

**Sokoine University of Agriculture**



**MSc Dissertation**

**Livestock Farmer's Awareness of Tick-Borne Diseases and Susceptibility Profile of Selected Cattle Ticks to Acaricides in Selected Areas of Eastern Tanzania**

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**May 2024**

**LIVESTOCK FARMER'S AWARENESS OF TICK-BORNE  
DISEASES, AND SUSCEPTIBILITY PROFILE OF SELECTED  
CATTLE TICKS TO ACARICIDES IN SELECTED AREAS OF  
EASTERN TANZANIA**

**A Dissertation Submitted in Partial Fulfilment of the Requirements  
for the Degree of Master of Science in Parasitology of Sokoine  
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## EXTENDED ABSTRACT

Ticks are arthropods that are responsible for the transmission of tick-borne diseases in cattle. The use of acaricides is the most preferred method for the control of ticks and tick-borne diseases in cattle. However, this method is challenged by ticks' resistance to different classes of acaricide. This study aimed to assess livestock farmers' awareness of tick-borne diseases (TBDs) and acaricide resistance of cattle ticks against frequently used acaricides in Eastern Tanzania. Assessment of farmers' awareness was conducted by administering a structured questionnaire to 129 individuals. To determine the species composition of ticks infesting cattle and assess of acaricide resistance of cattle ticks, field collection of ticks was performed whereby a total of 384 cattle were examined. An experimental design was adopted to evaluate the effectiveness of the selected acaricides against F1 tick larvae from collected field tick species. A judgmental or purposive sampling method was applied whereby a total of 952 ticks were collected and stored in plastic bottles filled with 70% ethanol for tick identification, while female engorged ticks were placed onto 50 ml perforated plastic containers with moisture cotton wool inside to allow egg laying and subsequently obtain F1 tick larvae that were tested for acaricides resistance. Identification of ticks was achieved using prescribed macroscopic and microscopic morphological features. In-vitro acaricide susceptibility test using Larval Packet Test (LPT) was performed to assess acaricides resistance of eight commonly used acaricides with active ingredients; Amitraz (AM), Organophosphates (OPs); chlorfenvinphos & chlorpyrifos and Synthetic Pyrethroid (SP); alphacypermethrin and cypermethrin.

The data obtained were organized and analyzed for tick species occurrence and mean percentage mortalities by using a Microsoft Excel spreadsheet and the IBM Statistical Package for Social Science

version 20. Generalized linear models (GLMs) with univariate analysis were employed to compare the performance of acaricide resistance of collected tick strains at 95 % confidence and p-value  $\leq 0.05$ . The findings indicate that the general understanding of livestock farmers on tick and TBDs was low, as only 27.9%, 28.7%, and 29.5% of the respondents identified *Rhipicephalus (Bo) spp*, *Amblyomma*, and *Rhipicephalus* ticks species respectively. A proportion of respondents understood the specific tick-borne disease names such as East Coast Fever (17.1%), *Babesiosis* (7.0%), and *Anaplasmosis* (6.2%). Hand spray application of acaricide was the most preferred method (89.1%) for the control of ticks and TBDs by livestock farmers. About half of respondents (48.8%) applied the acaricide once per week, followed by those practicing irregularly (39.6%). The majority of the livestock keepers (69.8%) do prepare acaricide at higher concentrations than that recommended by manufacturers. Among the tick species found in the study areas, *R. (Bo) microplus* was the most predominant species (57.9%) followed by *Amblyomma variegatum* (25.9%), *R (Bo) decoloratus* (6.8%), *Hyalomma rufipes* (4.3%) while the least species were *R. everts* (3.8%) and *R. appendiculatus* (1.1%). On the other hand, acaricide susceptibility status appeared to be low for acaricide C (Cypermethrin 15g/l) and acaricide D (Alphacypermethrin 100 g/l) by 84.60% and 85.70% respectively followed by emergence resistance for acaricide E (Alphacypermethrin 10g/l) and acaricide B (Alphacypermethrin 100g/l + Chlorpyrifos 100g/l) by 91.30% and 93.90% while acaricide H (Amitraz 125g/l), acaricide A (Amitraz 12.5%), acaricide F (Chlorpyrifos 500g/l + Cypermethrin 50g/l) and acaricide G (Chlorfenvinphos 300g/l) showed no resistance as exhibited by 97.80%, 97.90% 99.80% and 99.90% percentage mortality

respectively. Acaricide malpractices observed including the use of wrong dilution rates, use of single acaricide over prolonged time, and uncontrolled application intervals are some of the factors which might contribute to acaricide failure therefore more effort should be directed at educating the livestock farmers on ticks and TBDs control practices using livestock field officers and frequent training on good tick and TBDs control practices.

**Keywords.** Awareness, control practices, Acaricide, Resistance status, Ticks, Tick-borne diseases

## IKISIRI KUU

Kupe ni wadudu ambao wanahusika na maambukizi ya magonjwa yanayoenezwa na kupe kwa ng'ombe. Matumizi ya viuatilifu ndiyo njia inayopendekezwa zaidi ya kudhibiti kupe na magonjwa yanayoenezwa na kupe kwa ng'ombe. Hata hivyo, njia hii inakabiliwa na upinzani wa kupe kwa aina mbalimbali za viuatilifu. Madhumuni ya utafiti huu yalikuwa ni kutathmini uelewa wa wafugaji kuhusu magonjwa yaenezwayo na kupe na upinzani wa kupe dhidi ya viuatilifu vinavyotumika mara kwa mara Mashariki mwa Tanzania. Jumla ya wafugaji 129 walihusika katika tathmini hii ya ufahamu wa wakulima dhidi ya kupe wa ng'ombe na magonjwa wanayoeneza. Kufaham aina ya kupe wanaovamia ng'ombe na tathmini ya upinzani wa kupe dhidi ya viuatilifu, ukusanyaji wa kupe ulifanyika ambapo zaidi ya ng'ombe 384 walichunguzwa kutoka katika kaya 129. Majaribio ya kutathmini ufanisi wa viuatilifu vilivyochaguliwa dhidi ya mabuu (larvae) wa kupe yalifanyika kwa kutumia aina nne za kupe zilizokusanywa. Kupe 952 zilikusanywa na kuhifadhiwa kwenye chupa za plastiki zilizojazwa ethanol 70% kwa utambuzi wa kupe, wakati kupe wenye mimba waliwekwa kwenye vyombo vya plastiki (petridish) na kuwekwa kwenye box lililotobolewa lenye pamba iliyotiwa unyevu ili kuwezesha upatikanaji wa hewa na mazingira safi kwa ajili ya kupe kutaga mayai na hatimaye kupata mabuu F1 (larvae) wa kupe ambao walijaribiwa namna wanavyostahimili dhidi ya aina nane ya viuatilifu vilivyotumika.

Majaribio ya kutambua ukinzani wa kupe dhidi ya viuatilifu ulifanyika kwa kutumia njia ya Larval Packet Test (LPT). Viuatilifu nane vyenye viambato hai; Amitraz (AM), Organophosphates (OPs); chlofenvinphos & chlorpyrifos na Synthetic Pyrethroid (SP); alphacypermetrin na cypermetrin. Taarifa iliyopatikana ilipangwa na kuchambuliwa kwa kutumia Microsoft excel na Kifurushi cha Takwimu cha IBM SPSS cha

toleo la 20 la Sayansi ya Jamii. Pia njia ya Generalized linear models (GLMs) ilitumika ili kulinganisha utendaji wa upinzani wa viuatilifu dhidi ya aina za kupe zilizokusanywa.

Matokeo ya utafiti yanaonesha kuwa, uelewa wa jumla wa wafugaji kuhusu kupe na magonjwa yanayoenezwa na Kupe ulikuwa mdogo, kwani ni asilimia 27.9 walitambua kupe wa blue (R. (Boophilus) spp) , 28.7 kupe wenye rangi mgongoni (Amblyoma), na 29.5% kupe wekundu (Rhipicephalus spp). Idadi ya waliohojiwa walielewa majina mahususi ya magonjwa yanayoenezwa na kupe kama vile Ndigana kali (17.1%), Ugonjwa wa kukojoa damu (7.0%) na Ndigana baridi (6.2%). Njia iliyotumiwa na wafugaji wengi kuweka kiuatilifu kwa ng'ombe ni kunyunyizia kwa mkono (hand sprayer)(89.1%). Takriban nusu ya waliohojiwa (48.8%) walitumia kiuatilifu mara moja kwa wiki, ikifuatiwa na wale ambao hawakuwa na mda maalum wa kutumia kiuatilifu (39.6%). Wafugaji walio wengi (69.8%) hutayarisha viuatilifu katika viwango vya juu kuliko inavyopendekezwa na watengenezaji.

Miongoni mwa aina za kupe zilizopatikana katika maeneo ya utafiti, R. (Bo) microplus ilitawala zaidi kwa 57.9% ikifuatiwa na Amblyomma variegatum 25.9%, R (Bo) decoloratus 6.8%, Hyalomma rufipes 4.3% huku aina chache zaidi zilikuwa R. everts 3.8% na R. appendiculatus 1.1%. Kwa upande mwingine, ukinzani dhidi ya viuatilifu ilionekana kuwa ya juu kwa kiuatilifu C (Cypermethrin 15g/l) kwa 84.60% na kiuatilifu D (Alphacypermethrin 100 g/l) kwa 85.70%. ikifuatiwa na ukinzani wa kiuatilifu E (Alphacypermethrin 100 g/l) na kiuatilifu B (Alphacypermethrin100g/l + Chlorpyriphos 100g/l) kwa 91.30% na 93.90% mtawalia. Hata hivyo kiuatilifu H (Amitraz 125g/l), kiuatilifu A (Amitraz 12.5%), kiuatilifu F (Chlorpyriphos 500g/l + Cypermethrin 50g/l) na kiuatilifu G (Chlorfenvinphos 300g/l) hazikuonyesha ukinzani kama ilivyoonyeshwa kwa asilimia 97.80, 97.90% 99.80% na asilimia 99.90 ya vifo mtawalia.

Matumizi mabaya ya viuatilifu ni pamoja na matumizi ya viwango vibaya vya kuyeyusha kiuatilifu katika maji, matumizi ya kiuatilifu kimoja kwa muda mrefu na kutokuwa na mda maalum wa kuogesha mifugo ni baadhi ya mambo ambayo yanaweza kuchangia kushindwa kwa viuatilifu hivyo. Juhudi zaidi zinapaswa kuelekezwa katika kuwaelimisha wafugaji kuhusu kupe na kanuni za udhibiti wa magonjwa yanayoenezwa na Kupe kwa kutumia maafisa mifugo na mafunzo ya mara kwa mara juu ya mbinu bora za udhibiti wa kupe na magojwa yanayoenezwa na kupe.

**Neno kuu;** Ufahamu, mbinu za udhibiti, Viuatilifu, Hali ya Upinzani wa kupe dhidi ya viuatilifu, Kupe, magonjwa yanayoenezwa na Kupe,

## DECLARATION

I, Mwita Marwa Wambura do at this moment declare to the Senate of Sokoine University of Agriculture that this dissertation is my original work and that to the best of my knowledge, it has neither been submitted for an MSc degree award in any other institution.



