions in the four districts were different from each other. The differences among the districts were considered significant at P ≤ 0.05.

Results
Household social characteristics
Most of the farmers in the surveyed districts were above 35 years old (Table 1), indicating that livestock farming is commonly practiced by old people in the study areas. More than
half of the respondents had at least basic education. There were more farmers with primary education in Meatu, Serengeti and Tarime compared to Maswa. Most of the respondents had experience of more than 10 years in livestock keeping.

**Livestock production constraints**

Livestock diseases were ranked as the topmost problem affecting cattle production in Serengeti and Tarime districts and were reported to cause high mortalities and reduced production (Table 2). The farmers in Maswa and Meatu ranked lack of livestock feeds as the major constraints for cattle production, especially during the dry season. The reason is due to the fact that most land in the district is allocated to crop production and forest reserve leaving small marginal land for livestock keeping. Shortage of feeds and water were ranked 2nd and 3rd, respectively. In Serengeti and Tarime districts it was observed that most farmers depended on the great Mara river, although high crocodile population in the river was reported to pose a threat to livestock when drinking water. Other constraints included high price of veterinary drugs, low genetic potential of zebu cattle, and lack of market for their animals especially during the rainy season when roads to livestock markets are not passable and these problems were ranked 5th, 6th and 7th, respectively.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Maswa</th>
<th>Meatu</th>
<th>Serengeti</th>
<th>Tarime</th>
<th>Average</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age of household head</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>15-34 Years</td>
<td>21.7%</td>
<td>21.7%</td>
<td>10.0%</td>
<td>10.0%</td>
<td>17.1%</td>
<td>0.105</td>
</tr>
<tr>
<td>35-64 Years</td>
<td>70.0%</td>
<td>65.0%</td>
<td>76.7%</td>
<td>71.7%</td>
<td>68.7%</td>
<td></td>
</tr>
<tr>
<td>Above 64 yrs</td>
<td>8.3%</td>
<td>13.3%</td>
<td>13.3%</td>
<td>18.3%</td>
<td>13.3%</td>
<td></td>
</tr>
<tr>
<td><strong>Level of education</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No formal education</td>
<td>41.7%</td>
<td>14.8%</td>
<td>13.6%</td>
<td>10.0%</td>
<td>20.0%</td>
<td>0.0001</td>
</tr>
<tr>
<td>Standard four education</td>
<td>0.0%</td>
<td>6.6%</td>
<td>6.8%</td>
<td>20.0%</td>
<td>8.7%</td>
<td></td>
</tr>
<tr>
<td>Primary education</td>
<td>53.3%</td>
<td>67.2%</td>
<td>76.3%</td>
<td>66.7%</td>
<td>65.9%</td>
<td></td>
</tr>
<tr>
<td>Form four secondary</td>
<td>0.0%</td>
<td>6.6%</td>
<td>6.8%</td>
<td>20.0%</td>
<td>8.4%</td>
<td></td>
</tr>
<tr>
<td>education</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Form six and tertiary</td>
<td>0.0%</td>
<td>1.6%</td>
<td>0.0%</td>
<td>0.0%</td>
<td>0.4%</td>
<td></td>
</tr>
<tr>
<td>education</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Experience on cattle keeping</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>≤ 10 years</td>
<td>10.0%</td>
<td>19.7%</td>
<td>37.3%</td>
<td>36.7%</td>
<td>25.9%</td>
<td>0.001</td>
</tr>
<tr>
<td>&gt; 10 years</td>
<td>90.0%</td>
<td>80.3%</td>
<td>62.7%</td>
<td>63.3%</td>
<td>75.8%</td>
<td></td>
</tr>
</tbody>
</table>

(Kosgey 2004)

**Table 2: Ranking of constraints to cattle production in the study areas**

<table>
<thead>
<tr>
<th>Constraints</th>
<th>Ranking order</th>
<th>Chi square</th>
<th>df</th>
<th>P - value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diseases</td>
<td>(0.192) 2</td>
<td>(0.229) 2</td>
<td>(0.174) 1</td>
<td>(0.170) 1</td>
</tr>
<tr>
<td>Shortage of feeds and grazing land</td>
<td>(0.262) 1</td>
<td>(0.250) 1</td>
<td>(0.152) 2</td>
<td>(0.165) 2</td>
</tr>
<tr>
<td>Shortage of water</td>
<td>(0.147) 3</td>
<td>(0.160) 3</td>
<td>(0.126) 4</td>
<td>(0.145) 3</td>
</tr>
<tr>
<td>Low genetic potential of the animals</td>
<td>(0.068) 7</td>
<td>(0.083) 4</td>
<td>(0.117) 5</td>
<td>(0.108) 5</td>
</tr>
<tr>
<td>Shortage of labour</td>
<td>(0.065) 8</td>
<td>(0.058) 8</td>
<td>(0.105) 6</td>
<td>(0.096) 7</td>
</tr>
<tr>
<td>Lack of markets for livestock products</td>
<td>(0.078) 5</td>
<td>(0.075) 6</td>
<td>(0.088) 8</td>
<td>(0.089) 8</td>
</tr>
<tr>
<td>High price of veterinary drugs</td>
<td>(0.118) 4</td>
<td>(0.067) 7</td>
<td>(0.150) 3</td>
<td>(0.127) 4</td>
</tr>
<tr>
<td>Poor fertility of the animals</td>
<td>(0.070) 6</td>
<td>(0.078) 5</td>
<td>(0.090) 8</td>
<td>(0.100) 6</td>
</tr>
</tbody>
</table>

Index in bracket calculated as = Sum of (5×rank1) + (4×rank2) + (3×rank3) + (2×rank4) + (1×rank5) 
(Totalr5×rank1)+(Totalr4×rank2)+(Totalr3×rank3)+(Totalr2× rank 4)+(Totalr5×rank5)
Important Diseases in the study areas

The respondents were asked to rank the diseases according to frequency of occurrence, morbidity and animal deaths resulting from each disease. The most predominant diseases (using pair wise score ranking) were ranked differently based on the farmers’ experiences in each district (Table 3). Blackquarter (BQ), lumpy skin disease (LSD) and foot and mouth disease (FMD) were ranked 1st, 2nd and 3rd in Maswa and Meatu while in Serengeti and Tarime districts babesiosis, heart water and FMD were ranked as 1st, 2nd and 3rd, respectively. However, based on the overall scores the result show that, the most important disease was FMD (with 32 scores), followed by babesiosis and LSD (with 31 scores) while ECF had a score of 23.

The majority of livestock keepers were aware of the symptoms of different TBDs in all the districts surveyed. The symptoms that were mentioned included swelling of the external lymph nodes and circling or high stepping for East Coast fever and Heartwater, respectively. Babesiosis was reported to be characterised by passing of red urine while Anaplasmosis was described by emaciation and production of hard dung. The livestock keepers interviewed had adequate knowledge about TBDs and they knew the different diseases in their vernacular languages. For instance, in Serengeti and Tarime districts ECF was called “Chintura” by the kurya people while in Maswa and Meatu it was called “Madundo” in sukuma local language. Although the respondents in Maswa and Meatu did not rank TBDs as the most important diseases, the mortality reported by the farmers in the area shows that TBDs, specifically Babesiosis and Heartwater, were the leading diseases in causing deaths (Table 4). Other diseases which were ranked high included BQ, LSD, FMD, trypanosomiasis, CBPP and helminthiasis.

Table 3: Most important diseases as perceived by farmers in the four districts

<table>
<thead>
<tr>
<th>Disease</th>
<th>Maswa</th>
<th>Meatu</th>
<th>Serengeti</th>
<th>Tarime</th>
<th>Overall score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anaplasmosis</td>
<td>8</td>
<td>8</td>
<td>7</td>
<td>7</td>
<td>14</td>
</tr>
<tr>
<td>Babesiosis</td>
<td>7</td>
<td>4</td>
<td>1</td>
<td>1</td>
<td>31</td>
</tr>
<tr>
<td>Blackquarter</td>
<td>1</td>
<td>2</td>
<td>8</td>
<td>8</td>
<td>25</td>
</tr>
<tr>
<td>Contagious Bovine</td>
<td>6</td>
<td>9</td>
<td>9</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Pleuropneumonia</td>
<td>4</td>
<td>7</td>
<td>4</td>
<td>6</td>
<td>23</td>
</tr>
<tr>
<td>East Coast Fever</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>32</td>
</tr>
<tr>
<td>Heart water</td>
<td>9</td>
<td>6</td>
<td>2</td>
<td>2</td>
<td>25</td>
</tr>
<tr>
<td>Helminthiasis</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>9</td>
<td>5</td>
</tr>
<tr>
<td>Lumpy skin disease</td>
<td>2</td>
<td>1</td>
<td>5</td>
<td>4</td>
<td>31</td>
</tr>
<tr>
<td>Trypanosomiasis</td>
<td>5</td>
<td>5</td>
<td>6</td>
<td>5</td>
<td>23</td>
</tr>
</tbody>
</table>

Note: Each disease was given a score as follows; 1st Disease =10 points, 2nd Disease = 9 points, 3rd Disease = 8 points, 4th Disease = 7 points, 5th Disease = 6 points, 6th Disease = 5 points, 7th Disease = 4 points, 8th Disease = 3 points, 9th Disease = 2 points and 10th Disease =1 point.

Table 4: Number of animal affected by Tick Borne Diseases and resulting mortality

<table>
<thead>
<tr>
<th></th>
<th>Maswa</th>
<th>Meatu</th>
<th>Serengeti</th>
<th>Tarime</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Died %</td>
<td>Died %</td>
<td>Died %</td>
<td>Died %</td>
<td>Died %</td>
</tr>
<tr>
<td>Anaplasmosis</td>
<td>50</td>
<td>52</td>
<td>60</td>
<td>37</td>
<td>62</td>
</tr>
<tr>
<td>Babesiosis</td>
<td>18</td>
<td>34</td>
<td>44</td>
<td>200</td>
<td>176</td>
</tr>
<tr>
<td>East Coast Fever</td>
<td>5</td>
<td>3</td>
<td>60</td>
<td>55</td>
<td>42</td>
</tr>
<tr>
<td>Heart water</td>
<td>5</td>
<td>3</td>
<td>60</td>
<td>112</td>
<td>88</td>
</tr>
</tbody>
</table>
Mortality rates of TBDs infected animals
Farmers were able to recall exactly the number of cases when death occurred as well as the clinical signs shown by the animals before death. They were also able to remember the months when the death occurred. In general most deaths occurred during or just after the rainy season. Babesiosis was the leading cause of death in Meatu (88%) and Tarime (89%) while heartwater and anaplasmosis were the main cause of death in Maswa and Serengeti (Table 4).

Tick species and control methods
The most common tick species found in the study areas (Table 5) were bont ticks (Amblyomma spp), blue ticks (Boophilus spp) and brown ear ticks (Rhipicephalus appendiculatus). Ticks were reported to be common after the rains. Most of the interviewed farmers mentioned that the brown ear ticks were commonly seen on both calves and adults while the other ticks were rarely seen on calves. The farmers’ knowledge of ticks was influenced by geographical location and the extent of animals’ exposure and susceptibility to ticks. Farmers in Serengeti and Tarime districts, where tick infestation was high, were able to identify different species of ticks by their vernacular languages e.g. brown ear ticks (Rhipicephalus) are called Bitotona, blue ticks (Boophilus) are known as Rituru and bont ticks (Amblyomma) called Engoha. In Maswa and Meatu districts, where the intensity of ticks was low, no specific names were mentioned and the farmers just identified all species as “ng’hundya” in Sukuma language, which means ticks.

The majority of the interviewed farmers practiced tick control using either dipping or spraying with acaricide as a means of controlling TBDs while the rest reported that they do not exercise tick control measures. Out of 240 respondents interviewed, close on half acknowledged that their animals are tolerant to ticks and ECF. However, in Maswa only few farmers said that their animals were tolerant to ticks.

Table 5: Common tick species, tick control methods and opinions of livestock keepers on tolerance of their breeds to ticks and tick-borne diseases

<table>
<thead>
<tr>
<th>Variable</th>
<th>Districts</th>
<th></th>
<th></th>
<th></th>
<th>Mean Percentage</th>
<th>P - value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type of ticks</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Brown ear ticks</td>
<td>70.8</td>
<td>66.7</td>
<td>76.9</td>
<td>86.7</td>
<td>70.3</td>
<td>0.22</td>
</tr>
<tr>
<td>Blue ticks</td>
<td>80.0</td>
<td>96.0</td>
<td>78.2</td>
<td>79.6</td>
<td>83.5</td>
<td>0.001</td>
</tr>
<tr>
<td>Bont ticks</td>
<td>90.2</td>
<td>81.8</td>
<td>86.2</td>
<td>83.8</td>
<td>85.5</td>
<td>0.29</td>
</tr>
<tr>
<td>Acaricide application method</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dipping</td>
<td>0.00</td>
<td>0.00</td>
<td>20</td>
<td>68.3</td>
<td>22.1</td>
<td>0.0001</td>
</tr>
<tr>
<td>Spraying</td>
<td>91.7</td>
<td>96.7</td>
<td>68.1</td>
<td>25.1</td>
<td>70.4</td>
<td>0.0001</td>
</tr>
<tr>
<td>Neither</td>
<td>8.3</td>
<td>3.3</td>
<td>11.9</td>
<td>6.6</td>
<td>7.5</td>
<td>0.35</td>
</tr>
<tr>
<td>Animals are tolerant to ticks</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>50</td>
<td>10</td>
<td>58</td>
<td>48</td>
<td>41.5</td>
<td>0.000</td>
</tr>
<tr>
<td>No</td>
<td>50</td>
<td>90</td>
<td>42</td>
<td>52</td>
<td>58.5</td>
<td>0.000</td>
</tr>
<tr>
<td>Animals are tolerant to ECF</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>50</td>
<td>38</td>
<td>54</td>
<td>32</td>
<td>43.5</td>
<td>0.000</td>
</tr>
<tr>
<td>No</td>
<td>50</td>
<td>62</td>
<td>46</td>
<td>68</td>
<td>56.5</td>
<td>0.000</td>
</tr>
</tbody>
</table>
Frequencies of spraying/dipping to control ticks
Dipping/spraying regime for tick control was irregular, ranging from once per week to no dipping/spraying at all (Table 6). The majority of the farmers reported that they apply acaricide once per month. Farmers who were applying acaricides by spraying knew the brands, price for each brand, concentration recommended for each brand and could compare the efficiency of different types of acaricides used. Some of the brands used were super dip (chlorhexidine gluconate), paranex (alphacypermethrin), dominex (alphacypermethrin), cybadip (cypermethrin) and tixfix (amitraz). Those who were controlling ticks by dipping did not know the type of acaricides used, but only knew the price of dipping animals (i.e. Tshs 200.00 per animal for cattle and Tshs 50.00 per animal for small ruminants). According to the respondents this price was prohibitive for those having larger herds. The farmers who were practicing dipping also did not know the concentration of acaricides in the dip.