Mwita Marwa Wambura  
(MSc. Candidate)

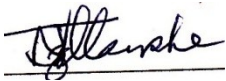
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10 / 05 / 2024  
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This thesis is dedicated foremost to God Almighty for His mercy and grace in my life, especially during two years at school where I succeeded in passing through many difficult challenges. I would also like to dedicate it to my late parents Mr. and Mrs. Mwitā M. Mwihechi for their invaluable ethical advice and care shown to me during their lifetime until their demise. To my dear Wife and our sons and daughter, their advice and guidance laid a foundation for the achievement of this work.

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#### **LIST OF ABBREVIATIONS**

- BOD Bio-Oxygen Demand (incubator)  
DD Discriminatory Dose

FAO	Food and Agriculture Organization
GDP	National Gross Domestic Product.
GLMs	Generalized Linear Models
LPT	Larval Packet Test
OPs	Organophosphate (acaricides)
SPs	Synthetic Pyrethroids (insecticides)
TBDs	Ticks and tick-borne diseases.
TPRI	Tropical Pesticides Research Institute

## CHAPTER ONE

### 1.0 GENERAL INTRODUCTION

#### 1.1 Background Information

Ticks and Tick-borne diseases can affect the health condition of an animal either directly or indirectly resulting in major losses in animal products such as eggs, meat, milk, and leather as well as the death of the affected animals. The direct effects are associated with the damage produced by the ticks during blood feeding on their hosts through injection of toxins which can cause paralysis and damage of hides, whereas the indirect effects are linked to the disease spread by the ticks such as East Coast fever, *Babesiosis*, *Cowdriosis* (heartwater) and *Anaplasmosis* as well as the treatment and control costs (Oscar and Cristian, 2018). Ticks and tick-borne diseases (TBDs) are the most prevalent and they cause the greatest economic barrier to livestock production in most countries (Rubaire-Akiiki *et al.*, 2004; Okuthe and Buyu 2006; Swai *et al.*, 2005).

Torr *et al.*, 2002 and Nagagi *et al.*, 2020, observed that there are many factors including different parasites and diseases that upset animal health and their productivity as main constraints in the growth and well-being of societies. In Tanzania, almost 80% of the cattle herds are at risk of being affected by TBDs every year, while the direct economic losses are likely to be US\$ 248 million of animals per year (Kivaria, 2006).

#### 1.2 Ticks and their classification

The ticks are insects that are found worldwide in tropical and subtropical regions. The hard ticks require blood meal from either wild or domestic animal hosts for survival and reproduction and are rarely encountered by humans. Some ticks are a nuisance by irritating and

even paralysis of the animal, weight loss, anemia, and even death in case of severe infestations due to high consumption of blood (Ahmad *et al.*, 2019). Ticks and tick-borne diseases are the cause of main economic losses through upsetting animal and human wellbeing. However, ticks are said to be the second vector of human diseases after mosquitoes (Lovis *et al.*, 2011).

Ticks are found in two main families, Argasidae which are called soft ticks due to lack of hard cover. The example of this family includes genera *Argas*, *Ornithodoros*, and *Otobius* (Jongejan and Uilenberg, 2004). The other family is Ixodidae; known as hard ticks due to the presence of scutum, which shields the whole dorsal surface of the adult male and only a small area in the larva, the nymph, and adult female. The Ixodidae family contains many genera including *Ixodes* which is the most prevalent genus containing 217 species. Other important genera include *Haemaphysalis*, *Dermacentor*, *Hyalomma*, *Rhipicephalus* (synonym *Boophilus*), and *Amblyomma* (synonym *Aponomma*) (Taylor *et al.*, 2016).

### **1.3 Tick life cycle, Transmission of Tick and TBDs**

Ticks usually prefer places on the body of animals where the skin is thin and short and have abundant blood supply, such as the inguinal region and external genitals. Ticks grow and develop best in hot and humid climates (Nejash and Tilahun, 2016).

Ticks normally go through four stages of life; egg, larva, nymph, and adult. Where the last three stages feed on the blood of animal hosts and can be infected with pathogens while feeding and transmitted to another animal host bitten by them Tick larval stage are very small in size hence difficult to see. Most of the larval tick's species are very similar in appearance with three pairs of legs when observed

microscopically by experts. Some tick-borne diseases can be transmitted through ovaries to the larvae that can also transmit the diseases to the animal host during their first blood feeding.

Nymphal stage ticks are characterized by having four pairs of legs and a clear dorsal shield (Scutum). Also, they have pale-colored abdomens. On the other hand, nymphs of many tick species look very similar causing a difficult identification process that needs only a professional person. Whereby, Adult ticks are characterized by four pairs of legs, visible mouthparts (shape and length), festoons, and a unique dorsal shield with either ornamentation or not. Ticks are classified into three categories depending on the number of hosts they involve during their life cycle. The three-tick life cycle includes One host tick: All three instars engorge on the same animal, the two ecdyses also taking place on the host e.g. *Boophilus annulatus*, *B. microplus*, and *Margaropus*. While two host ticks the larvae engorge and molt on the host and the nymphs drop off after having engorged. It molts on the ground and the imago seeks a new host e.g. *Rhipicephalus bursa*, *R. evertsi*. Where three host ticks require a different host for every instar, they drop off each time after having engorged and molt on the ground. e.g. *Ixodes ricinus*, *Ixodes*, *Hyalomma anatolicum anatolicum*, *Rhipicephalus appendiculatus* (Troughton and Levin, 2007).

Ticks and disease-causing organisms have co-evolved in balance with wildlife serving as hosts and reservoirs at the same time. Normally insecure circumstances occur only when these hosts or reservoirs interact with domestic animals, either by the movement of infested animals to non-infested regions or vice versa (Taylor *et al.*, 2016). On the other hand, Taylor *et al.* (2016) stated that ectoparasites have a role in the spread of zoonotic diseases to people, that cause high morbidity and mortality in the community.

TBDs of veterinary importance include *Theileriosis* such as East Coast fever and tropical bovine *theileriosis* that are transmitted by *R. appendiculatus* and *Hyalomma a. anatolicum* respectively. However, *Theileria* species such as *T. parva* and *T. annulata* are transcardially transmitted whereby the modal of transmission of *Babesia* spp is Transovarial transmission. However, *Babesiosis* caused by different *Babesia* species that are transmitted by tick species that are *Boophilus* spp, *Rhipicephalus* spp, and *Ixodes ricinus*. Other diseases include Bovine *anaplasmosis* caused by *Anaplasma marginale*, *A. centrale*–*Boophilus*, *Rhipicephalus*, *Ixodes*, and *Dermacentor* as well as Anthrax disease (Troughton and Levin, 2007).

#### **1.4 Acaricides and resistance status**

Acaricides are chemicals that kill an acarine group from members of the arachnid including mites and ticks (Abbas *et al.*, 2014). Conventional methods of killing ticks through the application of acaricides on the surface of animals are the most useful and economically feasible control measures for ticks and TBDs (Nagagi *et al.*, 2020). According to Kagaruki 1996; Nagagi *et al.*, 2020, acaricides are the control method applied through the surface of the animal either by plunge dipping, hand-washing, or spraying. The frequency of acaricide application depends on the acaricidal molecule but is assumed generally an average of twice per month. According to Vudriko *et al.* (2016); and Mbassa *et al* (2009), the practices of acaricide application are individually performed or through privately owned and commercially-run dip tanks. However, in Tanzania, due to cases of inappropriate use of acaricides, application methods, free acaricide market access, lack of periodic tick monitoring and increased acaricide pressure are among the factors that can accelerate the development of acaricide resistance (Vudriko *et al.*, 2016; Nagagi *et al.*, 2020)

On the other hand, Abbas *et al.*(2014) and Nagagi *et al.*(2020), discovered that the emergence of tick-resistant strains has been caused by selection pressure accelerated by the use of similar acaricides group for a prolonged period, lack of strategic alternation and monitoring of acaricide groups. In addition, Vudriko *et al.* (2016), noted that 90 % of tested tick populations were resistant against synthetic pyrethroids. Out of the tested ticks, 60 % and 63 % were super resistant (i.e., 0 % mortality) with cypermethrin and deltamethrin respectively. Resistance was also detected against organophosphates-synthetic pyrethroids co-formulation, OP chlorfenvinphos, and Amitraz (Vudriko *et al.*, 2016). Several studies have reported acaricide resistance in Tanzania. The study by Kagaruki, (1991) reported that about 45 % of the tested tick strains were resistant to organochlorides while Nagagi *et al.*(2020) reported the existence of ticks that showed variable levels of tolerance and resistance to alphacypermethrin, cypermethrin, and chlorpyrifos. It is therefore very important to conduct periodic monitoring of the ticks for the development of resistance against commonly used acaricides for improvement of livestock production.

### **1.5 Problem Statement and Justification**

Animal keeping is an important element of countryside incomes in African societies. There are many factors including different parasites and diseases that affect animal health and their productivity as main constraints in the growth and well-being of the societies (Torr *et al.*, 2002). Ticks cause stress and weaken the immune system leading to a decrease in animal products in both quantity and quality such as milk, meat, eggs, and leather as a result, the TBDs can limit the international trade of animals, their products, and sporting events (Oscar and Cristian, 2018).

Mbassa *et al.*(2009), have indicated that acaricide is the most useful and economically controlled measure of ticks and TBDs. Also, the study of Nagagi *et al.*(2020) which surveyed a few areas in Tanzania indicated that there is an emerging tick population that is resistant to the commonly used acaricides. However, the acaricide resistance is mostly caused by poor practices on acaricide applications that are performed individually through privately owned and commercially-run dip tanks. Likewise, the situation is promoted further by free acaricide market access, lack of periodic tick monitoring, and inadequate technical knowledge on acaricide application. Apart from the study of Kagaruki (1991) and Nagagi *et al.*(2020) which covered most regions of Tanzania and reported the presence of acaricide resistance, there is limited information on current tick-borne disease control practices, the prevalent tick species and the susceptibility status to the commonly used acaricides in different parts of Tanzania. The current study aimed at tracking the acaricide resistance in the Eastern zone of Tanzania to establish the list of drugs that can still be useful for the control of ticks and TBDs and those that need to be considered out of use. It was therefore important to survey to identify tick populations that have developed resistance against commonly used acaricides.

This information will contribute to minimizing the costs of production associated with ineffective acaricides which force farmers to either increase the acaricide concentration or reduce the interval of acaricide application. Also, minimize animal mortality caused by tick-borne diseases resulting from the failure of the tick control program and advise appropriate measures for controlling Tick-borne diseases in the country.

## **1.6 Objectives of the Study**

### **1.6.1 General objective**

The general objective of this study was to investigate farmers' awareness of tick-borne diseases and acaricide resistance in cattle ticks in the eastern zone of Tanzania.

### 1.6.2 Specific objective

- i. To assess farmers' awareness and control practices of Ticks and Tick-borne diseases in Eastern Tanzania.
- ii. To determine the prevalent tick species and their resistance status against frequently used acaricides in tick control programs.

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## CHAPTER TWO

### MANUSCRIPT I

#### **2.0 Commonly used Acaricides and their Application Methods Practiced in Eastern Tanzania**

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#### **Abstract**

Ticks are blood-feeding arthropods that transmit pathogens causing diseases of public and veterinary importance such as protozoa, helminths, and bacteria. Chemical control of ticks is the only simple and feasible method despite ticks developing resistance against specific commonly used acaricides. This study aimed to investigate commonly used acaricides and general tick control practices in the study areas where structured questionnaires were purposively administered to 129 cattle farmers from Morogoro and Chalinze districts in Morogoro and Pwani regions respectively.

The majority of respondent 63 (48.8%) applied the acaricide at the intervals of once per week followed by those who were not considering any interval (uncontrolled interval) by 51 (39.6%). Of most of the livestock keepers, 90 (69.8%) prepared acaricide solutions without following manufacturers' instructions. Application of acaricides by hand spray methods was mostly preferred by 115 (89.1%) over than dipping method due to various reasons including the availability of the dipping (tank) site. Most of the respondents 44 (34.1%) claimed to have used the same acaricide for more than ten years due to the availability in the market and high cost of other types of acaricides as described by several respondents 28 (21.7%) and 6 (4.7%) respectively, others were

purposively interchanging acaricides depending on some factors such as resistance suspicion to a certain acaricide by 63 (48.8%) of respondents. Based on simple observation and respondents' responses on tick control practices, it is highly suggestive that, there is resistance among the acaricides mentioned by respondents in their areas.

Finally, since the study was a structured questionnaire-based design, further experimental study should be conducted to observe the resistance status of common acaricides used in the study areas.

**Keywords.** Awareness, control practices, Ticks, Tick-borne disease,

## 2.1 Introduction

Hard ticks are arthropods of vertebrate that comprise several genera including, *Rhipicephalus (B) microplus*, *R. appendiculatus*, *Amblyomma variegatum*, and *R. (Boophilus) decoloratus* that are said to be more prevalent in tropical and sub-tropical regions including Tanzania (Mamiro *et al.*, 2016; Byaruhanga *et al.*, 2016). Ticks being blood-feeding arachnids can transmit pathogens of public and veterinary importance such as protozoa, bacteria and viruses (Tawana *et al.*, 2022; Mutavi *et al.*, 2018). The most prevalent TBDs affecting cattle include *babesiosis*, *theileriosis*, *cowdriosis*, and *anaplasmosis* (Nagagi *et al.*, 2020; Kivaria, 2006). Tick and tick-borne diseases (TBD) are serious constraints to many African farmer's economies including East African communities due to their effect on animal health and production (Oundo *et al.*, 2022; Haji *et al.*, 2022 ). In Tanzania, 80% of animals kept are at risk of being affected by ticks and TBD every year. TBDs affect livestock production either directly by reducing animal products such as milk and meat as well as increasing morbidity and mortality rates or indirectly through an increase in production costs ( ). Due to the great losses caused by ticks,

several efforts have been tried to control ticks and TBDs by use of local methods such as oil application, handpicking, and medicinal plants as well as chemical methods that appear to be more effective, time-saving, and economical. The most frequently applied acaricide groups include pyrethroids, amitraz, and organophosphates (Baffi *et al.*, 2005; Kagaruki, 1996). These acaricides are applied through hand washing, spraying, or dipping. This shows that the chemical control method is an important technique in controlling ticks and TBDs in the livestock production industry despite ticks developing resistance as well as environmental pollution (Nagagi *et al.*, 2020). The acaricide application is characterized by many factors such as knowledge of pesticide application, periodic tick monitoring, climatic conditions, and availability of pesticide in the free market as well as dipping sites (dip tanks) around the livestock keepers (Jongejan & Uilenberg, 2004; Mbassa *et al.* 2009). All these factors are responsible for increasing selection pressure that influences emergency and development of acaricide resistance (Abbas *et al.*, 2014; Vudriko *et al.*, 2018).

However, the study of Kagaruki (1991) and Nagagi *et al.*, (2020) reported the presence of malpractices and resistance to some acaricides from different parts of Tanzania. It was, therefore, important to survey the Eastern zone of Tanzania to evaluate the common acaricides currently used by livestock farmers and their general knowledge of acaricide application methods. The findings will be used as the baseline information required by different stakeholders involved in livestock production.

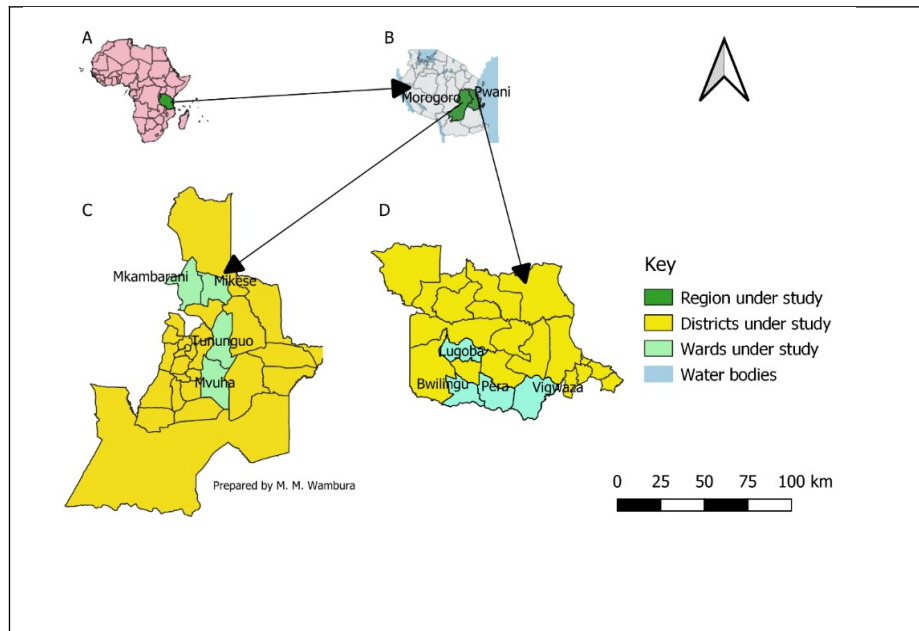
## **2.2 MATERIALS AND METHODS**

### **2.2.1 Study Area**

This study was conducted in Pwani and Morogoro regions. Pwani region is located between Latitude: -7° 00' 0.00" S and Longitude: 39° 00' 0.00" E and is bordered to the east by Dar es Salaam Region and

the Indian Ocean, to the west by Morogoro region, to the north by Tanga region and the south by Lindi region. On the other hand, the

Morogoro region is located between latitude 5° C 58" and 10° C 0" S and longitude 35° C 25" and 35° C 30" E. The region is bordered to the north by the Tanga and Manyara regions, to the south by the Ruvuma region, to the west by the Iringa and Dodoma regions, and to the east by the Pwani and Lindi regions. From each region, one district was selected Chalinze and Morogoro Districts where in each district four wards were purposively selected depending on cattle population whereby wards with large numbers of cattle were selected as indicated in Figure 2.1. Morogoro region is characterized by a tropical grassland climate having average temperatures of about 25 °C which vary from 18° C in high areas to 30° C in lowland areas. Likewise, regular rainfall ranges from 600mm to 1200mm on low and high land respectively. . Similarly, the climatic condition of the Pwani region is regarded as a tropical climate whereby temperature has a seasonal average temperature fluctuating from 13° C to 30° C and humidity as high as 98% (EPMS, 2006). Likewise, rainfall ranges between 800 – 1200 mm per annum with a bimodal rainfall pattern, a short rainy period from October to December, and a long rainy season between March and June. (Msuya *et al.*, 2013; Mushi, 2009; United Republic of Tanzania Coast Region, 2015). Generally, the livestock sector is dominated by smallholder livestock farmers, keeping mostly indigenous cattle, sheep, goats, pigs, and chicken which contributes to the National Growth Domestic Product (GDP). Apart from the livestock sector, these two regions deal with agricultural activities, forestry, fisheries, beekeeping, wildlife, and Industrial sectors (United Republic of Tanzania Coast Region profile 2015).



**Figure 2.1:** Study area map showing the distribution of eight wards from two regions each with one district and each district has four wards. 'A' represents the African continent, 'B' represents Tanzania, 'C' represents Morogoro district, and 'D' represents the Chalinze/Bagamoyo district.

### 2.2.2 Study Design and Data Collection

A cross-sectional study design was adopted in this study where the target study population was households involved in livestock keeping at eight purposively selected wards from Morogoro and Chalinze districts. The sample size was calculated according to Charan and Biswas, (2013) with a 95% confidence level.  $(n) = (Z_{1-\alpha/2})^2 \frac{P(1-P)}{d^2}$  Where; n = Sample size, P = Is the Expected proportion /estimated

percentage of livestock farmers in the population who are expected to be aware of Tick and TBDs (0.5),  $d^2 =$  Absolute (marginal) error (0.05),  $Z_{1-\alpha/2} =$  Critical value and a standard value for the corresponding level of confidence. Therefore, the estimated sample size was 384. However, due to the household accessibility and availability of the household members with their cattle only one hundred and twenty-nine (129) households, (66 and 63 households from Morogoro and Chalinze districts respectively) were interviewed based on their involvement in livestock keeping especially cattle and being above 15 years old after a verbal consent. The structured questionnaire was written in English and translated into a national language (Swahili) as well as accompanied with visual observations to confirm some respondent responses. The information obtained was properly recorded and finally used for analysis.

### **2.2.3 Data analysis**

The collected data were entered, coded, validated, and managed into a Microsoft Excel spreadsheet where descriptive statistics were performed with the help of IBM SPSS (Statistical Package for Social Science) version 20. The variations between demographic features and a general understanding of respondents on commonly used acaricides and their application methods were computed using the Chi-square test. However, any variation was considered statistically significant only if  $p\text{-value} \leq 0.05$ .

### **2.3 Ethical consideration**

The Research and Ethical clearance was approved by the Institutional review board of the Sokoine University of Agriculture with reference numbers SUA/DPRTC/R/126/VET/3/2021/4 and SUA/DPRTC/R/186/VOL. TV -7 respectively. Likewise, permission to conduct the study in the area was obtained from the Morogoro and Pwani regions with reference number AB.175/245/01/46 and FA.221/265/01F/102 respectively.

## 2.4 RESULT

### 2.4.1 Social demographic characteristics of the studied population

A total of 129 participants were interviewed whereby most of the participants 83 (64.3%) were above 37 years old and the number of males was 112 (86.8%) than the females. Married people formed a large number of respondents 110 (84.6%) of which most of them were from Chalinze District 58 (45.0%) and Morogoro District 52 (40.3%). In the case of education level, most of them had primary education 71 (55.0%) while respondent's ethnicity composed of Maasai 87 (67.5%) and non-Masai 42 (32.6%) as shown in Table 2.1.