<table>
<thead>
<tr>
<th>Spray frequencies</th>
<th>Maswa</th>
<th>Meatu</th>
<th>Serengeti</th>
<th>Tarime</th>
<th>Average (%)</th>
<th>P - value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Once per week</td>
<td>12</td>
<td>20</td>
<td>10</td>
<td>10</td>
<td>13</td>
<td>0.0001</td>
</tr>
<tr>
<td>Twice per month</td>
<td>0</td>
<td>0</td>
<td>25</td>
<td>23</td>
<td>12</td>
<td>0.0001</td>
</tr>
<tr>
<td>Once per month</td>
<td>5.7</td>
<td>66.7</td>
<td>58.4</td>
<td>41</td>
<td>55.9</td>
<td>0.0001</td>
</tr>
<tr>
<td>Once every three month</td>
<td>5</td>
<td>15</td>
<td>0</td>
<td>0</td>
<td>5</td>
<td>0.0001</td>
</tr>
<tr>
<td>Twice per year</td>
<td>5</td>
<td>10</td>
<td>0</td>
<td>15</td>
<td>7.5</td>
<td>0.0001</td>
</tr>
<tr>
<td>Once per year</td>
<td>12</td>
<td>15</td>
<td>0</td>
<td>0</td>
<td>6.8</td>
<td>0.0001</td>
</tr>
<tr>
<td>Not at all</td>
<td>8.3</td>
<td>3.3</td>
<td>6.6</td>
<td>11.9</td>
<td>7.3</td>
<td>0.0001</td>
</tr>
</tbody>
</table>

Discussion

Household demographic characteristics

The age, education and experience of the household heads observed in Maswa, Meatu, Serengeti and Tarime districts conform to those reported by other researchers elsewhere (Chenyambuga et al 2008; Kivaria et al 2012). These results indicate that the majority of household heads were in the active age group, attained formal basic education and have a long experience in livestock production. This suggests that the farmers interviewed have good knowledge and skills on livestock diseases and could provide relevant information on how to manage and control them. Nkonya et al (2004) reported that in any rural community education provide better opportunities to access information and services and enable farmers to take correct actions. This in turn makes it possible for the local farmers to shift from traditional to modern way of practices/life. Furthermore, farmers with formal education can have a good opinion on how their breed can be managed for sustainability (URT 2002).

Livestock production constraints

Livestock diseases were ranked as the major constraints of cattle production in Maswa, Meatu, Serengeti and Tarime districts. This observation is in agreement with the findings reported by other researchers (Chenyambuga et al 2008; Maingi and Njoroge 2010). Tick-borne diseases were the most important diseases in Serengeti and Tarime districts while BQ and LSD were ranked as the most important diseases in Maswa and Meatu districts. This is probably associated with the higher amount of rainfall in the former districts.
compared to the latter districts. Tick-borne diseases are more prevalent in areas with high rainfall due to high tick infestations caused by the conducive environment (Ohaga et al 2007).

**Knowledge and perceptions of livestock farmers on ticks, tick-borne diseases and their control measures**

The respondents’ good knowledge on diseases can partly be attributed to the art of diagnosis and treating livestock diseases acquired through experiences whereby all family members are involved in animals management. In the traditional societies knowledge and skills on livestock management are usually passed from fathers to children when they are still very young such that when one grows up he is able to take care of his herd. The traditional knowledge of disease management among pastoralists has been reported by FAO/links (2000), with the recommendation of the need to preserve this knowledge. In general the farmers were knowledgeable on ticks and TBDs, though there were some differences among the districts influenced by geographical location and the extent of exposure to tick infestation. The farmers’ opinion that ticks are most common after the rains is due to the fact that ticks occur throughout the year and during the rainy season farmers have other farm activities, hence being unable to make close observations to their animals. The same observation has been reported by Chenyambuga et al (2008).

Other studies based on participatory research methods (Mugisha et al 2005) in Uganda have reported ECF to be the most prevalent cattle disease, followed by trypanosomiasis. In Tanzania ECF, anaplasmosis and heartwater have been reported to be one of the most important killer diseases (Swai et al 2007). However, in this study the livestock farmers in Maswa, Meatu, Serengeti and Tarime districts considered ECF as a minor problem. This is because of the farmers’ perception that Tarime and Sukuma zebu cattle are tolerant to ticks and TBDs. While other TBDs were a problem to all age groups, farmers pointed out that ECF is a problem only to the calves. This is in agreement with Chenyambuga et al (2008) who reported that ECF is perceived by most farmers as the disease of calves.

Our observation show that, a number of diseases were treated as they occurred, including ECF, anaplasmosis, heartwater and internal parasites. However, cattle vaccinations against TBDs were not carried out in these areas. Treatments for disease were jointly administered by veterinarians and individual farmers who have acquired the knowledge through experience. In the four districts surveyed, it is common to see farmers procuring drugs in bulk and acaricides during the livestock auction days. The common drug used to treat TBDs was Oxytetracycline. In addition to Oxytetracycline treatment, the livestock keepers commonly practice lymph node burning with hot iron for the treatment of ECF, particularly in Maswa and Meatu. This is the evidence that the farmers in the study areas have adequate knowledge on ECF. These findings on farmers’ knowledge on disease and their control concurs with that reported by Jacob et al (2004) that pastoralists have superior diagnostic skills for animal diseases and their diagnosis conform to the veterinarian disease diagnosis criteria. This knowledge is orally passed on from one generation to the next, particularly from the elders to the young.

The results on the use of acaricide to control ticks and tick borne diseases disagree with Chenyambuga et al (2008) who reported that in some wards of Tarime district most farmers do not use acaricide to control ticks and TBDs due to the belief that their animals are tolerant. Our results showed that, even though most farmers still believe that their animals are tolerant to diseases, they have changed the attitudes with regard to
dipping/spraying of their animals. More farmers reported that they dip their animals, but this depended on the availability/accessibility of dips. The farmers living far away from the dips reported that they spray their animals. The reason for the change in attitude was that many farmers have been made aware about the effects of ticks and TBDs through extension services provided by the government using village extension officers (Laisser et al 2014). However, the spraying/dipping regime practiced by the farmers was different from the recommended routine dipping of once per two weeks for indigenous cattle in tick-borne disease endemic areas (Glass and Jensen 2007). In the present study the majority of the farmers were spraying/dipping once per month. The main reason given was that the high cost of acaricides prohibits the adoption of weekly or bi-weekly dipping. This observation is in agreement with the findings reported by Mugisha et al (2005) in pastoral societies of Uganda. Generally, the farmers have adopted irregular use of acaricide which is not based on a well planned tick control programme. This is due to the fact that, the intensive application (weekly application) of acaricide is uneconomical and unsustainable in indigenous cattle as pointed out by Pegram et al (1993). This study has revealed that most farmers use hand spray to apply acaricides to animals and they are aware that the regular acaricide application is the most effective way for TBDs control, but they do not apply regularly due to economic reason. This is in agreement with the observation made by Mugisha et al (2005) that spraying is the most preferred method for acaricide application in pastoral and agro-pastoral communities. However, the tendencies of some farmers to spray acaricide on the animals when sleeping do not guarantee that all parts of animal body become in contact with the acaricides. This practice is not efficient in killing all ticks and the problem is aggravated by the fact that inappropriate dilutions of acaricides are used.

The inappropriate application of acaricides coupled with the fact that animals sprayed intermingle with other animals which have not been sprayed/dipped when grazing in the communal lands can lead to development of tick resistance to acaricides. In the traditional livestock production systems where animals from different herds are mixed during grazing, it is suggested that dipping should be used for applying acaricides as it is more efficient and reliable in controlling ticks compared to spraying. When spraying is opted, farmers need to be trained on how to prepare the correct dilution of the acaricide and the sites for tick attachment on the body of the animal where spraying should be focused. It also important for livestock extension officers, before introducing any strict tick control system to consider farmers experience. In other study it was found that in some wards of Serengeti district where strict tick control measure was implemented, the levels of tick infestation decreased, but increased the incidences of animal to succumb to ECF infection compared to the wards in which there was no strict dipping/spraying (Laisser et al 2014).

Although farmers in Maswa and Meatu did not rank TBDs as the most important diseases the mortality percentages reported by the farmers in the area show that TBDs, specifically babesiosis was the leading cause of death. In Maswa and Meatu the high rank was given to BQ due to the fact that at the time of this survey there was an outbreak of BQ disease in the area and farmers tended to recall the event which was burning at that particular time.

The difference in ranking of tick-borne diseases between Serengeti and Tarime on one hand and Maswa and Meatu on the other hand is due to the different management systems. In Serengeti and Tarime cattle graze in communal land throughout the year. This grazing system makes it difficult to control animal diseases in pastoral and agro-pastoral communities due to mixing of herds from different households. Furthermore, this practice
tends to enhance the spread of ticks and TBD infections across the herds (Ogutu 2012). In Maswa and Meatu, animals from different herds do not mix and each farmer grazes his/her herd in a private grazing area. This is because of the shortage of grazing areas and each farmer needs to reserve a portion of grazing land known as “Kitiri” for using during the dry season. This kind of management helps to reduce tick burden and spread of disease incidences. Furthermore, grazing around the National park noted in Serengeti district is a predisposing factor for tick-borne diseases like ECF which can spread from wild buffalo to cattle (Marcellino et al 2012). It is suggested that farmers in all districts adopt controlled grazing and feed conservation as these can significantly reduce disease incidences of their livestock.

Farmers’ perception on tolerance of Tarime and Sukuma breeds to ticks and East Coast fever

In Serengeti and Tarime districts, the most common breed kept by local people is the Tarime zebu. This strain of TSHZ was the most preferred breed by the majority of the farmers in both districts. The farmers preferred this breed because they consider it to be more tolerant to ticks and ECF compared to the other TSHZ strains and they can survive without dipping/spraying with acaricide. This concurs with the findings reported by Chenyambuga et al (2008) in some wards of Tarime district. In Maswa and Meatu districts the predominant breed was the Sukuma zebu cattle. The livestock farmers in these districts preferred this breed because of its medium size, good shape and udder size, well placed teats and its docile temperament. The same findings have been reported by Akeyo (2012). In all four districts, disease resistance, low input in terms of treatment, feeding and ability of animals to survive under the harsh local conditions were the main reasons for keeping the local breeds. This observation concurs with the findings reported by Wambura et al (1998) who found zebu cattle to be relatively resistant to tick infestation compared to crossbreed animals.

The general perception of the livestock keepers in the four districts was that their local cattle are tolerant to ECF. The belief that Tarime and Sukuma cattle are tolerant to ECF is supported by the evidence provided by the respondents that their animals survive without regular dipping/spraying and that infected animals do recover without treatment using veterinary drugs. This implies that the indigenous breeds in the Lake zone of Tanzania possess survival traits which enable them to live and produce under tick challenges. This is in agreement with Rege and Tawah (1999) who stated that indigenous cattle are blessed with tick resistance and tolerance to vector-borne diseases and they perform better than exotic breeds under low-input conditions, climatic stresses, especially during times of drought. However, according to Chenyambuga et al (2008) and Kipronol (2009), ECF, which is caused by Theileria parva, has more serious effects on calves than in adult cattle. Therefore, calves need to be protected by regular dipping/spraying as they are important for herd growth and form the replacement stock.

Conclusions

Livestock diseases are the most important constraints to cattle production in the Lake zone regions of Tanzania. These are followed by shortage of forages and water during the dry season and shortage of grazing land. Among the diseases, tick-borne diseases rank first in the agro-pastoral communities of Lake Zone. The livestock farmers in the Lake zone have substantial knowledge on ticks and TBDs symptoms. Despite the extensive
knowledge, most livestock farmers do not apply acaricide regularly to control TBDs due to economic reason and the belief that their animals can tolerate ticks and TBDs. Tick control strategies should be harmonized with the indigenous knowledge of the livestock farmers on tolerance of local breeds to ticks and tick-borne diseases.

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Conflict of interest

The authors declare that they have no conflict of interest and this document is their original research work done in Lake Zone of Tanzania and no part of it has been submitted somewhere else for conference presentation or publication.