**Table 2.1: Demographic characteristics of study respondents from Chalinze and Morogoro districts**

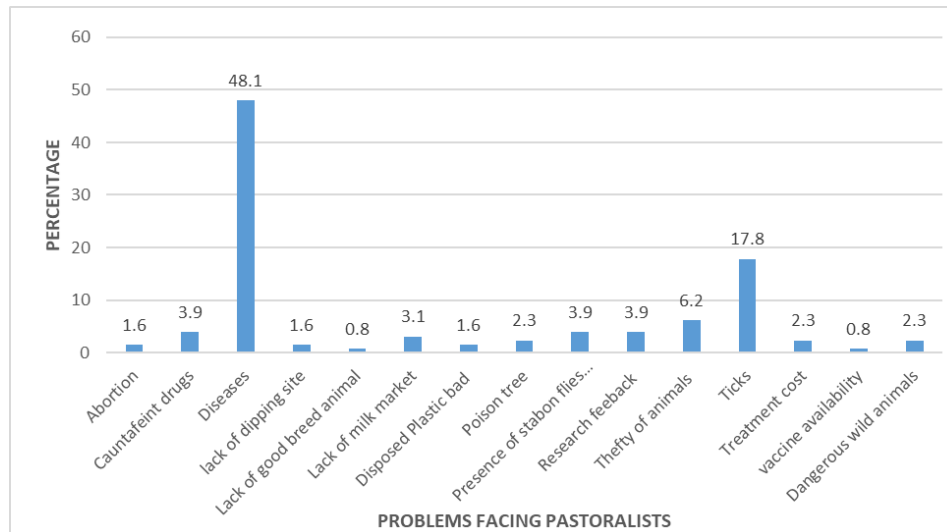
Variables		Chalinze District (n=63) Frequency (%)	Morogoro District (n=66) Frequency (%)	Total (N=129) Frequency (%)
Sex	Male	53 (41.1)	59 (45.7)	112 (86.8)
	Female	10 (7.8)	7 (5.4)	17 (13.2)
Age	15-25	7 (5.4)	13 (10.1)	20 (15.5)
	26-36	11 (8.5)	15 (11.6)	26 (20.2)
	Above 37	45 (34.9)	38 (29.5)	83 (64.3)
Education	None	21 (16.3)	22 (17.1)	43 (33.3)
	Primary	36 (27.9)	35 (27.1)	71 (55.0)
	High school	6 (4.7)	5 (3.9)	11 (8.6)
	College/University	0	4 (3.1)	4 (3.1)
Origin	Masai	41(31.8)	46 (35.7)	87 (67.5)
	Non Masai	22 (17.1)	20 (15.5)	42 (32.6)
Marital status	Married	58(45.0)	52 (40.3)	110 (85.3)
	Single	4 (3.1)	7 (5.4)	11 (8.5)
	Widowed	1 (0.8)	6(4.7)	7 (5.5)
	Divorced	0	1 (0.8)	1(0.8)

#### **2.4.2 Respondents' Livestock management practices**

The livestock management practices were evaluated through questionnaires and observation during farm visits. The results reveal that apart from keeping cattle the majority of respondents kept other animals such as goats 105 (81%), sheep 70 (54.3%), poultry 34 (26.4%), and donkeys 2 (1.6%). Of the high number of farmers 115 (89.9%) claimed to have kept animals for more than ten years while 116 (89.9%) of farmers provided fenced shelter to their animals, although all respondents that had both cattle and goat or sheep had also both fence and house for cattle and goat or sheep respectively. Most respondents 121(93.8%) used free-range grazing feeding mode while those using zero-grazing were only 8 (6.2%) as shown in Table 2.2. Among the challenges mentioned by respondents, diseases ranked high at 62 (48.1 %) followed by tick infestation at 23 (17.8%) as shown in Figure 2.2 and Table 2.3.

**Table 2.2:** Respondents' livestock management practices

Variables	Chalinze district N=53	Morogoro district N=76	Total N=129
<b>Other animals kept by Livestock farmers</b>			
<b>Goat</b>			
Farmers keeping <b>Goat</b>	40 (31.0)	65 (50.4)	105 (81.4)
Farmers not keeping <b>Goat</b>	13 (10.1)	11 (8.5)	24 (18.6)
<b>Sheep</b>			
Farmers keeping sheep	24 (18.6)	46 (35.7)	70 (54.3)
Farmers not keeping sheep	29 (22.5)	30 (23.2)	59 (45.7)
<b>Poultry</b>			
Farmers keeping <b>Poultry</b>	12 (9.3)	22 (17.1)	34 (26.4)
Farmers not keeping <b>Poultry</b>	41 (31.8)	54 (41.8)	95 (73.6)
<b>Donkey</b>			
Farmers keeping <b>Donkey</b>	0	2 (1.6)	2 (1.6)
Farmers not keeping <b>Donkey</b>	53 (41.1)	74 (57.4)	127 (98.5)
<b>Duration</b>			
1-2 years	2 (1.6)	1 (0.8)	3 (2.3)
3-5 years	3 (2.3)	1 (0.8)	4 (3.1)
6-10 years	2 (1.6)	5 (3.9)	7 (5.4)
More than ten years	46 (35.7)	69 (53.5)	115 (89.9)
<b>Animal shelter</b>			
Fenced shelter	48 (37.2)	68 (52.7)	116 (89.9)
House	5 (3.9)	8 (6.2)	13 (10.1)
<b>Feeding model</b>			
Free range Grazing	49 (38.0)	72 (55.8)	121 (93.8)
Zero grazing	4 (3.1)	4 (3.1)	8 (6.2)



**Figure 2.2:** Bar chart showing the percentage of respondents and the problems they are facing in livestock production

#### 2.4.3 General farmers' understanding of diseases affecting livestock in study areas

The farmers mentioned different diseases that were highly affecting their animals whereby East Coast fever (ECF) ranked second with frequency of 81 (62.8%) after CBPP which was 104 (80.6%). Nevertheless, when respondents were asked to mention the diseases they know, only a small percent could mention the specific names, ECF 22 (17.1%), *babesiosis* 9 (7.0%), and *anaplasmosis* 8 (6.2%) showing that most of the respondents (82.9%, 93.0% and 93.8% respectively) were not aware that these diseases are transmitted by ticks, as shown in the Table 2.3.

**Table 2.3: General understanding of respondents on diseases affecting livestock in the study areas**

Diseases	Frequency (%)	Chalinze District (n=63)	Morogoro District (n=66)	Overall (N=129) Frequency (%)	Chi-Square Tests p-value ≤ 0.05
		Frequency (%)	Frequency (%)		
FMD	Mentioned	17 (13.2)	34 (26.4)	51 (39.5)	0.004
	Not Mentioned	46 (35.7)	32 (24.8)	78 (60.5)	
CBPP	Mentioned	51 (39.5)	53 (41.1)	104 (80.6)	0.926
	Not Mentioned	12 (9.3)	13 (10.1)	25 (19.4)	
Theileriosis	Mentioned	50 (38.8)	31 (24.0)	81 (62.8)	0
	Not Mentioned	13 (10.1)	35 (27.1)	48 (37.2)	
Babesiosis	Mentioned	15 (11.6)	7 (5.4)	22 (17.1)	0.068
	Not Mentioned	48 (37.2)	59 (45.7)	107 (82.9)	
Anaplasmosis	Mentioned	8 (6.2)	1 (0.8)	9 (7.0)	0.013
	Not Mentioned	55 (42.6)	65 (50.3)	120 (93.0)	
Respondent's awareness of tick-borne parasitic diseases					
ECF	Aware	9 (7.0)	13 (10.1)	22 (17.1)	0.414
	Unaware	54 (41.9)	53 (41.1)	107 (82.9)	
Babesiosis	Aware	2 (1.6)	7 (5.4)	9 (7.0)	0.098
	Un aware	61 (47.3)	59 (45.7)	120 (93.0)	
Anaplasmosis	Aware	5 (3.9)	3 (2.3)	8 (6.2)	0.425
	Unaware	58 (45.0)	63 (48.8)	121 (3.8)	

#### 2.4.4 Tick and Tick control practices

The general understanding of respondents on different tick species identification was high and more than 75% of all respondents were able to mention at least one type of tick by stating the main features that distinguish specific tick genera such as *Boophilus* 36 (27.9%), *Amblyomma* 37 (28.7%) and *Rhipicephalus* 38 (29.5%). Almost all respondents mentioned acaricide as the method used to control ticks. The most preferred application method was hand spray which was mentioned by 115 (89.1%) of respondents and only 10 (7.8%) respondents claimed to use both dipping and hand spray. The majority of respondents 63 (48.8%) were applying the acaricide at intervals of once per week and 44 (34.1%) respondents claimed to have used one type of acaricide for more than ten years as shown in Table 2.4.

**Table 2.4: Ticks and tick control management**

Variables	Chalinze District (n=63) Frequency (%)	Morogoro District (n=66) Frequency (%)	Overall (N=129)	X <sup>2</sup> -Tests P ≤ 0.05
<b>Control Method</b>				
Chemical	60 (46.5)	62 (48.1)	122 (94.6)	
Handi picking	3 (2.3)	4 (3.1)	7 (5.4)	0.920
<b>Application interval</b>				
once per week	41 (31.8)	22 (17.1)	63 (48.8)	
Not controlled	13 (10.1)	38 (29.5)	51 (39.6)	
after 3-5days	7 (5.4)	0	7 (5.4)	
Fourteen day	1 (0.8)	6 (4.7)	7 (5.4)	0.001
after 3 week	1 (0.8)	0	1 (0.8)	

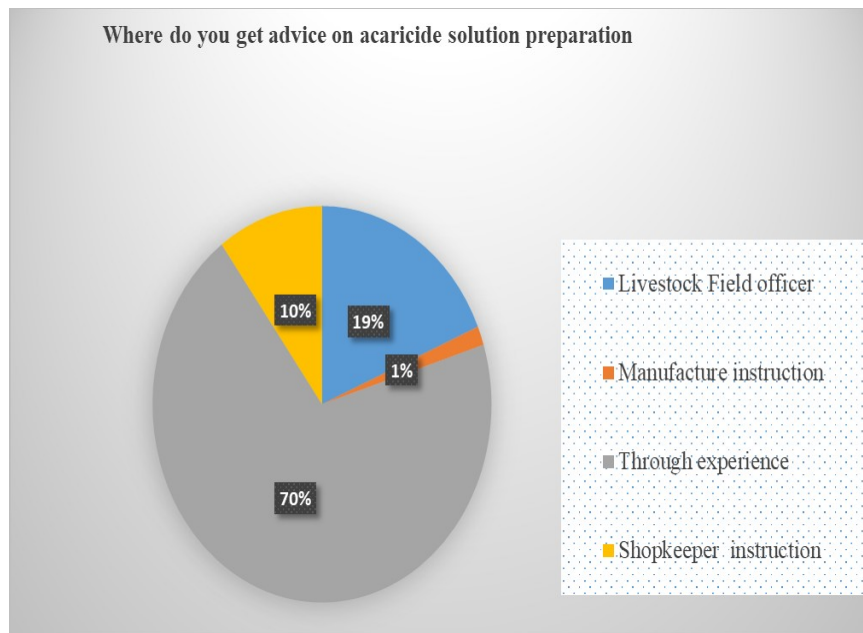
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<b>Duration of acaricide use</b>				
2-4 years	10 (7.8)	5 (3.9)	15 (11.6)	
5-7 years	7 (5.4)	2 (1.6)	9 (7.0)	
less than a year	7 (5.4)	2 (1.6)	9 (7.0)	
More than 10 years	10 (7.8)	34 (26.4)	44 (34.1)	0.001
8-10 years	7 (5.4)	22 (17.1)	29 (22.5)	
Not controlled	22 (17.1)	1 (0.8)	23 (17.8)	
Know Boophilus spp	16 (12.4)	20 (15.5)	36 (27.9)	
Don't know Boophilus spp	47 (36.4)	46 (35.7)	93 (72.1)	
Know Rhipicephelus spp	20 (15.5)	18 (14.0)	38 (29.5)	
Don't know Rhipicephelus spp	43 (33.3)	48 (37.2)	91 (70.5)	
Know Amblyomma spp	19 (14.7)	18 (14.0)	37 (28.7)	
Don't know Amblyomma spp	44 (34.1)	48 (37.2)	92 (71.3)	0.753
Apply acaricide by Dipping	7 (5.4)	3 (2.3)	10 (7.8)	
Don't apply by Dipping	56 (43.4)	63 (48.8)	119 (92.2)	
Apply acaricide by hand spraying	59 (45.7)	56 (43.4)	115 (89.1)	
Don't apply by hand spraying	4 (3.1)	10 (7.8)	14 (10.9)	0.256

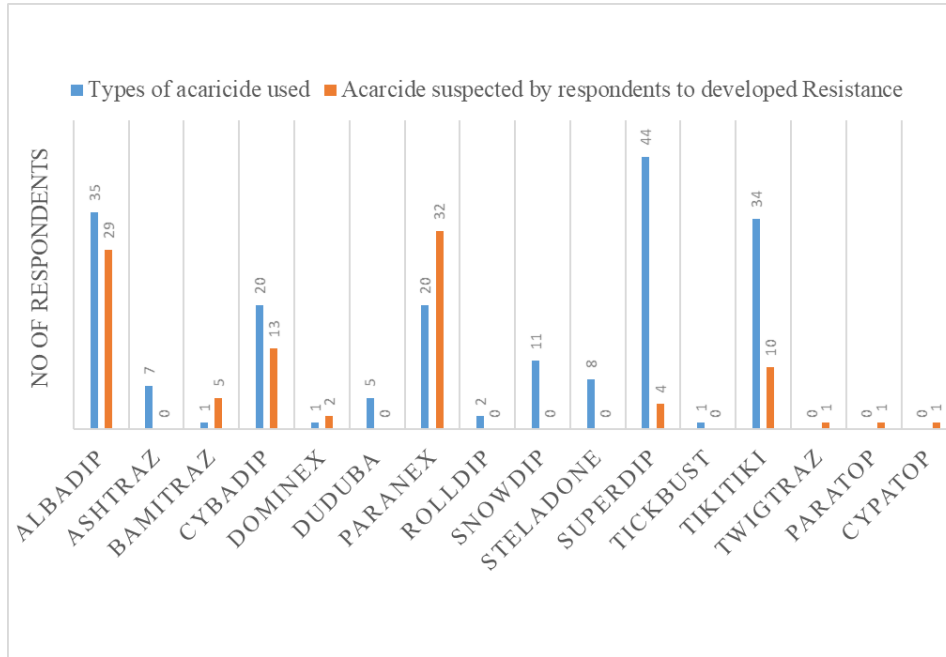
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### 2.4.5 Acaricide preparation and dilutions

The farmers were asked to state how they prepared the solution for acaricide application and where they got that knowledge. The majority 90 (69.8%) decide on the amount of chemical to use based on their long-term experience with animal keeping while 24 (18.6%) were taught by their field officers around them and 13 (10.1%) obtained the information from acaricide retail traders but only two respondents were using manufacturer's instructions on how to prepare the solution as shown in **figure 2.3** below. Along with this line, they listed some acaricides that they thought ticks have developed resistance whereby Alphacypermethrin 100g/l ranked high by 32 (24.8%) respondents followed by Alphacypermethrin 10g/l as shown in **figure 2.4 and 2.5**.

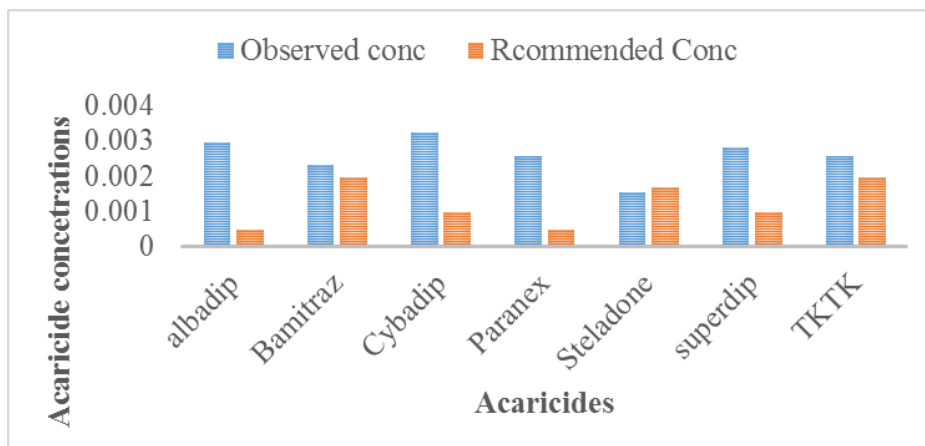


**Figure 2.3:** Showing the source of acaricide solution preparation information.



**Figure 2.4.** Showing types of acaricides used and those suspected to develop resistance against ticks

**Note:** Duduba® is not acaricide however, the livestock farmers were using it to control ticks and other stubborn flies such as stomoxis.



**Figure 2.5.** Showing acaricides observed concentration (concentration prepared by respondents) against expected (manufacturer) concentration

## **2.5 Discussion**

This study shows the general awareness of livestock farmers on ticks and tick-borne diseases affecting cattle and the control methods practiced in the eastern part of Tanzania. The findings show that most of the respondents were married males who attended primary education and aged above 37 years old. This is consistent with the observation made by (Chenyambuga *et al.*, 2010). Livestock farming was the main economic activity for most of the people around the surveyed areas. These findings correspond to the existing report from (Laisser *et al.*, 2014; Nkonya *et al.*, 2004).

In the pastoralist community, men only take basic education and sometimes they don't go to school at all instead they spend most of their time grazing their animals while learning from their elders how to

manage their livestock as has been revealed by most of the respondents by claiming to be involved in livestock keeping for more than ten years whereby similar findings were reported by Chenyambuga *et al.* (2010). Free-range grazing and fenced shelters were the main feeding and shelter practices used respectively by most of the respondents although few were using houses with some supplements to their animals where this finding is similar to that reported by Baltussen *et al.*, 2020 and Kavana *et al.*, 2005. The study shows that livestock diseases were the main challenge in the two districts followed by tick infestation, other problems such as the presence of counterfeit acaricides, lack of dipping sites, and high-cost acaricides collectively account for an increased number of ticks that are responsible for carrying and transmitting disease-causing agents to animals as also reported by Jaime Betancur Hurtado & Giraldo-Ríos, 2019 and Nagagi *et al.*, 2020.

The respondents were able to mention the signs of diseases affecting their animals although some of them used local disease names such as ndigana kali which means ECF and ndigana baridi meaning *Anaplasmosis*, while few of them were able to specify them by either recalling the signs or specific name of the disease. *Babesiosis* was described as “kukojoa damu” in the Swahili language which means haematuria while *anaplasmosis* disease was characterized by hard dung faeces and this finding concurs with the report of Chenyambuga *et al.* (2010) and Jacob *et al.* (2017). Nevertheless, the respondents had poor knowledge of TBD with an exceptional few respondents knowledgeable about diseases such as ECF 22 (17.1%), *Babesiosis* 9 (7.0%), and *Anaplasmosis* 8 (6.2%), similar results have been reported by (Laisser *et al.*, 2014; Jacob *et al.*, 2017). The findings indicate that, although respondents were able to mention different TBDs affecting their cattle, they were not aware that those diseases are transmitted by ticks (Table 2.3).

The results show that the respondents had some knowledge about ticks whereby, some of the respondents used key features of ticks for identification such as red and small in size representing *Rhipicephalus* genera (29.5%), while *Rhipicephalus (Boophilus)* spp identified as big and blue by 27.9% and *Amblyomma* spp known as colored tick by 28.7% of respondents. This is consistency with the observation made by Walker *et al.*(2014). Nevertheless, the presence of these tick species conforms to the types of TBDs listed by respondents as also reported by (Kerario *et al.*, 2017; Isack *et al.*, 2017).

The chemical method was the most common method used to control ticks while hand picking was used by women during milking time which is similar to reports (Hezron *et al.*, 2012; FAO, 2004). There were about fifteen acaricides and one pesticide that were listed to be used by respondents to control ticks. The mentioned pesticide (Duduba<sup>®</sup>) is designed to be used in crops but they claimed to use it for controlling ticks and stubborn flies (*Stomoxys*) because most acaricides were not able to kill the ticks as well as prevent annoying flies from biting their animals. Hence due to their experience, they were mixing Duduba and either of the acaricides such as Paranex, Superdip, Albadip, Cybadip, and Tikitik, to serve the dual purpose as also observed by Nagagi *et al.* (2020). Some of the respondents (48.8%) reported using the acaricide at the interval of once per week while others used them at an interval of 3-5 days (5.4%) or after two weeks (5.4%) as indicated in table 2.3. The majority used a single acaricide for more than ten (10) years due to different factors including, easy availability and low cost while suspicion of acaricide resistance was the key factor that led them to change from one acaricide to another as also reported by (Hezron

*et al.*, 2012; Ogden *et al.*, 2005). The commonly used acaricide application method was hand spray although, a small number of respondents were using both dipping and hand spray which concurs with the report of Laisser *et al.*(2014). Although the hand spray method is easy and feasible for small-scale farmers, it is less efficient as it needs special care to apply chemicals thoroughly in areas such as ears, under tail, udder, axillae, and other inaccessible areas of the animal. Indeed, most farmers did not have structures for immobilizing the animals during the acaricide application but all animals were just confined in a fence making it difficult to achieve thorough acaricide application.

The majority of interviewees reported that they prepared acaricide solution based on their own experience such as increasing the acaricide concentration until the solution turned into a strong milky color or using 2 or 3 caps (a special container for measuring the chemical during acaricide preparation in 15L or 20L knapsack sprayer) whereby one cap has 20 to 30 ml capacity volume depending on manufacturer. However, few of them were following the manufacturer's instructions.

Githaka *et al.* (2022) suggested that livestock farmers in Africa are normally not aware of the different chemical compositions between acaricide brands as a result the same concentrations are used for all acaricide brands other than those recommended by the manufacturers likewise, they practice incorrect combination or rotation of different chemical classes.