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CHAPTER THREE

3.0 PAPER II

Tick burden and prevalence of *Theileria parva* infection in Tarime zebu cattle in the lake zone of Tanzania

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Tick burden and prevalence of *Theileria parva* infection in Tarime zebu cattle in the lake zone of Tanzania

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Abstract This study was carried out to assess the distribution, abundance of different tick genera and prevalence of *Theileria parva* infection in Tarime zebu cattle kept in selected wards of Serengeti and Tarime districts in Mara region. Adult ticks were identified and counted from half body parts of 360 animals which were extensively managed in communal land with natural pastures. Concurrently, blood samples were collected and thereafter DNA extracted and a nested polymerase chain reaction (nPCR) was done using primers specific for p104 gene to detect the presence of *T. parva* DNA. Ticks were identified into four groups: *Amblyomma* genus, *Boophilus* sub-genus of *Rhipicephalus* genus, other species of *Rhipicephalus* and *Hyalomma* genus. *Rhipicephalus* genus accounted for 71.8 % of the total ticks, whereas *Amblyomma*, *Boophilus* sub-genus of *Rhipicephalus* genus and *Hyalomma* constituted 14.1, 14.0 and 0.1 %, respectively. There were more animals (*p* < 0.05) infected with ticks in Tarime district (96.1 %) than in Serengeti (61.7 %). The average counts of ticks were higher in adult animals (*p* < 0.05) than in young animals. The overall prevalence of *T. parva* was 27.7 % and was higher (*p* < 0.05) in Serengeti (38.3 %) than in Tarime district (16.7 %). However, all animals tested positive for *T. parva* did not show any clinical signs of East Coast fever (ECF), suggesting the existence of subclinical infection in Tarime zebu. These results suggest that Tarime cattle can tolerate ECF infection and are likely to serve as potential carriers of *T. parva* to other less-tolerant cattle breeds in mixed herds. Since Tarime cattle are preferred by most farmers with mixed herds, routine screening for *T. parva* is highly recommended to minimize introduction of infected cattle into an immunologically naive population.

Keywords East coast fever · Tarime zebu · Tick species · Tolerance

Introduction

In Tanzania, about 80 % of the total cattle herds are at risk of being infected with tick-borne diseases (TBDs) annually, and the direct economic losses are estimated at US$ 248 million with mortality rate estimated at 920,000 animals/year (Kivaria 2006). East Coast fever (ECF) is one of these diseases and causes substantial losses in cattle and therefore is a socioeconomic threat to the development of the livestock sector (Mbasa et al. 1994; Swai et al. 2007; Chenyambuga et al.)
The study involved six wards from Tarime and Serengeti districts namely Matongo, Komoso, Kibasa, Kubanjbaru, Rung’abure and Kisaka. Two villages were purposely selected from each ward making a total of 12 villages for both districts. In each village, 10 households (120 households in total) were randomly selected using random numbers from the list of all households keeping more than 10 Tarime zebu. From these households, a total of 360 animals (three per household, i.e. one calf, one ewe and one adult) were selected for tick identification and counting as well as blood sampling. The desired sample size for the study was calculated according to the procedure described by Thusfield (1995), and the animals were selected based on no evidence of admixture with other strains of zebu.

Tick identification and counts

Tick counts were done according to Lundt et al. (1979), counting ticks at predilection sites. The predilection sites were the head, sternum and hind quarters (Brown and Ducasse 1967). Due to limited resources and time, visible adult ticks were counted from one side of the animal body and identified using the keys of Mathysse and Colbo (1987). The half-body counts were doubled to estimate the number of ticks for the whole body. The ticks were identified to genus level and recorded for each animal examined; animal category, tick attachment sites, household head, village, ward and district were also recorded.

Blood sampling, genomic DNA extraction and detection of Theileria parva

Whole blood samples were collected by jugular vein puncture using Vacutainer tubes with EDTA. The samples were labelled and stored in a cool box with ice packs while in the field and later put into a refrigerator until when they were transferred to the laboratory in the Department of Microbiology and Pathology at SUA for further analyses.

Genomic DNA was extracted from 354 whole blood samples using the Puregene Blood-Core Kit (Qing, Minnesota, USA) according to the manufacturer’s instructions (six samples clotted before DNA extraction and they were therefore omitted). The nested p104 PCR was used to screen all field samples for the presence of T. parva. Primers derived from the T. parva-specific 104-kDa antigen (p104) gene were used in the PCR amplification as previously described by Odongo et al. (2010) and Larsen et al. (1990). The sequences of the forward and reverse primers were 5’-ATT TAA GGA ACC TGA CGT GAC TGC 3’ and 5’ TAA GAT GCC GAC TAT TAATGA CAC C 3’, respectively, for first round and 5’ GGC CAA GGT CTC CTT CAG AAT AGC 3’ and 5’T GG TGT GTG GGT TTC CTC GTC ATC TGC 3’, respectively, for the second round. The nested polymerase chain reaction (nPCR) amplifications were performed in a total volume of 20 μl containing 14 μl nuclease-free water, 0.5 μl (10 pmol) each of forward and reverse primers and 5 μl of genomic DNA (20 ng/μl) template added into the lyophilized pellet (Bioneer PCR Premix—Korea), followed by vortexing and brief spin down to dissolve the pellet. For the second round, the amount of water was 18.5 μl, and 0.5 μl of the primary PCR product was used as a template. Reaction conditions for the primary PCR included initial denaturation at 94 °C for 5 min, denaturation at 94 °C for 60 s, annealing at 60 °C for 60 s and extension at 72 °C for 60 s, and the amplification was done in 30 cycles. The cycling profile condition for the second PCR was the same as the primary amplification, except for the annealing temperature which was 50 °C. The nPCR reactions were carried out in a thermocycler (Takara Bio Inc., Japan).
The nPCR products were separated on 1.5 % agarose gel and visualized in an ultraviolet (UV) trans-illuminator.

Data analysis

The data obtained from tick count and PCR were coded and analysed using SPSS version 16 (SPSS 2008). The percentage of animals infested with different tick species and the prevalence of T. parva in animals were compared using a chi-square to test the significance of the differences in tick infestation and prevalence of T. parva between villages, wards and districts. Analysis of variance was used to assess the statistical significance differences in tick counts between wards and between districts. The fixed effects assessed were district, ward within district, animal category and attachment site. All results were considered significant at \( p \leq 0.05 \).

Results

Tick species and abundance in Serengeti and Tarime districts

Four genera of ticks were identified from a total of 360 animals. The genera recorded were Rhipicephalus (71.8 %), Amblyomma (14.1 %), Boophilus sub-genus of Rhipicephalus genus (14.0 %) and Hyalomma (0.1 %). Three of these (Rhipicephalus, Amblyomma and Boophilus sub-genus of Rhipicephalus genus) were found in both districts whereas Hyalomma was found only in Tarime district. The most abundant and widely distributed ticks in both districts belonged to Rhipicephalus, followed by Amblyomma and Boophilus sub-genus of Rhipicephalus genus as shown in Table 1. With an exception to Amblyomma, the distribution of all other tick species differed significantly \( (p \leq 0.05) \) between the two districts, with Tarime district having the highest counts for each species. Of the examined animals in Serengeti and Tarime, 61.7 and 96.1 %, respectively, were found to be infested by ticks. The proportion of animals infested with ticks differed significantly \( (p \leq 0.05) \) between districts.

<table>
<thead>
<tr>
<th>District</th>
<th>Parameter</th>
<th>Rhipicephalus</th>
<th>Amblyomma</th>
<th>Boophilus</th>
<th>Hyalomma</th>
<th>( p ) value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serengeti</td>
<td>Tick count (meanSE)</td>
<td>32.6±3.7*</td>
<td>15.8±1.6*</td>
<td>9.6±2.4*</td>
<td>0.0</td>
<td>0.00</td>
</tr>
<tr>
<td></td>
<td>% of total ticks</td>
<td>42.8</td>
<td>45.6</td>
<td>25</td>
<td>0.0</td>
<td>0.00</td>
</tr>
<tr>
<td>Tarime</td>
<td>Tick count (meanSE)</td>
<td>89.5±6.3*</td>
<td>20.7±2.7*</td>
<td>30.3±5.9*</td>
<td>4.7±1.5</td>
<td>0.00</td>
</tr>
<tr>
<td></td>
<td>% of total ticks</td>
<td>60.0</td>
<td>52.2</td>
<td>33</td>
<td>1.7</td>
<td>0.014</td>
</tr>
<tr>
<td>Overall</td>
<td>Tick count (meanSE)</td>
<td>71.8±4.3*</td>
<td>18.6±1.7*</td>
<td>23.7±4.1*</td>
<td>4.7±1.5</td>
<td>NA</td>
</tr>
<tr>
<td></td>
<td>% of total ticks</td>
<td>68.9</td>
<td>52.8</td>
<td>38.6</td>
<td>1.7</td>
<td>0.00</td>
</tr>
<tr>
<td>( p ) value</td>
<td>0.00</td>
<td>0.151</td>
<td>0.00</td>
<td>NA</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Means bearing different superscripts across rows are significantly different \( (p < 0.05) \)

Prevalence of T. parva in Tarime cattle

The overall prevalence of T. parva in Tarime cattle was 27.7 % (Table 3). It is shown here that the prevalence of T. parva in cattle blood was significantly \( (p \leq 0.05) \) higher in Serengeti district (38.3 %) than in Tarime district (16.7 %). The prevalence of T. parva infection in different animal categories was not significantly \( (p > 0.05) \) different. There were no significant \( (p > 0.05) \) differences in terms of the prevalence of T. parva among wards of Tarime district, and in most wards, the prevalence was less than 30 %. However, a significant \( (p < 0.05) \) difference in T. parva infection was observed among the wards of Serengeti district. The highest number of animals that tested positive for T. parva infection was found in Rung'abure ward while the lowest was found in Kisaka.

Discussion

The main focus of this study was to establish the tick burden and the prevalence of T. parva, the causative agent of ECF, in

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Table 2. Factors influencing tick burden in Serengeti and Tarime districts.

<table>
<thead>
<tr>
<th>Factors</th>
<th>Level</th>
<th>Tick count (mean±SE)</th>
<th>% of animals infected</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Category</td>
<td>Young</td>
<td>683±68.8±</td>
<td>77.5</td>
<td>0.007</td>
</tr>
<tr>
<td></td>
<td>Young</td>
<td>88.0±11.1</td>
<td>80.0</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Adult</td>
<td>100±11.2</td>
<td>79.1</td>
<td></td>
</tr>
<tr>
<td>2. Attachment site</td>
<td>Head</td>
<td>66.7±1.5</td>
<td>68.1</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>Sternum</td>
<td>160±1.6</td>
<td>51.9</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Hind quarter</td>
<td>23±1.9</td>
<td>63.3</td>
<td></td>
</tr>
<tr>
<td>3. Ward</td>
<td>Kebanjanja</td>
<td>17±2.7</td>
<td>75.0</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>Kisaaka</td>
<td>62±5.8</td>
<td>91.7</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Runghabure</td>
<td>5.2±3.2</td>
<td>18.7</td>
<td></td>
</tr>
<tr>
<td>Tarime</td>
<td>Kibussa</td>
<td>132±16.0</td>
<td>93.3</td>
<td>0.016</td>
</tr>
<tr>
<td></td>
<td>Komato</td>
<td>81±10.5</td>
<td>95.0</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Mutongo</td>
<td>139±17.8</td>
<td>100.0</td>
<td></td>
</tr>
<tr>
<td>4. District</td>
<td>Serengeti</td>
<td>383±3.8</td>
<td>61.7</td>
<td>0.00</td>
</tr>
<tr>
<td></td>
<td>Tarime</td>
<td>117±8.9</td>
<td>96.1</td>
<td></td>
</tr>
</tbody>
</table>

Means bearing different superscripts across columns are significantly different (p<0.05).

Clinically healthy Tarime cattle reared. Tick genem observed in this study conformed to those reported by Chenyambuca et al. (2008) in other wards of Tarime district. In both districts, *Rhipicephalus* was the most prevalent tick genus infecting Tarime cattle. Knowledge of tick species available in a certain area is very important in predicting the occurrence of specific TBDs in such an area and thus helps in planning proper diseases control programme.

In this study, calves had the smallest tick counts compared to weaners and adults due to the nature of calf rearing systems. In Serengeti and Tarime districts, calves are kept around homestead separately from adult animals, this reduces the risk of tick infestation because of the limited exposure. Adult animals are continually grazed in natural pastures in communal lands throughout the year and thus are more prone to tick infestation. Our observation concurs with the findings obtained by Kivaria et al. (2012) in the pastoral community of Southern Sudan where calves and other young animals are left to graze near the homestead. However, our findings are contrary to the findings obtained by Mwambene et al. (2012). In their study, they found that in Fipa communities, calves and adult animals are grazed together; hence, both are exposed to the same tick infestation risk.

High tick burden was observed in Tarime compared to Serengeti; this is attributed to the existence of different tick control systems in the two districts. A strictly community-managed dipping practice has been instituted in Serengeti district and this has reduced significantly the level of tick infestation in Serengeti compared to Tarime district. Also, ecologically, Tarime district is more humid and receives more rainfall than Serengeti district; these climatic conditions are more favourable for tick growth and multiplication. These findings are consistent with previous reports by Mathee et al. (1997) who found that ecological differences, grazing system, extent and frequency of acaricide usage had large influence on tick counts in any ecological zone. Tarime district, therefore, needs to establish strict tick control programme, but it should not exceed the endemic stability. Any control programme should aim at providing sufficient tick control to increase cattle productivity while at the same time not reducing tick numbers to the extent that endemic stability is adversely affected.