Overdosing as well as persistent use of the acaricide are the main aspects that accelerate resistance selection of acaricide in ticks (Abbas *et al.*, 2014). There is a relationship between either underdosing or

overdosing on the drug and the degree of resistance as revealed by Vudriko *et al.* (2018) and Lees *et al.* (2013) that, a species controlled by a recommended dose of acaricide may show resistance when a low dose of the same acaricide is applied. Nevertheless, most of the respondents admitted not seeking advice from veterinary professionals in their localities as also reported by (Mugisha *et al* 2005). Moreover, the weekly application of acaricide, inappropriate acaricide dose, seemed to be uneconomical in indigenous cattle due to the high price of acaricides (Pegram *et al* 1993; Okello-Onen and Rutagwenda 1998).

## **2.6 Conclusion**

In this study ticks and TBDs are the most common cattle production constraints identified by livestock keepers where East Coast fever (ECF) disease ranked second after CBPP followed by tick infestation. However, the respondents had low knowledge about tick species identification as well as TBDs indicating that most of the respondents were not aware that these diseases are transmitted by ticks. The chemical method was the most used by almost all respondents through hand spray although few respondents used the dipping method. About sixteen acaricides were used by respondents in the study area and the majority agreed to use one acaricide for more than ten years at the interval of once per week. The majority of livestock keepers obtained knowledge of acaricide solution preparation through their long-term experience in animal keeping while few were taught by their field officers, acaricide retail traders, and manufacturer's instructions. As a result, most of the respondents were using acaricide concentrations which were above the manufacturers' recommendation. These acaricide malpractices and lack of dipping sites may increase the risk of emergency and the development of acaricide-resistant tick species. Indeed the livestock farmers listed some acaricides that they thought to be resisted by ticks whereby Alphacypermethrin 100g/l ranked high followed by Alphacypermethrin 10g/l.

## **2.7 Recommendations**

This study has revealed that there are malpractices in acaricide usage and low knowledge of TBDs. Education should be provided to the pastoralists and agro-pastoralists on acaricide application and tick control practices in general through livestock field officers as well as constructing dipping sites. On the other hand, the government should provide acaricide subsidies to lower their cost allowing the farmers to have an equal chance of buying any acaricides he /she want to prevent the use of one acaricide for a prolonged time. Finally, since the study was based on a structured questionnaire design, the experimental study should be conducted to observe the resistance status of commonly used acaricides in the study areas.

## **2.8 Acknowledgment**

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## **2.9 Conflict of interest**

The authors declare that there is no conflict of interest on the matter of publication of this research paper.

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**CHAPTER THREE**  
**MANUSCRIPT II**

**3.0 OCCURRENCE OF TICK SPECIES IN CATTLE AND THEIR  
SUSCEPTIBILITY STATUS AGAINST FREQUENTLY USED  
ACARICIDES IN MOROGORO AND PWANI REGIONS**

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**Abstract**

Ticks are ectoparasites which are important vectors of various tick-borne pathogens including protozoa, bacteria, and viruses that cause life-threatening diseases in animals and humans. Proper knowledge of tick species distribution and their susceptibility status against commonly used acaricides is important for the successful control of ticks and tick-borne diseases. This study aimed to determine the prevalent tick species and resistance status of the frequently used acaricides in tick control programs in Eastern Tanzania. A cross-sectional study design was conducted to collect ticks and information on tick control practices. A purposive sampling method was adopted whereby a total of 952 ticks were collected from naturally-infested cattle herds. None engorged ticks were stored in plastic bottles filled with 70% ethanol while engorged or semi-engorged ticks were put in special perforated plastic containers placed on moisture cotton wool inside the box. Tick species were identified by using macroscopic and microscopic morphological features according to Walker *et al* (2014).

To assess the tick's susceptibility status, an experimental study design was adopted to evaluate the effectiveness of the selected commonly used acaricides whereby an In-vitro acaricide susceptibility test using the Larval Packet Test (LPT) was performed according to FAO (2004) with some modifications. Eight commonly used acaricides with active ingredients; AM (Amitraz), OPs (chlorfenvinphos and chlorpyrifos), and SP (alphacypermethrin and cypermethrin) were used. Two concentrations were prepared that are Discriminatory doses formulated by doubling the recommended dose and control using diluents composed of olive oil and acetone at a 1:2 ratio where both dead and live larvae were counted according to Shyma *et al.* (2015). The findings indicate that out of all ticks collected from the study area, *R (Bo) microplus* was the predominant species making a total of 552 (57.9%) followed by *Amblyomma variegatum* by 247(25.9%), *R (Bo) decoloratus* by 65(6.8%), *Hyalomma rufipes* 41(4.3%) while the least species were *R. evertsi* and *R. appendiculatus* with 37(3.8%) and 10(1.1%) respectively. On the other hand, acaricide susceptibility status appeared to be low for acaricide; acaricide C (Cypermethrin 15g/l) and acaricide D (Alphacypermethrin 100g/l) by 84.60% and 85.70% respectively followed by acaricide E (Alphacypermethrin 10g/l) and acaricide B (Alphacypermethrin100g/l + Chlorpyrifos 100g/l) by 91.30% and 93.90% while acaricide H (Amitraz 125g/l), acaricide A (Amitraz 12.5%), acaricide F (Chlorpyrifos 500g/l + Cypermethrin 50g/l) and acaricide G (Chlorfenvinphos 300g/l) showed no resistance as exhibited by 97.80%, 97.90% 99.80% and 99.90% percentage mortality respectively. Generally, the study revealed that almost all tick species were susceptible to Amidines (amitraz) and OP (Chlorpyrifos and Chlorfenvinphos) while SP (Cypermethrin and Alphacypermethrin) showed signs of developing resistance.

### 3.1 Introduction

Ticks are important vectors of tick-borne diseases (TBDs) of major animals that constrain livestock production as a result of morbidity and mortality caused by those diseases. Ticks (class: Arachnida, Order: Ixodida) parasitize animals by feeding on blood thereby causing anemia and reduced live weight gain as well as hide and skin quality because of local necrosis attributed to tick bites. Tick bites can also result in a reduction in meat and milk production and increased cost of production because of tick control operations (Mamiro *et al.*, 2016). Furthermore, it indirectly affects cattle by causing tick worry which eventually weakens the immune system leading to increased susceptibility to various diseases (Nagagi *et al.*, 2020; Peter *et al.*, 2005).

The abundance and distribution of tick and TBD are contributed by many factors such as operation (biotic) factors including the presence of a host, and drug application practices as well as abiotic factors that include temperature, rainfall, and humidity. Any change from normal circumstances of one or more of these factors leads to a change in the abundance and distribution of ticks and TBDs (Jongejan & Uilenberg, 2004; Ledwaba *et al.*, 2022).

In Africa, the main tick species upsetting animal production include *Amblyomma hebraeum*, *Ixodes rubicundus*, and *Rhipicephalus appendiculatus* while the most prevalent tick species was *Rh. (Bo) decoloratus* and *Rh. (Bo) microplus*, (Horak *et al.*, 2015). In Tanzania, (Kerario *et al.*, 2017) reported seven tick species namely *A. variegatum*, *A. lepidum*, *R. microplus*, *R. appendiculatus*, *R. evertsi*, *R. decoloratus*, and *H. rufipes*) in all the three regions. The most prevalent species in Mara, Mbeya, and Singida were *R. appendiculatus* (50.5%), *R. evertsi* (35.6%), and *A. lepidum* (31.2%) respectively. Lynen *et al.*, (2007) reported that the distribution range of *B. microplus* extended

more to the northern regions and other areas of Tanzania that were previously not-occupied, while *B. decoloratus* was largely confined to highlands in central and northern Tanzania and several other high-altitude refuges. The study conducted by Mamiro *et al.* (2016) in the Pwani region reported only four prevalent tick species namely *R. microplus* (27.40%), *R. evertsi* (10.25%), *R. appendiculatus* (6.19%) and *A. variegatum* (56.10%). Walker *et al.*, (2014); Hezron *E et al.*, (2012); Yeoman and Walker (1967) have reported different tick species and TBDs in different parts of Tanzania such as *Rhipicephalus (B) decoloratus* and *R. (B) microplus* which transmit *Anaplasma marginale* and *Babesia bigemina* that cause bovine *anaplasmosis* (gall sickness) and bovine *babesiosis* (redwater) respectively, Other ticks such as *Rhipicephalus appendiculatus* transmit East Coast fever and benign bovine *theileriosis* in cattle. *Rhipicephalus evertsi* transmit pathogens that cause bovine *anaplasmosis*. *Amblyomma variegatum* and *A. lepidum* transmit *Ehrlichia ruminantium* and *Theileria mutans* that cause *Cowdria ruminantium*, (*heartwater*) and *Theileriosis*, respectively. Also, *Hyalomma rufipes* transmit *Anaplasma marginale* (bovine *anaplasmosis*).

Acaricide application has been a major means of tick control and hence TBDs on livestock, although other methods such as handpicking, oil application, biological method and medicinal plants are sparingly used (Moyo & Masika, 2009; Nagagi *et al.*, 2020; George, 2000). The most frequently used acaricides belong to organophosphates, amitraz, and pyrethroids (Baffi *et al.*, 2005; Kagaruki, 1996). However, it is well noted that intensive use of acaricides is one of the positive risk factors for the development of resistant tick species. This is due to imposed selection pressure where susceptible tick populations are removed and at the same time increases the proportion of the resistant strains (Abbas *et al.*, 2014).

Indisputably, ticks develop resistance to different acaricide groups depending on the acaricide used, concentration, and frequency of application intervals (Drummond, 1983; Spickett, 1998). Other factors such as free acaricide market access, lack of periodic tick monitoring, and inadequate technical knowledge of acaricide application are contributing factors to tick resistance. For instance, Castro-Janer *et al.* (2010) in Uruguay and Vudriko *et al.* (2018) in Southern and Northwestern Uganda reported *Rhipicephalus appendiculatus*, *R.(B) decoloratus* and *R. (B.) microplus* to be tolerant to almost all chemical acaricides.

Despite the presence of several reports of acaricide resistance in different parts of the world (Castro-Janer *et al.* (2010)) ; Obaid *et al.* (2022); Githaka *et al.*, (2022). There is a paucity of knowledge on resistance status in Tanzania only a few studies have reported resistance of ticks against commonly used acaricides. Apart from the study of Kagaruki (1991) and Nagagi *et al.* (2020) which covered most regions of Tanzania and reported Organochloride resistance, there is limited information on the occurrence, distribution of tick species and the susceptibility status of commonly used acaricides in different parts of the country. The current study aimed at tracking the acaricide resistance in the Eastern zone of Tanzania to establish the list of acaricides that can be useful for the control of ticks and TBDs. This will contribute towards the rational use of acaricides and minimize costs associated with ineffectiveness.

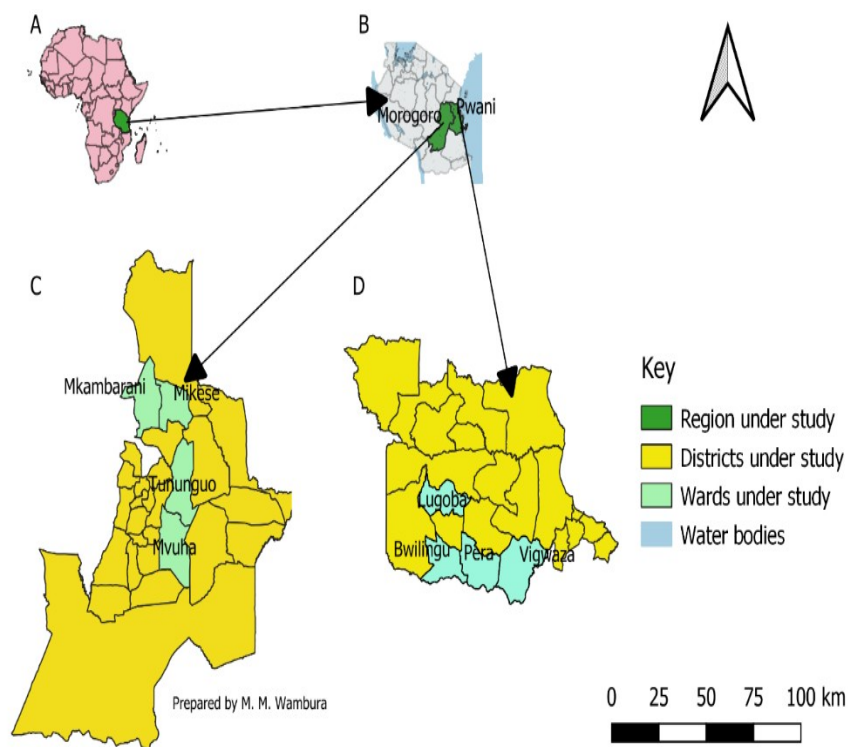
## **3.2 Materials and methods**

### **3.2.1 Description of the study area**

The study areas from which the tick samples were collected were the Morogoro and Pwani regions. Morogoro region is between latitude 5° C 58" and 10° C 0" to the South of the Equator and longitude 35° C 25" and 35° C 30" to the East. The region is bordered by the Tanga region

in the north, to the south by the Ruvuma region, to the west by the Iringa and Dodoma regions, and to the east by the Pwani and Lindi regions. Morogoro region is characterized by a tropical grassland climate having average temperatures of about 25 °C which vary from 18° C in high areas to 30° C in lowland areas. It is characterized by regular rainfall ranging from 600mm to 1200mm in low and high lands respectively . Pwani region is between Latitude: -7° 00' 0.00" S and Longitude: 39° 00' 0.00" E and it is bordered by Dar es Salaam and the Indian Ocean in the east, to the west by the Morogoro region, to the north by the Tanga region and to the south by the Lindi Region. Similarly, the climatic condition of the Pwani region is regarded as a tropical climate whereby temperature has a seasonal average temperature fluctuating from 13° C to 30° C and humidity as high as 98% (EPMS, 2006). Likewise, rainfall ranges between 800 – 1200 mm per annum with a bimodal rainfall pattern, a short rainy period from October to December, and a long rain season between March and June (Msuya *et al.*, 2013; Mushi, 2009; *United Republic of Tanzania Coast Region*, 2015). Generally, in both regions livestock sector is dominated by smallholders' livestock farmers, keeping mostly indigenous cattle, sheep, goats, pigs, and chicken. Apart from the livestock sector, these two regions deal with agricultural activities, forestry, fisheries, beekeeping, wildlife, and Industrial sectors (*United Republic of Tanzania Coast Region profile* 2015).

From each region, one district was selected namely Chalinze and Morogoro Districts. In each district, four wards were purposively selected depending on cattle population as indicated in Figure 3.1.



**Figure 3.1.** Map showing the distribution of eight wards from which the study was done 'A' represents the African continent, 'B' represents Tanzania, 'C' represents Morogoro district, and 'D' represents the Chalinze/Bagamoyo district

### 3.2.2 Study Design

A cross-sectional study design was used to collect ticks and a laboratory experimental design was adopted to evaluate the effectiveness of frequently used acaricides against F1 tick larvae from collected field tick species under controlled laboratory conditions.

### **3.2.3 Tick collection and identification**

Eight wards namely; Mvuha, Tununguo, Mikese, Mkambalani, Bwilingu, Lugoba, Vigwaza, and Pera were purposively selected. Furthermore, in each ward, three villages were selected based on cattle population only four households involved in cattle keeping were selected and at least four infested cattle per household were sampled. Cattle were restrained properly by owners and ticks were collected using small forceps starting from the head towards the tail and placed in a Petri dish for sorting. Collected ticks were handled carefully to avoid shedding of the legs and de-capitulation. None engorged ticks were transferred into plastic bottles with 70% ethanol and covered with plastic caps ( Nonga *et al.*, 2012), and the engorged ticks were put in 50 ml perforated plastic container placed on moisture cotton wool in the box with small holes to allow air circulation. All tick samples from one household (both engorged and non-engorged) were pooled together and labeled accordingly based on the household number, village, ward, and district collected. Thereafter, collected ticks were transported to the entomology laboratory at the College of Veterinary Medicine and Biomedical Sciences, Sokoine University of Agriculture for laboratory analysis. The collected ticks were identified to genus and species level based on morphological features using tick identification keys as described by Walker *et al.* (2014).

### **3.2.4 Tick Rearing**

One to three semi and full-engorged female ticks collected from similar locations or one herd were placed on one filter paper in a well-labeled Petri dish at optimum conditions which are 27 to 28°C and 75% to 85% Relative humidity in the BOD incubator. The eggs were then transferred in a small glass bottle with muslin cloth covering the mouth for hatching. The larvae were maintained under similar conditions in a BOD incubator until 12 to 14 days old (Kok, 2015; Shyma *et al.*, 2015).

### 3.2.5 Ticks acaricides-susceptibility Test

An in-vitro tick susceptibility test was done against eight (8) frequently used acaricides labeled as; Acaricide A (Amitraz 12.5%), acaricide B (Chlorpyrifos 100g/l + Cypermethrin 100g/l), acaricide C (Cypermethrin 15g/l), acaricide D (Alphacypermethrin 100g/l), acaricide E (Alphacypermethrin 10g/l), acaricide F (Chlorpyrifos 500g/l + cypermethrin 50g/l), acaricide G (Chlofenvinphos 300g/l) and acaricide H (Amitraz 125g/l). The test was done using tick larvae 12 to 14-day-old in packets prepared according to the Larval Packet Test (LPT) (FAO, 2004). The concentration used was a discriminatory dose as described by FAO (2004). Twice a normal dose (from the manufacturers' instruction guide) was prepared for each acaricide product using a diluent comprised of olive oil and acetone at a ratio of 1:2. Approximately, 0.7 mL of each prepared concentration was applied on one side of filter paper (Whatman number 1) (3.75 × 8.5 cm) in duplicate. Control packets were treated with diluent only. The filter papers were dried in an incubator at 37°C for 30 minutes. The dried filter papers were folded in half forming a rectangular packet with adhesive tapes sealing the sides forming an open-ended packet. Then, the open end was closed with adhesive tape after placing approximately 100 larvae and later placed in a BOD incubator that was maintained at 28°C and 75–85% relative humidity for 24 hours. After 24 hours, both dead and live tick larvae were counted. Tick larvae with the ability to move were considered life (Shyma *et al.*, 2015). The percentage mortality at each concentration was obtained after being corrected by Abbott's formula (Abbott 1925).

A total of 274 LPT tests/trials were performed to determine the susceptibility status of four different larvae from engorged tick species obtained from study areas namely *Amblyomma variegatum* (96 trials), *R. (Bo) microplus* (127 trials), *Rhipicephalus* spp (35 trials) and *Hyalomma rufipes* (16 trials) as shown in Table 3.1.

### 3.3 Data analysis

Data on the occurrence of tick species and percentage mortalities were recorded, sorted, and compiled using Microsoft Excel where appropriate percentages and frequencies were obtained. Descriptive results were presented in tables and figures. On the other hand, the IBM SPSS (Statistical Package for Social Science) version 20 was employed to analyze susceptibility status using generalized linear models (GLMs) with univariate analysis to compare the performance of acaricides of collected tick strains at 95 % confidence and p-value  $\leq 0.05$ .

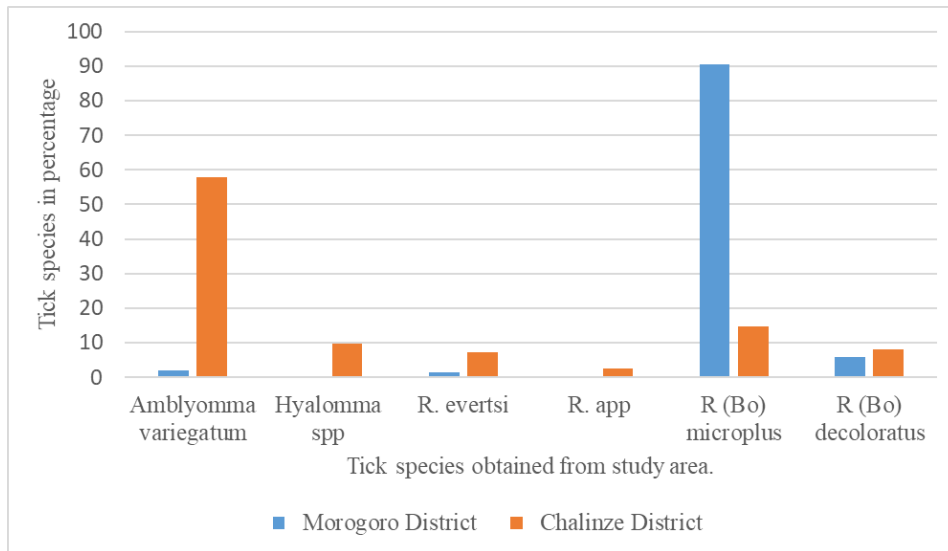
### 3.4 Ethical consideration

The Research Ethical clearance was approved by the Institutional review board of the Sokoine University of Agriculture with reference numbers SUA/DPRTC/R/126/VET/3/2021/4 and SUA/DPRTC/R/186/VOL. IV -7. Likewise, permission to conduct the study in the area was obtained from the Morogoro and Pwani regions with reference number AB.175/245/01/46 and FA.221/265/01F/102, respectively.

### 3.5 Results

#### 3.5.1 Occurrence of tick species

A total of 952 tick species were collected from the study area, *Rh. (Bo.) microplus* was the predominant species making a total of 552 (57.9%) followed by *Amblyomma variegatum* 247(25.9%), *Rh. (Bo.) decoloratus* by 65(6.8%) and *Hyalomma rufipes* 41(4.3%). The least species were *R. evertsi* and *R. appendiculatus* with 37(3.8%) and 10 (1.1%) respectively. More tick species 544 (57.1%) were encountered in Morogoro District compared to Chalinze District 408 (42.9%). Additionally, *Amblyomma variegatum* was the dominant species in the Chalinze District while *R. (Bo) microplus* species was predominant in Morogoro District as indicated in Figure 3.2.