The prevalence of *T. parva* infection observed in this study is almost similar to that observed by Gachohi et al. (2012) in pastoral herds in Kenya and Marcellino et al. (2012) for pastoral livestock keepers in Southern Sudan. However, our findings disagree with the previous results reported in different wards of Tarime district (Chenyambuca et al. 2008). The difference observed is due to variation in vector densities from year-to-year due to changes in climatic conditions, socioeconomic activities, seasons and management practices such as dipping practices. The observed high prevalence of *T. parva* in cattle of Serengeti district compared to those of Tarime district is in agreement with the findings by Kivaria et al. (2012) who reported that regular dipping reduces vector intensity which in turn reduces the bovine immune stability. Another reason is that cattle in Serengeti district graze in areas bordering the Serengeti National Park during the dry season due to shortages of pastures and water in the common communal grazing lands. Most areas in the parks are FCF-endemic zones due to the abundance of *Rhipicephalus appendiculatus*, particularly during the end of the wet season (Marcellino et al. 2012). Thus, cattle populations in Serengeti district seem to easily pick up the *T. parva* pustules when grazing in the national park where there are plenty of buffalos which are good carrier of the protozoan (Marcellino et al. 2012).

Our observations of cattle samples revealed that all animals that tested positive for *T. parva* did not show any signs of
Table 3. Factors influencing association with *T. parva* infection (percentage of animals infected in each category, ward and district)

<table>
<thead>
<tr>
<th>Factor</th>
<th>Level</th>
<th>% of animals infected</th>
<th>Chi-square</th>
<th>df</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Negative</td>
<td>Positive</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1. Category</td>
<td>Young</td>
<td>72.9</td>
<td>27.1</td>
<td>2.145</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>Weaver</td>
<td>76.3</td>
<td>23.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Adult</td>
<td>67.8</td>
<td>32.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2. Wards</td>
<td>Serengeti</td>
<td>Kebejebe /Koros</td>
<td>60.0</td>
<td>40.0</td>
<td>4.371</td>
</tr>
<tr>
<td></td>
<td>Kisaika</td>
<td>71.7</td>
<td>28.3</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Rongo /Koros</td>
<td>55.3</td>
<td>46.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tarime</td>
<td>Kibiko</td>
<td>95.0</td>
<td>5.0</td>
<td>9.693</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>Matongo</td>
<td>74.1</td>
<td>25.9</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3. District</td>
<td>Serengeti</td>
<td>61.7</td>
<td>38.3</td>
<td>20.746</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Tarime</td>
<td>83.3</td>
<td>16.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall prevalence</td>
<td>72.3</td>
<td>27.7</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*p < 0.05 (significantly different at this level)*

illness, indicating being infected with *T. parva* but remained healthy and thus are potential carriers. Furthermore, our study has demonstrated that animals having high tick burden do not necessarily become infected with *T. parva*, the parasite that causes ECF. This is because some ticks do not harbour or do not carry the protozoan parasites causing ECF. Generally, our findings on tick burden and *T. parva* prevalence in Tarime zebu concurs with the farmers’ belief that Tarime cattle can tolerate ECF infection and may, therefore, serve as carriers of *T. parva* and other hemoparasites (Ngowi et al. 2008; Chenyambuga et al. 2008). This is because high tick infestation and *T. parva* infection were found in Tarime cattle, but none of the animals examined showed either signs of ill-health as a result of tick infestation or clinical signs of ECF despite the fact that the DNA of *T. parva* was detected in 27.7% of the animals sampled. We can hypothesize that the Tarime zebu can be infected with *T. parva* but cannot develop signs of ECF; hence, they may become a source of infection to other less-tolerant ecotypes of indigenous cattle in mixed herds.

Since Tarime cattle are preferred by most farmers because of their perceived resistance to ticks and ECF, community conservation programmes for the ecotype should be promoted. Studies elsewhere have shown that indigenous cattle are tolerant to ticks and other vector-borne diseases and they perform relatively better than exotic breeds under low-input conditions, climatic stresses and diseases outbreaks, especially during times of drought (Rege and Tawab 1999). Murula (2008) observed that the Nguni cattle are more tolerant to ticks and TBDs than nondescript cattle in the traditional sector in South Africa and that communal farmers keeping Nguni cattle did not require regular treatment with costly veterinary pharmaceuticals for tick and TBD control. A similar scenario may apply in Tarime zebu cattle; this provides opportunity for exploiting the genetic potential of Tarime cattle for tolerance to TBDs. This may be beneficial for the sustainability of the livelihoods of pastoral and agro-pastoral communities, especially now that climate change and variability has frustrated livestock production in many countries in the world, necessitating adoption of more climate and disease-resistant breeds.

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Conflict of interest The authors declare that they have no conflict of interest.

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CHAPTER FOUR

4.0 PAPER III

Tick burden and acquisition of immunity to *Theileria parva* by Tarime cattle in comparison to Sukuma cattle under different tick control regimes in the lake zone of Tanzania

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Tick burden and acquisition of immunity to *Theileria parva* by Tarime cattle in comparison to Sukuma cattle under different tick control regimes in the Lake Zone of Tanzania


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This study was conducted to determine tick burden and immunological parameters of resistance to East Coast fever (ECF) in Tarime and Sukuma cattle. Tick load, packed cell volume (PCV), *Theileria parva* (*T. parva*) specific antibody percent positivity (PP), and prevalence of *T. parva* parasites were studied in relation to dipping regime, strains, and season. A total of 50 experimental cattle were included in this study. Tick load was determined by whole body counts, antibody percent positivity was determined by the polymorphic immunodominant molecule (PIM)-based *T. parva* enzyme-linked immunosorbent assay (ELISA), and prevalence of *T. parva* parasites was detected by a nested polymerase chain reaction (PCR) based on the p104 gene. Dipping frequency on tick burden showed no statistically significant differences when cattle of either strain were dipped either once every 2 or 3 weeks in the dry and wet seasons. However, Tarime cattle had higher (p<0.05) tick count than Sukuma cattle and non-dipped groups maintained high tick infestation throughout the experimental period. The PCV values were within the physiological range, although this parameter was lower in Tarime cattle (p<0.05). All cattle regardless of strain were seropositive, although Tarime cattle maintained higher PP compared to Sukuma by 15%. Conversely, the prevalence of *T. parva* parasites was lower in Tarime (38%) compared to Sukuma cattle (38.5%), but the difference was not significant (p>0.05). During the study period, 20% (5/25) of Sukuma cattle contracted ECF, but none of the Tarime cattle showed clinical signs for the disease. The differences between the two strains shown in terms of PP and *T. parva* parasite prevalence may indicate the ability of individual cattle to resist tick infestation and ECF infection under natural challenge. Higher antibody levels but lower parasite prevalence attained by Tarime cattle suggests inherent ability of Tarime cattle to resist clinical development of ECF infection, but to remain as *T. parva* carriers.

Key words: Carrier state, *Theileria parva*, ticks, seropositivity.
INTRODUCTION

Tanzania is endowed with valuable indigenous strains of cattle (Das and Mkony, 2003). These livestock resources, apart from offering direct food products like meat and milk, also provide draught animal power as labor saving technologies; manure for fertilizing crop fields and biogas for electrification or cooking fuel which has a potential for reducing deforestation (MLFD, 2015). About 80% of the indigenous cattle strains in Tanzania are exposed to vector-borne infections, among which is East Coast fever (ECF) which is a major killer disease (Kivana, 2005) causing substantial losses in terms of morbidity in adult cattle of improved breeds and calf mortality, and therefore, hinders development of the livestock sector (Swai et al., 2007; Cheryambuga et al., 2008). Control strategies for ECF are based on the use of acaricides to control the vector ticks, chemotherapy of sick animals as well as immunization of cattle by the infection and treatment method (ITM) (Norval et al., 1992; Musoke et al., 2004; Oura et al., 2004; Acaricide use and chemotherapy are often limited by high costs, development of resistance by the vector ticks, and the parasites as well as environmental impacts (Mugisha et al., 2005a; Kvarla, 2006; deCastro, 1997; George et al., 2004; Ministry of Water and Livestock Development, 2004). On the other hand, ITM offers a valuable alternative for ECF control (Oura et al., 2004) however, its widespread application has faced many challenges. These include the requirement of cold chain mode of delivery to remote areas and high cost of the vaccine (up to US$10 per animal), which is unaffordable to most smallholder herders (Di Giulio et al., 2009). Furthermore, ITM does not completely eliminate the need for acaricide application due to the potential existence of other tick-borne diseases.

Indigenous strains of cattle found in ECF endemic areas around the Lake zone of Tanzania (that is, areas around Lake Victoria), such as the Tarime cattle are reported to be more tolerant to ECF than exotic cattle and other zebu in the same region (Faling et al., 1991; Cheryambuga et al., 2008). This is probably due to natural selection, because of the long-time exposure to the disease in the region. However, much of available evidence is based on anecdotal information regarding tolerance of Tarime cattle to ECF (Cheryambuga et al., 2008).

The aim of present study was to assess tick burden and immunological parameters of resistance to ECF in Tarime in comparison to Sukuma cattle found in the same zone. Cattle from the two strains were subjected to acaricide application using two different frequencies, either once every 2 or 3 weeks. The findings from this study will make it possible to make strong recommendation about the potential of indigenous strains for rational utilization in ECF endemic areas.

MATERIALS AND METHODS

Study area

This study was conducted at Tanzania Livestock Research Institute (TAILRI), Mabuku centre in Mwanza region, Lake Zone of Tanzania. The centre is located between latitudes 2° 58′ 84° South and longitudes 33° 56′ 12° East at an altitude of 1170 m above sea level. Temperatures at the centre range between 25 and 35°C and rainfall ranges from 600 to 800 mm per annum. The rainfall pattern is bimodal, with short rains starting in November and ending in February and long rains starting in mid March and ending in May.

Study design and animals

A total of fifty cattle (26 Tarime and 25 Sukuma), aged 9 to 12 months were randomly selected from a cohort of 110 cattle, which were previously purchased from farmers around the Lake Zone of Tanzania, where the animals have been kept for many years under smallholder management systems with poor tick control practices. Farmers around the Lake Zone believe that the Tarime and Sukuma strains possess resistance to tick infestation and/or ECF infection. The original cohort of 110 cattle had been kept at Mabuku Research Centre in Mwanza, Tanzania for a year-long monitoring of tick-borne diseases, during which time the cattle were kept under similar management conditions including weekly dipping. At the onset of this study, cattle from each type, shown to be free of ECF (by absence of blood parasites) were divided into groups of eight, eight, and nine. The cattle were individually identified by different colored ear tags and allocated to experimental groups, which were distinguished by different dipping regimes. Cattle in group 1 were dipped once after two weeks while those in group 2 were dipped once every three weeks. The third group, comprising 9 cattle served as control group without dipping throughout the study period. Dipping was done using alphaphenpermethrin (Dominex®) with an initial dip filling of 1 L of Dominex® for 2000 L of water, and dip replenishing by adding 1 L of Dominex® for 1269 L of water. The experiment was conducted during the dry season (mid-August to mid-November 2014) and the wet season (mid-November 2014 to mid-February 2015).

The three cattle groups were grazed together and monitored throughout the study period for tick counts and any ECF clinical signs (fever, that is body temperature above 39.5°C for more than 3 consecutive days; swelling of parotid and prescapular lymph nodes, presence of a macrocytosis index ≥5% on collected smears and anemic respiratory distress). Any diseased animal was promptly treated using buparvaquone (2.5 mg/kg) and all animal biodata was recorded on field sheets.
Sampling
Whole body tick counts were done once every week just before dipping. Only adult ticks were counted and identified to genus level in situ. Once every three weeks blood samples were collected by jugular vein puncture into plain vacutainer tubes for serology and into EDTA-containing vacutainer tubes for whole blood (Poly Medicura Ltd. Fandriken, India) for packed cell volume (PCV) and DNA extraction. A total of 400 blood samples were therefore collected during 6 months in 8 sampling periods at three week intervals. The blood samples were temporarily stored in cool boxes containing ice packs and then transferred to Tanzania Veterinary Investigation and Laboratory Agency (TVLA) in Mwanza for initial processing (PCV and serum extraction) and then transferred to the Faculty of Veterinary Medicine laboratories at Sokone University of Agriculture, Morogoro for further analyses.

Tick identification and counts
Tick counts were done according to Lomot et al. (1979). Visible adult ticks were counted from whole animal body and identified using the keys of Mathysse and Catoto (1987). The ticks were identified to genus level and recorded for each animal in relation to frequency of dipping season and strain.

Genomic DNA extraction and detection of *Theileria parva*
Genomic DNA was extracted from 400 whole blood samples using the Pure Gene Blood Core (QIAGEN) Kit (Minnesota, USA) according to the manufacturer’s instructions. The nested p104 polymerase chain reaction (nPCR) was used to screen all field samples for the presence of *T. parva*. Primers derived from the *T. parva*-specific 104-KDa antigen (p104) gene were used in the PCR amplification as previously described by Odongo et al. (2010) and Kim et al. (1990). The sequences of the forward and reverse primers were 5’ATT TAA GGA ACC TGA CTT GGT AAC S3’ and 5’TAA GAT GCC GAC TAT TAA TGA CAC C3’, respectively, for the first round and 5’GCC CAA GGT TCT CTT CAG AAT AGG S3’ and 5’TGG TGT TCT CTC GTC ATG TGC S3’, respectively, for the second round. The nPCR amplifications were performed in a total volume of 20 μl containing 14 μl nuclelease-free water, 0.5 μl (10 pmol) of each of forward and reverse primers and 5 μl of genomic DNA (20 ng/μl) template added into the lyophilized pellet (Blonier PCR Pre-Mix - Korea), followed by vortexing and brief spin down to dissolve the pellet. Reaction conditions for the primary PCR included initial denaturation at 94°C for 5 min, denaturation at 94°C for 30 s, annealing at 60°C for 30 s and extension at 72°C for 60 s and the amplification was done in 30 cycles. For second round, amount of water was 16.5 and 0.5 μl of the primary PCR product was used as a template. The cycling profile condition for the second PCR was the same as the primary amplification, except for the annealing temperature which was 50°C. The nPCR reactions were carried out in a thermocycler (TAKARA Bio Inc., Japan). The nPCR products were separated on 1.5% agarose gels and visualized on a UV trans-illuminator.

Packed cell volume (PCV)
The PCV was determined using whole blood drawn from the jugular vein into EDTA-containing vacutainer tubes. Blood was drawn from the tubes using capillary tube filled to ¼ of its length. One end of the tube was sealed with cistaseal and then placed into a micro-haematocrit centrifuge (ex UK with 9 cm rotor radius) for 5 min at 12,000 rpm (RCF: 14,462.28 g). Reading of the PCV was performed on the Micro-haematocrit Reader Scale. The PCV was determined from the reading expressed as percentage of packed red cells in the total volume of blood.

Indirect ELISA for *T. parva* antibodies
The PIM-based enzyme-linked immunosorbent assay (ELISA) described by Katende et al. (1999) was used to measure specific antibodies to *T. parva* (sensitivity > 99%, specificity 94 to 98%). Optical density (OD) of each sample was measured at 405 nm using an Erba Luxscan II ELISA reader (Erba diagnostics, Mannheim GmbH, Germany). The OD readings were used to compute antibody percent positivity (PP) for each sample using the formula:

\[
PP = \frac{\text{Mean OD of sample}}{\text{Mean OD of strong positive}} \times 100
\]

The PP of 20% or higher was considered positive.

Data analysis
The data obtained from tick count, PCV, ELISA and PCR were coded and analysed using the statistical package for social sciences (SPSS) research software version 16 (SPSS, 2008). The percent of tick species, PCV, PP, and the prevalence of *T. parva* were compared using a chi-square test to test the significance of the differences in tick infestation and prevalence of *T. parva* between strains, dipping regimes, and seasons. Analysis of variance (ANOVA) was used to analyze the data on tick count to assess the statistical significance of differences. The three effects assessed were strain, dipping regime and season. All results were considered significant at p<0.05.

RESULTS

Ticks infestation among Sukuma and Tarime cattle strains

Three tick genera, Amblyomma genus, Boophilus sub-genus of *Rhipicephalus* genus and other species of *Rhipicephalus* were identified in the study area (Tables 1 and 2). *Rhipicephalus* genus accounted for 98.2% of the total ticks whereas *Boophilus* sub-genus of *Rhipicephalus* genus and Amblyomma constituted 1.3 and 15.5%, respectively. Significant differences were observed between Tarime and Sukuma cattle in terms of tick counts (p<0.05). Tarime cattle had relatively higher (11.8 ± 0.9) number of ticks per animal compared to Sukuma cattle (7.3 ± 0.9). However, dipping frequency of once every 2 or 3 weeks did not reveal significant differences in tick burden in both strains in both seasons. As expected non-dipped cattle had the highest (14.7 ± 1.1) tick burden compared to cattle dipped at either intervals (p<0.05). When tick infestations on the two cattle strains were compared during between the seasons, higher tick
Table 1. Factors influencing tick burden in Sukuma and Tarime cattle.

<table>
<thead>
<tr>
<th>Categories</th>
<th>Level</th>
<th>Whole body Tick count (Mean ± SE)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dipping frequency</td>
<td>Dipping after 2 weeks</td>
<td>5.8±1.2</td>
<td>0.0001</td>
</tr>
<tr>
<td></td>
<td>Dipping after 3 weeks</td>
<td>8.1±1.2</td>
<td></td>
</tr>
<tr>
<td></td>
<td>No dipping</td>
<td>14.7±1.1</td>
<td></td>
</tr>
<tr>
<td>Strain</td>
<td>Sukuma</td>
<td>7.3±0.6</td>
<td>0.0013</td>
</tr>
<tr>
<td></td>
<td>Tarime</td>
<td>11.8±0.8</td>
<td></td>
</tr>
<tr>
<td>Season</td>
<td>Dry season</td>
<td>9.1±0.9</td>
<td>0.5368</td>
</tr>
<tr>
<td></td>
<td>Wet season</td>
<td>11.9±0.8</td>
<td></td>
</tr>
</tbody>
</table>

*Means bearing different superscripts within column in each category are significant difference (p<0.05).

Table 2. Tick genera and confirmed ECF cases in Sukuma and Tarime cattle.

<table>
<thead>
<tr>
<th>Strain</th>
<th>N</th>
<th>Rhipicephalus (%)</th>
<th>Boophilus (%)</th>
<th>Amblyomma (%)</th>
<th>ECF cases (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sukuma</td>
<td>25</td>
<td>68.1</td>
<td>16.1</td>
<td>15.6</td>
<td>20</td>
</tr>
<tr>
<td>Tarime</td>
<td>25</td>
<td>68.3</td>
<td>16.5</td>
<td>15.2</td>
<td>0</td>
</tr>
<tr>
<td>Overall</td>
<td>50</td>
<td>68.2</td>
<td>16.3</td>
<td>15.5</td>
<td>10</td>
</tr>
</tbody>
</table>

Table 3. Percentage of *T. parva* PCR positive samples in Sukuma and Tarime cattle at different sampling periods.

<table>
<thead>
<tr>
<th>Sampling period</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tarime PCR positive results (n=25)</td>
<td>24</td>
<td>32</td>
<td>36</td>
<td>40</td>
<td>40</td>
<td>44</td>
<td>44</td>
<td>44</td>
<td>38.5</td>
</tr>
<tr>
<td>Sukuma PCR positive results (n=25)</td>
<td>32</td>
<td>32</td>
<td>32</td>
<td>36</td>
<td>40</td>
<td>40</td>
<td>40</td>
<td>40</td>
<td>38.5</td>
</tr>
<tr>
<td>Overall PCR positive results (n=50)</td>
<td>28</td>
<td>32</td>
<td>34</td>
<td>38</td>
<td>40</td>
<td>42</td>
<td>48</td>
<td>48</td>
<td>38.25</td>
</tr>
</tbody>
</table>

Counts were observed during the wet season but the difference between wet and dry seasons was not statistically significant (p>0.05).

**Clinical ECF manifestation and PCV among Sukuma and Tarime cattle strains**

During the entire experimental period, animals were monitored for clinical signs of ECF. Although no mortality was recorded, however, five animals of Sukuma cattle (20%) showed clinical signs of ECF infection as confirmed by microscopy of blood and lymph smears. All the ECF cases occurred in group 3 animals (Sukuma control group) during the wet season and the sick animals recovered after treatment with buparvaquone (2.5 mg/kg). No cattle from the Tarime strain suffered from ECF during the study period (Table 2).

Significant differences were also observed between Tarime and Sukuma cattle in terms of packed cell volume (p<0.05). Tarime cattle had relatively lower (29.9 ± 0.2) PCV mean per animal compared to Sukuma cattle (31.9 ± 0.4). However, dipping frequency either once every 2 or 3 weeks did not lead to significant differences in PCV means in the strains. When PCV means were compared between the seasons, higher PCV means were observed during the dry season and the difference between wet and dry seasons was significant (p<0.05) for both strains. Clinically sick animals (ECF) had lower PCV below their respective group means but their PCV level was within the physiological range of 24 to 46 (Table 4).

**Prevalence of *T. parva* parasites among Sukuma and Tarime cattle strains**

The prevalence of *T. parva* parasites was determined at 3-week intervals from mid August 2014 to mid February 2015, a period representing dry and wet seasons. *T. parva* prevalence was found to be variable over different sampling periods as well as between strains (Table 3). Thus, regardless of dipping regime, *T. parva* parasite prevalence was 24% at the beginning of the study (sampling period 1) and increased to 44% six
Table 4. Influence of strain, dipping regime and season on prevalence of T. parva parasites, antibody percentage positivity and packed cell volume in Sukuma and Tarime cattle.

<table>
<thead>
<tr>
<th>Categories</th>
<th>Strain</th>
<th>Dipping regime</th>
<th>Season</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Sukuma N=25</td>
<td>Tarime N=25</td>
<td>N=32</td>
<td>N=18</td>
</tr>
<tr>
<td>ELISA (PP means)</td>
<td>30.3±0.9²</td>
<td>34.9±1.1²</td>
<td>0.000</td>
<td>0.944</td>
</tr>
<tr>
<td>T. parva PCR percent (%)</td>
<td>38.5³</td>
<td>38²</td>
<td>0.836</td>
<td>0.539</td>
</tr>
<tr>
<td>Packed Cell Volume (means)</td>
<td>31.9±0.4²</td>
<td>29.6±0.7²</td>
<td>0.001</td>
<td>0.439</td>
</tr>
</tbody>
</table>

Means bearing different superscripts within rows in each category are significantly different (p<0.05).

months later (sampling period 8) for Tarime cattle, whereas it was 32 and 48%, respectively for Sukuma cattle during the two sampling periods. Between the two strains, T. parva prevalence was 28 and 46% at the beginning and at the end of the study period, respectively. Mean prevalence for the entire study period was 38.2% in the study area. However, the difference in parasite prevalence between Tarime and Sukuma cattle strains was not statistically significant.

Prevalence of antibody percentage positivity in Sukuma and Tarime cattle strains

Levels of specific antibodies to T. parva detected in the Tarime and Sukuma cattle strains are shown in Table 4.

All cattle regardless of strain were positive for T. parva antibodies. Tarime cattle maintained significantly higher antibody percent positivity compared to Sukuma cattle by 13% (p<0.05). This was further demonstrated when cattle from the two strains were clustered into seropositivity categories (Figure 1). Thus, higher proportion of Sukuma strain cattle clustered to lower antibody PP (20 to 40) whereas most Tarime strain cattle fell into the higher antibody cluster PP (41 to 60). None of the cattle displayed antibody PP above 60%. Figure 1 depicts the distribution of cattle by strain across seropositivity categories. Interestingly, only a few Sukuma cattle showed antibody PP above 41% whereas Tarime cattle were evenly distributed across the seropositivity categories.

DISCUSSION

This study confirms anecdotal information regarding tolerance of Tarime cattle to ECF as compared to Sukuma cattle kept in the same lake zone of Tanzania. This conclusion is based on the clear findings emanating from this study. Firstly, our study clearly indicated that Tarime cattle carried significantly higher tick burdens than Sukuma cattle. Most (> 68%) of the ticks found on the cattle of both strains were of Rhipicephalus appendiculatus species, vectors of T. parva, the causative agent of ECF. Furthermore, none of the Tarime cattle showed signs of clinical ECF whereas signs were evident in 20% of the Sukuma cattle. These findings provide the first testimony to hitherto farmers' assertions that Tarime cattle possess tolerance to both ticks and clinical ECF. Farmers keeping Tarime cattle do not dip their cattle frequently; however, animals of this strain rarely show clinical ECF signs as compared to cattle of other strains commonly grazing in the same area (Laisser et al., 2014). Ability of the Tarime cattle to carry more ticks and suffer less ECF may, therefore, be ascribed to an inherent feature of this strain of cattle, which has a small body size and seem to tolerate harsh local environmental conditions, including pressure from tick infestation. Our result on tick burdens and incidence of ECF in Tarime cattle concur with the findings by Taylor (2006), who reported a significant negative correlation between animal body weight and tick counts. Animals with an average body weight below 250 kg had 42% more ticks compared to animals with higher body weight.

Also the high tick infestation observed on non-dipped cattle agrees with the report by Mathee et al. (1997) who showed that regardless of breed, if ticks are not controlled, animals will always carry more ticks throughout the year and if the animals are not tolerant, there will be high mortality. However, the number of ticks per animal observed in this study differs from that observed by Laisser et al. (2014) in Serengeti and Tarime districts of
Mara region. The difference observed may be attributed by several factors, which include management practices and animal factors in these ecological areas. Generally, our findings demonstrate that animals which are adapted to a given environment usually carry fewer ticks as previously reported by Wambura et al. (1998).

Secondly, when we compared Tarime and Sukuma cattle in terms of prevalence of *T. parva* parasites, our study has revealed that Tarime cattle had slightly lower parasite prevalence, although the difference from Sukuma cattle was not statistically significant. These results agree with previous findings by Gachohi et al. (2012) and Marufu (2008) who reported that when different breeds of cattle are kept together in *T. parva* endemic area, less resistant cattle tend to acquire more parasites. Lower *T. parva* parasite prevalence observed in Tarime cattle may indicate a higher ability of these animals to clear *T. parva* parasites compared to more susceptible cattle breeds. Our results have further shown that parasitemia increased gradually in both strains from the beginning of the study (period 1) to the end of the study (period 8) by almost 150 to 180%. These data provide an indication of inherent *T. parva* carrier state of cattle of both strains and that the carrier state increased incrementally throughout the study period. Our data also show that cattle of both strains picked up the infection gradually since they co-grazed on same pastures, however, it is interesting to point out that Tarime cattle did not develop clinical signs to the extent shown by Sukuma cattle. The Lake zone in Tanzania is an endemic area for ECF and as such it would be expected that cattle kept in this area are under constant exposure to the *T. parva* parasites. Zebu cattle usually acquire immunity to *T. parva* following recovery after primary ECF infection. It is also evidently reported that natural tick challenge incrementally boosts the immunity acquired by zebu cattle kept in endemic areas (Kazungu et al., 2015), therefore, resistant individuals or breeds are likely to attain higher immunity compared to susceptible breeds. Our result on the differences between Tarime and Sukuma cattle strains further confirms the preference of farmers in the Lake Zone for Tarime cattle than other strains of cattle (Laiser et al., 2015). As such, farmers’ knowledge on unique attributes of their cattle breeds is usually passed on through generations (FAO/IFB, 2000) and thus our result has demonstrated evidence to farmers’ beliefs and paves way to designing improvement programmes for Tarime cattle for not only ECF tolerance.

Figure 1. Comparison of cattle strains by antibody percentage positivity categories.
but also in terms of productivity. Thirdly, this study has revealed a broad range of antibody percent positivity levels for both strains, but the majority of the Tarime cattle clustered to higher antibody PP categories. Besides being an epidemiological indicator, the level of antibodies in animals in an endemic area may also indicate a measure of resistance to infection pressure in the study area. It is possible that the higher level of specific antibodies reflects the response of resistant individuals in a *T. parva* endemic area under constant natural tick challenge. Probably, these findings could also suggest the ability of Tarime cattle to mount a stronger antibody response, which primarily play a crucial role in neutralization of *T. parva* sporozoites in early stages before ECF infection. This is also supported by our finding of lesser clinical ECF infections in Tarime compared to Sukuma cattle. The findings of lower parasite prevalence, but higher antibody levels shown by Tarime cattle suggest a potential state of endemic stability, whereby cattle develop a carrier state of *T. parva* parasites without clinical disease (Norval et al., 1992; Deen et al., 1993; Perry and Young, 1995; Malovele et al., 2003). Our result concurs with Martins et al. (2010) who reported a significant negative association between antibody percent positivity levels in cattle and incidences of ECF cases.

This study has revealed, high seroprevalence which was indicated for both, Sukuma and Tarime cattle strains. The 100% seroprevalence indicates that all study animals had previous exposure to *T. parva*, although differences existed between the strains and individual cattle in terms of level of antibodies produced. The high seroprevalence obtained in this study area further demonstrates the potential development of a state of endemic stability in the Lake zone, whereby the majority of cattle are expected to be carriers as previously shown by Kazungu et al. (2015). Dipping regime did not show any influence on seroprevalence results.

**Conclusion**

The present study has demonstrated differences in terms of tick burdens between Tarime and Sukuma cattle, whereby Tarime cattle carried significantly higher tick load. Tarime strain of cattle was also different from Sukuma cattle in terms of *T. parva* parasite prevalence and antibody percent positivity. The higher antibody levels but lower parasite prevalence shown by Tarime cattle suggests a higher proportion of resistant individuals in this strain, which may potentially support development of a *T. parva* carrier state. Our study provides evidence to farmers’ beliefs in the Lake zone of Tanzania regarding resistance of Tarime compared to Sukuma cattle. This paves a way to designing improvement programmes for the Tarime cattle for rational utilization.

**Conflict of interest**

The authors have not declared any conflict of interest.

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CHAPTER FIVE

5.0 PAPER IV

A Review on prevalence, control measure and tolerance of Tanzania Shorthorn Zebu cattle to East Coast fever in Tanzania

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A review on prevalence, control measure, and tolerance of Tanzania Shorthorn Zebu cattle to East Coast fever in Tanzania

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Abstract In Tanzania, control of East Coast fever (ECF) has predominantly relied on tick control using acaricides and chemotherapy, little on ECF vaccination, and very little on dissemination regarding animal immunization. In this paper, the prevalence, control measure, and tolerance of Tanzania Shorthorn Zebu (TSHZ) cattle to ECF are reviewed. In addition, the opportunities available for reducing the use of acaricides for the benefit of the farmers in terms of reduction of costs of purchasing acaricides and environmental pollution are described. The tick distribution and epidemiological factors for ECF such as the agro-ecological zones (AEZ), livestock production systems (LPS), strain, and age of the animals are also described. These factors influence the epidemiology of ECF and the distribution of TSHZ strains in different geographic locations of Tanzania. We have further showed that there is a tendency of farmers to select among the strains of TSHZ for animals which can tolerate ticks and ECF and cross-breed them with their local strains with the aim of benefiting from the inherent characteristics of the most tolerant strains. Generally, many strains of TSHZ cattle are tolerant to tick infestation and ECF infection and can be bred to respond to the needs of the people. In this review paper, we recommend that in future, ECF epidemiological studies should account for factors such as livestock production systems, agro-climate, breed of animal, tick control strategy, and the dynamic interactions between them. In conclusion, we have demonstrated that an integrated control method involving use of acaricides, immunization, and ECF-tolerant/resistant animals is required.

Keywords East Coast fever · Tanzania shorthorn zebu · *Theileria parva* infection · Tolerance

Introduction

The total number of cattle raised in Tanzania is 25.8 million and most of them are indigenous cattle belonging to the Tanzania Shorthorn Zebu (TSHZ). The TSHZ has several strains which include Iringa Red, Masaai, Singida White, Gogo, Chaga, Pare, and Mbulu (Manga et al. 2001; Das and Mkombe 2003). These animals are used for various functions, especially for provision of animal protein food (meat and milk), draft power as labor saving technology, manure for fertilizing crop fields, and biogas for electrification or cooking fuel which has a potential for reducing deforestation. Furthermore, they play important social functions in traditional societies, including payment of dowry and sacrifices in traditional ceremonies (MLFD 2015). They also provide employment and income for the majority of the smallholder...
farmers living in rural areas. The TSHZ animals usually survive and reproduce under harsh climatic, nutritional stress, and poor management conditions under the smallholder production systems (MLFD 2015).

East Coast fever (ECF) is a tick-borne disease (TBD) of cattle which is caused by a protozoan parasite namely *Theileria parva*. The parasite is transmitted by a three-host tick called *Rhipicephalus appendiculatus*, which have dropped from infected cattle during the preceding stage of the life cycle (Norval et al. 1992). In cyclopropagative and transient transmission, the *T. parva* parasite multiples and undergoes cyclical changes within two developmental stages (nymph and adult) of the vector. In Tanzania, ECF is the major cause of cattle deaths and costs the government a huge financial resource for its control. For example, the costs of acaricide application, which is the primary means of tick control, has been estimated to range between US$ 6 and US$ 36 per adult animal in the East African countries including Tanzania (Munjauw and McLeod 2003; Mugisha et al. 2005; Kivaria 2006). In addition, the disease prevents the introduction of more productive but ECF-susceptible exotic breeds of cattle, hence, hampering the development of the livestock sector considerably.

This review is based on the recognition that the application of acaricides as the most common method used to control ticks in the country has caused the development of tick resistance to most acaricides available in the market and has reduced these products’ useful lifetime due to improper use. Also, the improper use of acaricides results into the presence of chemical residues in meat, milk, and environmental pollution and this prompts the need for better monitoring of their application (Graf et al. 2004; Mugisha et al. 2005; Castro-Janer et al. 2010).

Like other indigenous African cattle, the TSHZ cattle are credited for their inherent ability to tolerate endemic diseases such as East Coast fever (Marufu 2008). As such, most livestock keepers do not apply acaricide to control ticks because they believe that their animals can tolerate ticks and TBDs. The TSHZ animals have survived for many years without or with minimum ticks and ECF control, yet they contributed significantly to the welfare of rural people, who accounts for 80% of the population in Tanzania. Farmers in one community understand the degree of tolerance of their animals to tick infestation and ECF infection compared to those of other communities (Laisser et al. 2015). This can be evidenced by a situation where one community hires or buys a bull from another community due to a belief that the strain of the animal bought is superior in terms of ticks and ECF tolerance compared to the one in their community (Chenyambahga et al. 2010). Although it is obvious that TSHZ animals can tolerate ticks and ECF compared to exotic breeds and there is increasing awareness on the subject, there are no compiled data to support the farmer’s belief and confirm this evidence for various strains of TSHZ. We had five specific objectives for this article;

1. To review the prevalence of ECF in various production systems in Tanzania
2. To highlight the epidemiological factors influencing the prevalence of ECF
3. To highlight the tick and ECF control methods in various production systems in Tanzania
4. To review the tolerance of African indigenous cattle to ticks and ECF infection
5. To review the mechanisms for tick and ECF resistance in cattle

Review methodology

A search of peer-reviewed publications on ECF, ticks, and ECF-tolerant cattle in Tanzania was conducted from comprehensive databases including PubMed, ScienceDirect, Swetswise, and CABI direct. The search was extended to available theses, conference proceedings, and project reports. Keywords were standardized across the databases to produce comparable searches and these were East Coast Fever, *Theileria parva*, *Rhipicephalus appendiculatus*, epidemiology, prevalence, incidence, cattle, tolerance, and Tanzania. References of all relevant articles were also searched to identify articles that could have been missed in the search. The search was conducted for all available years in each database. The keyword search produced 356 articles. We screened all the articles and then identified the most relevant ones for Tanzania.

Prevalence of ECF in various production systems in Tanzania

Up to date, several studies have been conducted and others are still underway regarding the incidence of ticks and ECF disease across different production systems in Tanzania. Table 1 shows the findings reported by different researchers from Tanzania on ticks, TBDs, ECF, and epidemiology of *T. parva* in pastoral societies. *R. appendiculatus* is widely distributed in Tanzania in those climatic and vegetational areas that are suitable for it (Fig. 1) and the most abundant tick species in those areas (Table 1). So far in large areas of Tanzania, this species of tick is either completely absent or has never been recorded. In Tanzania, there are several systems for livestock production and management which influence the prevalence of ECF. This is because in each production system, different measures are taken to control the vector ticks which are responsible for the transmission of parasites.
<table>
<thead>
<tr>
<th>Study N</th>
<th>Study</th>
<th>Strain targeted</th>
<th>Ecological zone</th>
<th>Research output</th>
<th>References</th>
</tr>
</thead>
</table>
| 1       | Baseline study on farmers knowledge and attitude ticks and tick-borne diseases | Sukuma and Tarime zebu           | The lake zone              | (i) Farmers have substantial knowledge on ticks and tick-borne disease (they even name ticks and TBDs on their vernacular language)  
(ii) Tick and tick-borne diseases are not important than other animal diseases and ECF is only a problem to calves  
(iii) Calves need to be protected than adult animals  
(iv) Dipping regime is irregular  
(i) Common tick genera are *Rhipicephalus appendiculatus*, *Boophilus*, *Amblyomma*, and *Hyalomma*  
(ii) Tick burden is high  
(iii) *Theileria parva* prevalence in the area is relatively high but *Theileria parva*-infected cattle do not show clinical signs of ECF  
(iv) Tarime cattle tolerate more tick burden than Sukuma zebu cattle  
(v) Tarime cattle unlike Sukuma cattle can be infected by *Theileria parva* but do not show ECF clinical signs  
(vi) Tarime cattle show high immune response to *Theileria parva* than Sukuma cattle | Laiosser et al. (2015) |
| 2       | A study on Tarime zebu cattle to investigate ticks species, tick burden, and prevalence of *Theileria parva* | Tarime zebu                      | The lake zone              | (i) Farmers have substantial knowledge on ticks and tick-borne disease (they even name ticks and TBDs on their vernacular language)  
(ii) Tick and tick-borne diseases are not important than other animal diseases and ECF is only a problem to calves  
(iii) Calves need to be protected than adult animals  
(iv) Dipping regime is irregular  
(i) Common tick genera are *Rhipicephalus appendiculatus*, *Boophilus*, *Amblyomma*, and *Hyalomma*  
(ii) Tick burden is high  
(iii) *Theileria parva* prevalence in the area is relatively high but *Theileria parva*-infected cattle do not show clinical signs of ECF  
(iv) Tarime cattle tolerate more tick burden than Sukuma zebu cattle  
(v) Tarime cattle unlike Sukuma cattle can be infected by *Theileria parva* but do not show ECF clinical signs  
(vi) Tarime cattle show high immune response to *Theileria parva* than Sukuma cattle | Laiosser et al. (2016) |
| 3       | A comparative study between Sukuma and Tarime zebu on ticks and *Theileria parva* to assess | Sukuma and Tarime zebu           | The lake zone              | (i) Farmers have substantial knowledge on ticks and tick-borne disease (they even name ticks and TBDs on their vernacular language)  
(ii) Tick and tick-borne diseases are not important than other animal diseases and ECF is only a problem to calves  
(iii) Calves need to be protected than adult animals  
(iv) Dipping regime is irregular  
(i) Common tick genera are *Rhipicephalus appendiculatus*, *Boophilus*, *Amblyomma*, and *Hyalomma*  
(ii) Tick burden is high  
(iii) *Theileria parva* prevalence in the area is relatively high but *Theileria parva*-infected cattle do not show clinical signs of ECF  
(iv) Tarime cattle tolerate more tick burden than Sukuma zebu cattle  
(v) Tarime cattle unlike Sukuma cattle can be infected by *Theileria parva* but do not show ECF clinical signs  
(vi) Tarime cattle show high immune response to *Theileria parva* than Sukuma cattle | Laiosser et al. (2016) |
| 4       | A study on farmers knowledge and attitude on ticks and tick-borne diseases | Tarime, Ankole, Boran, and Kenya shorthorn zebu | The lake zone of East Africa (Tanzania, Kenya, and Uganda) | (i) Farmers have substantial knowledge on ticks and tick-borne disease (they even name ticks and TBDs on their vernacular language)  
(ii) Tick and tick-borne diseases are not important than other animal diseases and ECF is only a problem to calves  
(iii) Calves need to be protected than adult animals  
(iv) Dipping regime is irregular  
(i) Common tick genera are *Rhipicephalus appendiculatus*, *Boophilus*, *Amblyomma*, and *Hyalomma*  
(ii) Tick burden is high  
(iii) *Theileria parva* prevalence in the area is relatively high but *Theileria parva*-infected cattle do not show clinical signs of ECF  
(iv) Tarime cattle tolerate more tick burden than Sukuma zebu cattle  
(v) Tarime cattle unlike Sukuma cattle can be infected by *Theileria parva* but do not show ECF clinical signs  
(vi) Tarime cattle show high immune response to *Theileria parva* than Sukuma cattle | Laiosser et al. (2016) |
| 5       | A study to determine seroprevalence of *Theileria parva*, knowledge, attitude, and practices of livestock keepers on East Coast fever (ECF) on indigenous cattle | Mixed strains, Maasai, and Sukuma zebu | Eastern zone---Kilosa district | (i) Improper dipping results in tick resistance  
(ii) Pastures are the source of ticks which infect animals due to communal grazing  
(iii) *Theileria parva* parasites are widely distributed throughout the two geographical sites  
(iv) The seroprevalence of *T. parva* is relatively high in Tanga and Iringa regions  
(v) Pasture-grazed animals are more likely to be seropositive than those that are zero-grazed | Tarimo (2013) |
| 6       | A study to investigate the cause of animal death | Mixed strains, Maasai, and Sukuma zebu | Kilosa district            | (i) Improper dipping results in tick resistance  
(ii) Pastures are the source of ticks which infect animals due to communal grazing  
(iii) *Theileria parva* parasites are widely distributed throughout the two geographical sites  
(iv) The seroprevalence of *T. parva* is relatively high in Tanga and Iringa regions  
(v) Pasture-grazed animals are more likely to be seropositive than those that are zero-grazed | Kambarage (1995) |
| 7       | A study to determine the distribution and prevalence and to quantify risk factors for *Theileria parva* | Iringa Red and Masai zebu         | Northern and highland zone | (i) *Theileria parva* parasites are widely distributed throughout the two geographical sites  
(ii) The seroprevalence of *T. parva* is relatively high in Tanga and Iringa regions  
(iii) Pasture-grazed animals are more likely to be seropositive than those that are zero-grazed | Swai et al. (2007) |
| 8       | A study to determine prevalence of tick genera and prevalence of *Theileria parva* in adult ticks | Mixed strains                     | Northern zone              | (i) Common tick genera are *Rhipicephalus appendiculatus*, *Boophilus*, *Amblyomma*, and *Hyalomma*  
(ii) *Theileria parva* infection prevalence in adult R. appendiculatus is low  
(iii) Seroprevalence was significantly higher in vaccinated than unvaccinated cattle  
(iv) *Prevalence of T. parva* parasites is significantly higher in vaccinated than unvaccinated cattle | Swai et al. (2005) |
| 9       | A study to assess the incremental effect of natural tick challenge on the infection and treatment method-induced immunity against | Masai zebu cattle                  | Northern zone              | (i) Seroprevalence was significantly higher in vaccinated than unvaccinated cattle  
(ii) *Prevalence of T. parva* parasites is significantly higher in vaccinated than unvaccinated cattle | Karungu et al. (2015a) |
The livestock production systems in Tanzania mainly include pastoralism (traditional extensive livestock production system), agro-pastoralism (traditional crop-livestock production system), and ranching (commercial/insensitive livestock production system). These cattle production systems can be simply divided into extensive (low input) and intensive (high input) (MLFD 2011). About 97% of the Tanzanian cattle are reared under the traditional extensive livestock production system (MLFD 2015).

In the traditional extensive production system, which is the most predominant in Tanzania, indigenous cattle are the main type of animals kept (MLFD 2015). Since these animals are adapted to and selected for ability to withstand harsh environmental conditions and diseases, little efforts are taken by farmers to control ECF; hence, the prevalence of this disease is high (Ngowi et al. 2009). In the agro-pastoral production systems where both crop and extensive livestock production systems are practiced, cattle are herded to graze in different sites during dry and wet seasons, and in the backyard production system where cattle are kept around farmers’ homesteads, the prevalence of ECF is variable (Chenyamba et al. 2010). For example, when the animals are shifted to other places during dry season to search for pasture, they tend to pick infested ticks which will, in turn, increase the prevalence of ECF in their areas (Laisser et al. 2014). Tick infestation is a common problem in pastoral and agro-pastoral areas due to the fact that most livestock farmers do not control ecto-parasites through dipping or spraying. The prevalence of ECF observed in the agro-pastoral areas of the lake zone of Tanzania by Chenyamba et al. (2010) and Laisser et al. (2014) was 80 and 27.7%, respectively. In Tanga region where most of the farmers graze their cattle around their households or practice zero grazing, a very low prevalence of 2.7% has been reported (Swai et al. 2007). Moreover, the observed lower tick infestation and ECF infection in Tanga could be attributed to regular application of tick and tick-borne disease control methods.

Higher prevalence of *T. parva* has been reported by Kazungu et al. (2015) in pastoral areas of the Masai communities of Simanjiro district, northern Tanzania. According to Kazungu et al. (2015), apart from lack of tick control measures in pastoral areas, proximity to national parks and game reserves contributes to higher prevalence of *T. parva* in pastoral areas. Furthermore, the control measure practiced in an area has much influence on the prevalence of *T. parva*. Kazungu et al. (2015) reported a prevalence of 50.39 and 19.69% for *T. parva* infection in vaccinated and unvaccinated Masai cattle, respectively, in Simanjiro district. Another study done by Haji et al. (2014) in five villages (pastoral areas) of Monduli District namely Msawaki, Lake Manyara, Naitolia, Makuyuni, and Ngena revealed an overall *T. parva* prevalence of 12.5%. Komwihangelo et al. (2009) reported *T. parva* prevalence of 30% in Mpwapwa breed kept in the central zone of Tanzania under the semi-
Table 2  Tick-tolerant zebu in Africa

<table>
<thead>
<tr>
<th>S/N</th>
<th>Breed</th>
<th>Country</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Africkander</td>
<td>South Africa</td>
<td>Bonnem (1940) in Bonnem (1981)</td>
</tr>
<tr>
<td>2</td>
<td>Brahman</td>
<td>South Africa</td>
<td>Rechav (1987), Rechav et al. (1990)</td>
</tr>
<tr>
<td>3</td>
<td>Bonamara</td>
<td>South Africa</td>
<td>Scholtz et al. (1991)</td>
</tr>
<tr>
<td>4</td>
<td>Ngumis-Nkoni (Sanga)</td>
<td>South Africa</td>
<td>Scholtz et al. (1991)</td>
</tr>
<tr>
<td>5</td>
<td>Boran</td>
<td>Kenya</td>
<td>de Castro (1991)</td>
</tr>
<tr>
<td>6</td>
<td>East African Zebu</td>
<td>Kenya</td>
<td>Laff et al. (de Castro 1991)</td>
</tr>
<tr>
<td>7</td>
<td>Kisanja</td>
<td>Sudan</td>
<td>Laff (1984)</td>
</tr>
<tr>
<td>8</td>
<td>Butana</td>
<td>Sudan</td>
<td>Laff (1984)</td>
</tr>
<tr>
<td>11</td>
<td>Nkoni (Sanga)</td>
<td>Zimbabwe</td>
<td>Norval et al. (1988)</td>
</tr>
<tr>
<td>12</td>
<td>Tanine zebu</td>
<td>Tanzania</td>
<td>Laisser et al. (2016)</td>
</tr>
</tbody>
</table>

Source: adapted from de Castro (1991) and Laisser et al. (Laisser et al. 2016)

intensive system while Swai et al. (2007) found the prevalence of *T. parva* to be 77.5% in Iringa. Red zebu raised under the agro-pastoral production system in Iringa Rural and Kilolo districts in the southern highland zone of Tanzania. The lower *T. parva* prevalence in Mpyawpa cattle is because the breed is an improved breed kept by small-scale farmers who practice regular dipping/spraying with acaricide to control ticks.

Epidemiological factors influencing the prevalence of ECF

The epidemiology of ECF is largely influenced by varying environmental conditions such as agro-ecological zones, livestock production system, and animal breed and age (Gachohi et al. 2012). There are several factors that may influence the epidemiology of ECF disease, but the main one is the control measures against the transmitting vector of *T. parva* parasite, a tick of the genus *R. appendiculatus*. The use of acaricides to control tick infestation through dipping or spraying is the main method used in Tanzania. However, the acaricides used in this method are very expensive and acaricide application is uneconomical to most farmers (Kivaria 2006); hence, farmers tend to stop dipping/spraying their animals, use below recommended dilution, or apply acaricide irregularly (Laisser et al. 2014). In addition, many farmers do not apply acaricides properly due to lack of knowledge and skills of application, and this causes high tick infestation in cattle (Chenyambuga et al. 2010) and increases tick resistance to acaricide. This trend has led *T. parva* parasites to persist and hence high number of ECF cases for calves in traditional livestock production systems.

Change in climate is another factor influencing epidemiological of ECF. Tanzania has faced continuous climatic changes in various agro-ecological zones and this has affected the distribution of tick vectors. Climate change includes change in rainfall pattern and distribution, occurrence of drought, and low and high temperatures (Kimaro and Chibinga 2013). The change in these climatic factors influences the abundance or distribution of *T. parva* parasites in a given agro-ecological zone by affecting the survival of the parasite or the intermediate vector (Kimaro and Chibinga 2013). In the northern highlands of Tanzania (Kilimanjaro, Arusha, Usambara in Tanga region), *R. appendiculatus* is said to be numerous high during the rainy season; this is quite similar to the situation in the southern highlands which also have a relatively high number of *R. appendiculatus* ticks during the wet season (Iringa, Mbeya, and Ruika regions) (Chenyambuga et al. 2008).

Large numbers of ticks, mainly *R. appendiculatus*, are usually observed during the period between the end of rainy season and start of dry seasons. The amount of rainfall is the principal stimulus to *R. appendiculatus* activity (Laisser et al. 2016). The low number of tick counts in some of agro-pastoral communities could be related to the low number of traditional cattle stock and improved management aspect on tick control. The local cattle grazing in communal areas is the main source of ticks in most areas because of irregular dipping or absence of tick control in traditional communities (Chenyambuga et al. 2010; Laisser et al. 2014). However, the prevalence of *T. parva* in some agro-ecological zones is low due to the development of endemically stable condition (Kazungu et al. 2014). Most research findings show that under traditional management system, there is a condition of endemic stability due to repeated contact of the animals with parasite (Kivaria et al. 2012). Endemic stability here means presence of more animals with higher (>70%) serum antibodies to *T. parva* where no or few cases of ECF. In contrast where effective and intensive tick control is practiced, it induces a high state of endemic instability whereby animal serum antibody is less or equal to 30% and more animals succumb to ECF (Kivaria et al. 2012).
Tick and ECF control methods in various production systems in Tanzania

Tanzania has a tick control policy that emphasizes on controlling the spread of ticks from endemic zones to non-endemic zones and protecting infiltration of ticks from other neighboring countries through movement or exportation of cattle. The control strategy for this policy includes dipping regimes or spraying in all traditional and private livestock farms, construction of stock routes and grounds, and lastly conducting research for tick and tick-borne diseases with focus on immunology and tick ecology (MLFD 2015). Generally, the methods used for controlling ticks and tick-borne diseases such as ECF in Tanzania involve the use of acaricides for vector ticks and chemotherapy for sick animals as well as immunization of cattle by the infection and treatment method (ITM) to a small extent (Perry 1994; Museke et al. 2004; Oura et al. 2004). However, tick control methods differ depending on production system. These production systems include traditional extensive livestock production, traditional crop-livestock production, and intensive livestock production system.

Traditional extensive livestock production system

In this system, livestock keepers own many indigenous zebu cattle which are grazed together under free range or herded grazing management conditions in communal rangelands with minimal production input. Under this system, there is little or no tick control at all. However, T. parva infections occur in this system and result in little loss in productivity and/ or mortality. In early 1980s, the government of Tanzania constructed many dip tanks in pastoral and agro-pastoral communities. The dip tanks were run by the government for a short time before being handled to the societies who did not afford to run them due to cost and lack of knowledge (Swai et al. 2007). As a result, many dip tanks in rural areas were abandoned and become non-functional. Consequently, spray method became a common method of tick control among the livestock farmers in rural areas (Laisser et al. 2015). Even though this method is easy to apply, it is not a good method to control ticks in communal areas because some farmers do not use appropriate dilution dose of acaricides while others apply irregularly; this results in tick developing resistance. Furthermore, under a spray method, the acaricides do not reach to all parts of the animal; hence, not all ticks are killed (Laisser et al. 2014). It has also been reported that in the lake zone of Tanzania, livestock keepers do not spray their animals regularly and some dips/spray their animals only when they have money to buy the acaricide; others apply the acaricide only twice a year while others do not apply acaricide at all (Chenyumba et al. 2010; Laisser et al. 2015). In those areas, ticks are available throughout the year and ECF prevail in many animals kept in communal rangeland. Nevertheless, most pastoralists and agro-pastoralists have local knowledge and experience on how to treat ECF infection by burning the lymph node of sick animals using hot iron and administering oxytetracycline (OTC) injection (Chenyumba et al. 2010; Laisser et al. 2015).

Another ECF control method which has been introduced recently in Tanzania is ITM. This involves infecting cattle with a calibrated number of T. parva parasites and concurrently giving them an injection of a long acting OTC treatment. OTC slows down the multiplication of T. parva parasite and provides animals' immune system to fight against the disease. The vaccine used in Tanzania was the Muga cocktail from Kenya (Patel et al. 2016). This method has been introduced in Masihi community in Simanjiro district (Kazungu et al. 2015). The method seems to be cost-effective for the control of ECF and reduces significantly mortality rate in the immunized animals (Martins et al. 2010). Although ITM offers a valuable alternative for ECF control (Oura et al. 2004), its widespread application has faced many challenges. These include the requirement for cold chain mode of delivery to remote areas and high cost of the vaccine (up to US$10 per animal), which is unaffordable to most smallholder herders (Di Giulio et al. 2009). Furthermore, ITM does not completely eliminate the need for acaricide application due to the potential existence of other tick-borne diseases. The method is not widely used by farmers; hence, efforts are needed to educate...
the livestock-keeping communities to accept this method. In traditional livestock-keeping societies, anything involving payment for livestock disease control before the outcome of it is seen, the local communities usually do not accept, especially those with many animals. Most traditional livestock keepers do not see ticks or ECF as a burning issue because adult animals suffering from ECF do recover without treatment.

Traditional crop-livestock production system

Traditional crop-livestock system integrates livestock production with other subsistence farm enterprises, particularly traditional crop agriculture. The population of cattle in this system is smaller compared to the extensive system because of land limitation. Usually, farmers are forced to have few animals in order to be able to carry out other agricultural activities; usually, a small portion of land is left uncultivated to be used for grazing animals during crop-growing season. Alternatively, animals are herded away from the farm during cropping season and returned back during the dry/harvesting time. This farming system exists in the highland areas as well as Lake Victoria basin of Tanzania in which zebu cattle are predominantly kept.

In this system, tick and ECF control methods vary depending on management practices existing in a particular community. Moreover, the control of ticks varies from one farmer to another; some farmers control ticks regularly, others on irregular basis while others do not control at all. This situation results in high tick burden in most animals (Laisser et al. 2014). In some areas, for example in Serengeti, the local government imposes a bylaw forcing each farmer to dip or spray his/her animals at least once every 2 weeks. This has reduced tick burden to a large extent in animals grazed in communal rangelands but increased T. parva prevalence (Laisser et al. 2014). This observation agrees with Kivaria et al. (2012) who observed an increase of T. parva prevalence in areas where strict tick control regime was practiced compared to the areas with less tick control measure. Usually, crop-livestock farmers do not graze their animals together, instead, each farmer has his/her own land to graze his/her animals (Laisser et al. 2015). This grazing system tends to minimize ticks and ECF. Generally, under crop-livestock system, tick and ECF control is taken seriously compared to the extensive system. This is probably due to the fact that there are fewer animals to manage and farmers have economic power to purchase acaricides.

Intensive livestock production system

In Tanzania, commercial systems consist of beef or dairy cattle farms in which highly productive exotic breeds of cattle and their crosses with indigenous cattle are kept. In the context of ECF control, these systems are characterized by intensive weekly application of acaricide that leads to the disruption of T. parva transmission. This system has become less important in Tanzania because the majority of the large farms has collapsed or has been privatized. We could not find any observational study conducted in this system in Tanzania from the literature.

Alternative tick control methods

Breeding of tick-resistant cattle, pasture spelling, pasture burning, and planting some special grasses have also been considered as means for tick control. Gladney et al. (1974) concluded that insect control based on sterile males or genetic manipulations offers little promise while pheromone attractants could be useful for domestic pets or for ticks attached on specialized sites. Mbai et al. (2002) reported that farmers also used alternative methods such as used engine oil (12%), Jeyes fluid (24%), chickens (4%), and manual de-ticking (2%). Kaaya and Hassan (2000) reported that the use of entomopathogenic fungi to control ticks may reduce the frequency of chemical acaricide use and the need for treatment of tick-borne diseases. They also concluded that mycopesticides are safer for the environment than conventional acaricides. However, in Tanzania, this method does not exist and, hence, there is a need to explore the suitability of the method under farmers’ conditions.

Due to the disadvantages of chemical acaricide products, the adoption of alternative control methods can minimize such problems. Wharton (1983) briefly reviewed the alternative methods of tick control and concluded that the utilization of host resistance, while offering an attractive approach to tick control, raises many questions even with the relatively simple Boophilus microplus and Babesia association. Resistance is an acquired characteristic and each animal develops its own level of resistance in response to tick challenge; the level may be high (as in most zebu cattle) or low (as in most European cattle), but a wide range of resistance occurs in all breeds of cattle. It is irritable, and selection and breeding for tick resistance are possible not only in zebu × European breeds but also within European breeds. However, selection for resistance or culling for susceptibility at present is based on tick numbers surviving on cattle exposed either naturally or artificially to tick challenge. This makes obvious problems for the cattle producer who is concerned about the effects of these ticks on production. Wharton (1983) showed that the most logical method of alleviating tick predation would be to capitalize on host-parasite relationships that evolved in nature. Cattle have survived in Asia and Africa despite the prevalent of Babesia, Theileria, and their Boophilus and Rhipicephalus vectors. Host resistance, expressed by an animal’s ability to prevent the maturing of large numbers of ticks, and disease immunity are survival mechanisms for the host and for external and internal parasites. The problem is not only to utilize
these attributes but also to increase productivity. Resistance to *Boophilus microplus* is associated primarily with zebu (*Bos indicus*) cattle (Bock et al. 1997). Considerable progress has been made in developing resistant *B. indicus × B. taurus* beef and dairy cattle that limit the effects of ticks while retaining high productivity (Hayman 1974; Turner 1975). Moreover, improvement of the nutritive value of pasture would allow cattle to develop a better resistance to tick infestation (MLFD 2015).

**African cattle breeds tolerant to ticks and ECF**

Resistance to tick infestation varies among individuals and breeds of cattle. It is known that in many subtropical and semi-arid environments in Africa, indigenous breeds are highly resistant to ticks and ECF (Norval et al. 1991). The phenomena of host resistance to ticks and enzootic stability to tick-borne diseases are well documented by Perry et al. (1985) and Latif and Pegram (1992). The ability of cattle to resist ticks and tick-borne diseases such as therioesthesia depends on the strength of their body immunity to respond against the infectious sporozoites, schizonts, and piroplasms parasitic antigens. Development of a stronger immunity system comes about through exposure to such diseases (Kivaria et al. 2012). Studies have shown that the advantages of indigenous cattle with regard to tolerance to ECF disease are results of profound exposure of the animals to *T. parva* parasite. The prolonged exposure to the parasite makes the animal develop immunity to the parasite, a condition known as carrier state (Kivaria et al. 2012). Furthermore, the ability to tolerate *T. parva* parasites by indigenous cattle can be artificially increased by vaccination (Kazungu et al. 2015). Once an animal gains immunity following exposure to infected ticks or through calves borne to immune dams, tick control frequency can be reduced so as to maintain endemic stability in a population of indigenous cattle (Laissner et al. 2016).

In East Africa and Southern Africa, Zebu and Sanga cattle and their crossbreeds are considered to be tolerant to ECF (Ndungu et al. 2005; Marufu 2008). Table 2 shows African cattle breeds that are resistant to ECF including Afrikander cattle, Brahman, Bonsmara, and Nguni-Nkoni (Sanga) all from South Africa, Boran, and East African Zebu found in Kenya, Karama, and Butana found mainly in Sudan. Others breeds are Boran and Horro from Ethiopia, Tarime zebu from Tanzania, and Nkoni/Sanga from Zimbabwe (de Castro 1991; Chenyambega et al. 2010; Laissner et al. 2015, 2016).

Wambura et al. (1998) also conducted a research to determine the difference in tick infestation and tick susceptibility between TSHZ and their crosses with exotic breed. The TSHZ strains were Meru, Mbulu, and Iringa Red and the crosses were Meru × Friesian and Iringa Red × Friesian. In their study, they found that TSHZ strains had less tick infestation compared to the crossbreeds. Furthermore, a high level of ECF infection in crossbred populations was clearly observed. These findings are in accordance with the findings reported by Perry (1994) that improved breeds including European taurines are susceptible to ECF.

In Tanzania, most traditional livestock keepers believe that their local cattle are tolerant to ticks and diseases. As a result of this belief, some strains are moved from their place of origin to other places (Chenyambuga et al. 2010; Mwambene et al. 2012). Although only one study has been done to compare two strains of TSHZ on tolerance to ticks and ECF (Laissner et al. 2016), all survey studies conducted show that most traditional livestock farmers claim that their cattle are tolerant to ticks or TBD diseases (Msanga et al. 2012; Mwambene et al. 2012; Laissner et al. 2015). The belief that TSHZ cattle are tolerant to ticks and ECF is supported by the evidence provided by findings in the literature which shows that zebu cattle survive without regular dipping/spraying and that infected animals do recover without treatment using veterinary drugs (Laissner et al. 2014; Laissner et al. 2015; Kazungu et al. 2015). This implies that the indigenous strain of zebu cattle in Tanzania possess genes which enable them to live and produce under high tick and ECF challenges. This is in agreement with Rege and Tiwah (1999) who stated that indigenous cattle are blessed with tick resistance and tolerance to vector-borne diseases and they perform better than exotic breeds under low-input conditions and climatic stresses, especially during times of drought. However, according to Chenyambuga et al. (2010) and Kiprono (2009), most livestock keepers in rural areas feel that ECF, which is caused by *T. parva*, has serious effects on calves than in adult cattle. Therefore, calves need to be protected by regular dipping/spraying as they are important as replacement stock.

**Mechanisms for tick and ECF resistance**

The immune responses at age related such that calves born to immune dams become more resistant than calves born to non-immune cows. Other factors that influence the ability of cattle to resist tick infection include heritability of the trait where cattle acquire the tolerance ability through inheritance, natural exposure to infected ticks, immunization by infection and treatment method, increasing tick challenge, and increased calf recovering rate after a disease challenge.

**Conclusion**

It is therefore recommended (1) to conduct further investigations on un-dipped herds of cattle to clarify the status of endemic stability to TBDs and the possibility of allowing a non-
dipping strategy to be extended much more widely in other agro-ecological zone, (2) to investigate the T. parva carrier state and establish whether or not resistant cattle constitute a practical danger to other breeds, (3) to define appropriate tick control regimes which integrate vaccination of the high-grade breed, and (4) to develop revised tick control strategies in an integrated package for each agro-ecological zone.

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Compliance with ethical standards Conflict of interest The authors declare that they have no conflict of interest.

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CHAPTER SIX

6.0 Research Output of the Thesis, General Conclusions and Recommendations

6.1 Research Output of the Thesis

The overall objective was to establish whether Tarime cattle are tolerant to tick infestation and ECF infection. The research outputs in this thesis are summarized and concluded as follows:

Livestock diseases are the most important constraints to cattle production in the Lake zone regions of Tanzania. These are followed by shortage of forages and water during the dry season and shortage of grazing land. Among the diseases, tick-borne diseases rank first in the agro-pastoral communities of Lake Zone. The livestock farmers in the Lake zone have substantial knowledge on ticks and TBDs signs. Despite the extensive knowledge, most livestock farmers do not apply acaricide regularly to control TBDs due to economic reason and the belief that their animals can tolerate ticks and TBDs. Their perception toward ECF is that, it is not more important than other livestock diseases. This is a reflection of the capability of their animals to tolerate the disease. It is therefore, important that tick control strategies should be harmonized with the indigenous knowledge of the livestock farmers on tolerance of local zebu strains to ticks and tick-borne diseases (Paper I).

Observations of Tarime cattle populations at two geographical locations revealed that; although management of animals can alter tick infestation and reduce animal ability to succumb to the parasites, the animals’ ability to withstand the diseases remains high. This is due to the fact that, all animals that tested positive for *T. parva* in both localities did not show any signs of illness, indicating that Tarime cattle can be infected with *T. parva* but
remain healthy and thus are potential carriers. Furthermore, the study has demonstrated that animals having high tick burden do not necessarily become infected with *T. parva*, the parasite that causes ECF. This is because some ticks do not harbour or do not carry the protozoan parasites causing ECF. High tick infestation and *T. parva* infection were found in Tarime cattle, but none of the animals examined showed either signs of ill-health as a result of tick infestation or clinical signs of ECF despite the fact that the DNA of *T. parva* was detected in 27.7% of the animals sampled. It can be said that the Tarime zebu can be infected with *T. parva* but do not develop signs of ECF; hence, they may become a source of infection to other less tolerant ecotypes of indigenous cattle in mixed herds (Paper II).

The present study found differences in terms of tick burden between Tarime and Sukuma strains, whereby Tarime cattle carried significantly higher tick load compared to Sukuma cattle. Tarime strain was also different from Sukuma strain in terms of *T. parva* parasite prevalence and antibody percent positivity. The higher antibody levels but lower *T. parva* prevalence shown by Tarime strain compared to Sukuma strain suggests that animals belonging to Tarime strain are relatively tolerant to ECF compared to Sukuma. The study provides evidence to farmers’ beliefs in the Lake zone of Tanzania regarding tolerance of Tarime strain compared to Sukuma strain. This paves a way for designing improvement programmes for the Tarime strain for sustainable utilization (Paper III).

The review study on the prevalence, control measure and tolerance of Tanzania Shorthorn Zebu cattle to East Coast fever in Tanzania revealed that ECF is not seen by most livestock keepers as the most serious disease compared to other TBDs. This is because TSHZ can be kept for long time with irregular or no tick control and manage to survive, despite the high tick burden and ECF infection rate. The review also shows that each society entrusts their strain for different reasons and they can compare their strain of
cattle with that of other society. Our study provides evidence for Tarime cattle from the Lake zone of Tanzania to be accepted by other livestock farmers due to its tolerance to ECF disease (Paper IV).

6.2 General Conclusion

It can be concluded from the present study that Tarime cattle tolerate high tick infestation and East Coast fever infection than the Sukuma cattle which are also kept in the Lake Zone. It can also be further concluded that, once per month dipping (using Dominex acaricide) of indigenous cattle found in the Lake Zone is satisfactory for tick and ECF control under traditional livestock production system.

6.3 Recommendations

Based on the findings obtained from this study, the following recommendations are made:

i. Further, research is required, especially on genomic related studies for Tarime cattle to understand the genetic uniqueness of the strain and the mechanisms or genes which make the strain be able to tolerate ECF infection.

ii. Community conservation programmes for the Tarime strain should be promoted.