**Figure 3.2:** Showing distribution of tick species in percentage from Morogoro and Chalinze districts

### 3.5.2 Susceptibility status

The mean percentage mortality of acaricide B, C, D, and E to *Amblyomma variagatum* was relatively low (less than 96%) from both Chalinze and Morogoro districts except acaricide B and E from Bwilingu and Tununguo wards showed 97±5.1 and 100 percentage mortality, respectively. However, acaricide A, F, H, and G all showed 100% mortality to *A. variagatum* from both districts. Also, the performance of acaricides B, C, D, and E to *Rhipicephalus (Boophilus) microplus* was relatively less than 96% with exception to *R. (Bo) microplus* from Mikese ward that showed 100% mortality to acaricides B, C, and D and E. Likewise, acaricide A, F, H, and G performed above 96% mortality with exception to acaricide A and H that showed 82±13% and 75±0.4% respectively from Mikese ward. On the other hand, most

of the acaricides percentage mortalities of *Rhipicephalus* spp were above 96% except acaricides C, D and E showed  $69 \pm 9.7$ ,  $81 \pm 7.8$  and  $90 \pm 7.6$  percentage mortality respectively for the species from Vigwaza, 87.2% and 77.8% mortality of acaricide C and D for the species from Tununguo. *Hyalomma rufipes* was the only species that was highly susceptible to almost all acaricides except acaricide D with a percentage mortality of  $79 \pm 29.9$  as shown in Table 3.1.

**Table 3.1** Mean Percent Mortality of tick species against a discriminatory dose of Acaricide B (Alphacypermethrin 100g/l + Chlorpyrophos 100g/l), Acaricide C (Cypermethrin 15g/l), Acaricide D (Alphacypermethrin 100g/l), Acaricide E (Alphacypermethrin 10g/l) Acaricides A(Amitraz 12.5%), Acaricide F(Chlorpyrifos 500g/l + Cypermetrin 50g/l), Acaricide H (Amitraz 125g/l) and Acaricide G (Chlorfenvinfos 300g/l).

Tick species	Place of Collection		Percentage Mortality (%) (Mean ± STDEV %)								There are no Tests per tick species
	District	Ward	Acaricide B	Acaricide C	Acaricide D	Acaricide E	Acaricide A	Acaricide F	Acaricide H	Acaricide G	
<i>A. variegatum</i>	Chalinze	Bwilingu	97±5.1	73±38.0	88±14.4	92 ± 6.0	100.0	100.0	100.0	100.0	96
		Lugoba	84 ±8.1	79 ±17.3	89 ±22.5	88±21.5	100.0	100.0	100.0	100.0	
		Vigwaza	92±10.6	85 ±12.6	80 ±17.2	86 ± 9.7	100.0	100.0	100.0	100.0	
<i>Boophilus</i> species	Morogoro	Tununguo	94.0	77.6	87.1	100.0	100.0	100.0	100.0	100.0	127
		Chalinze	Bwilingu	97 ±6.9	89 ±11.3	91 ±12.6	91± 8.2	100.0	100.0	100.0	
	Morogoro	Lugoba	89 ±15.5	75 ±36.1	63 ±34.0	77 ±25.3	100.0	100.0	100.0	100.0	
		Vigwaza	96±5.6			89 ± 9.8	100.0	100.0	100.0	100.0	
		Mikese	100 ±0.0	100.0	100.0	100 ±0.0	82±13	100.0	75 ±0.4	100.0	
<i>Rhipicephalus</i> spp	Chalinze	Vigwaza	99 ±1.2	69 ±9.7	81 ± 7.8	90 ± 7.6	98±4.9	100.0	100.0	100.0	35
		Morogoro	Tununguo	100. ±0.0	87.2	77.8	97.9	100.0	97.1	100.0	
<i>H. rufipes</i>	Chalinze	Vigwaza	100 ±0.0	96 ±5.6	79 ±29.9	95 ± 7.5	100.0	100.0	100.0	100.0	16

### 3.5.3 Overall ranking of tick susceptibility status

Ticks species that showed at least 96% mortality were considered not resistant whereas those that showed between 80-95 % mortality were considered susceptible while those that showed between 60-80 % mortality against a given acaricide were considered resistant (Vudriko *et al.*, 2016; Yawa *et al.*, 2022). The percentage mortality of tick species against different acaricides was classified into four categories as shown in Table 3.2. The least performers (Category 1) comprised acaricides C and D with mean percentage mortality (%) of 84.60 and 85.72 respectively, and the highest performers (Category 4) acaricides F and G with percentage mortalities of 99.83% and 99.90 % respectively.

**Table 3.2:** Overall ranking of tick susceptibility status against used acaricides in the surveyed areas

Acaricide type	Given Name	N	Percentage Mortality (%)				P value P ≤ 0.05
			Category 1	Category 2	Category 3	Category 4	
Cypermethrin 15g/l	Acaricide C	34	84.5976				a, b, f, g, h
Alphacyp. 100g/l	Acaricide D	35	85.7286				a, f, g, h
Alphacyp 10g/l	Acaricide E	34		91.2938			a, f, g, h
Alpha100g/l + Chlorp 100g/l	Acaricide B	34		93.9012			a, c, f, g, h
Amitraz 125g/l	Acaricide H	34			97.7609		b, c, d, e
Amitraz 12.5%	Acaricide A	34			97.8894		b, c, d, e
Chlor 500g/l + Cyp 50g/l	Acaricide F	35				99.8251	b, c, d, e
Chlorfen 300g/l	Acaricide G	34				99.9029	b, c, d, e

Acaricide types with capital letters A, B, C, D, E, F, G, and H are statistically significantly different with one or more acaricides labeled with their corresponding small letters (a, b, c, d, e, f, and g).

### 3.4 Discussion

The information on tick species distribution and resistance status of a particular area is an important part of controlling ticks and TBDs. In the current study, six tick species were found namely *Rhipicephalus (Boophilus) microplus*, *Rh. (Bo) decoloratus*, *Rh. evertsi*, *Rh. appendiculatus*, *Amblyomma variegatum* and *Hyalomma rufipes*. These findings corroborate with what was reported by (Mamiro *et al.*, 2016; Nagagi *et al.*, 2020) who reported the presence of all these tick species from different parts of Tanzania. While *Amblyomma variegatum* was the predominant tick species in the Chalinze District, the *Rhipicephalus (Boophilus) microplus* was found in the Morogoro and Chalinze districts. The findings from this study are contrary to what was reported by (Mamiro *et al.*, 2016) where *A. variegatum* was the most abundant species by (56.6%) followed by *Rh. (Bo) microplus* by (27.3%). On the other hand, other species such as *Hyalomma rufipes*, *Rh. Appendiculatus*, *Rh. evertsi* and *Rh. (Bo) decoloratus* were encountered at a relatively low percentage in all districts. The distribution of these species depends on different factors such as a climatic condition that favor certain vegetation, host availability, and tick and TBDs tolerant breeds (Tatchell & Easton, 1986; Laisser *et al.*, 2015; Chenyambuga *et al.*, 2010).

In addition, human factors such as the movement of infested livestock from one area to another for trading and search of pastures especially during drought season as well as poor knowledge of acaricide application might influence the tick distributions in different areas. This finding corresponds to Vudriko *et al.* (2016) who found resistant *Rh. appendiculatus* collected from cattle bought from different parts of Uganda for slaughter.

In the experimental acaricides, the results of susceptibility tests of different acaricide groups against tick species from different areas indicate that acaricides with classes; H (Amitraz 125g/l), A (Amitraz 12.5%), F (Chlorfenvinfos 500g/l + Cypermethrin 50g/l) and G (Chlorfenvinfos 300g/l) performed above 96%. These results indicate that these groups of acaricides are effective in the control of ticks and TBDs in those areas as most ticks have not developed resistance against them. However, acaricide A (Amitraz 12.5%) and H (Amitraz 125g/l) were less effective against *R. (Bo) microplus* from Mikese ward in Morogoro district this may be due to acaricides with amitraz molecule being frequently used in Mikese ward compared to other acaricide classes as exhibited by Abbas *et al*, (2014) who stated that frequent acaricide application is a risk factor for the emergence of resistant tick strains. Generally, acaricide classes C (Cypermethrin 15g/l), D (Alphacypermethrin. 100g/l), E (Alphacypermethrin 10g/l), and B (Alphacypermethrin100g/l + Chlorpyrifos 100g/l) were less effective to almost all subjected tick species, having percentage Mortality of 84.6%, 85.7%, 91.3% and 93.9% respectively. This finding corresponds with a previous report (Vudriko *et al.*, 2016) which stated that most of the tick species tested had resistance to at least one class of acaricide molecule in Uganda. In addition, the acaricides which were reported by farmers to be repeatedly used and applied for a long period showed less efficacy in the susceptibility test than those which were not frequently used due to either being recently introduced in the market or being too expensive for the farmer to buy (Mwita *et al.*, 2023 unpublished data). This corresponds to the report of Obaid *et al.* (2022) which stated that the repeated use of a single acaricide is often responsible for the selection of fast-developing acaricide resistance.

On the contrary, the Formamidines class acaricide H (Amitraz 125g/l) and A (Amitraz 12.5%) were among the acaricide groups that had a high mortality rate above 98% mean mortality rate although, it was mentioned by the farmers as widely used and less effective during

questionnaire survey (Mwita *et al.*, 2023 unpublished data). This result suggests that for ticks to develop resistance against amitraz it might involve complex mechanisms which are multi-factorial. Indeed the study of de La Canal *et al.* (2021) suggested that amitraz resistance in the *R. microplus* strain resulted from multiple factors including cytochrome P450 detoxification and mutation in the octopamine receptors while Li *et al.* (2004) suggested target site insensitivity as the mechanism responsible for amitraz resistance instead of a detoxification mechanism.

Based on susceptibility test results, the acaricide derived from Synthetic Pyrethroid (SP) molecules (Cypermethrin and alphacypermethrin) showed the emergence of resistance to developing resistance status compared to acaricides formulated from Formamidines (Amitraz) and OP (Chlorpyrifos and Chlorfenvinfos) that showed susceptible status to almost all tick species. However, this study was in disagreement with work reported by Hezron *et al.*, (2012) that a cypermethrin with 10% w/v was 99% effective in killing multi-host ticks after treatment.

The resistance of these tick species against SP (Alphacypermethrin and Cypermethrin) can be due to the spread of resistant strains of ticks through livestock movement or trade as well as the use of the same SP and coformulation (CoF) for prolonged periods and acaricide misuse adopted by livestock farmers. In addition, insecticides from agriculture with similar classes to acaricides increase selection pressure which is usually regarded as an early driver of acaricide resistance in tick control programs (Diabate *et al.*, 2002; Hien *et al.*, 2017).

The current study found that SPs ( Cypermethrin 50g/l ) had a high level of resistance compared to AM (amitraz) and OPs (Chlorfenvinfos and Chlorpyrifos) although, when combined with OPs (Chlorpyrifos) appeared to increase the tick mortality rate as

also reported by Obaid *et al.*(2022) and Thullner *et al.*(2007) that the combination of OPs (Chlorpyrophos) with SPs (cypermethrin) has been successfully applied for the control of *Rh. microplus* in different parts of the world. However, Shyma *et al.*(2015) suggested that, for those acaricides that are becoming ineffective the manufacturer-recommended dose should be revalidated.

Finally, acaricide G (Chlorfenvinphos 300g/l) performed best however this is contrary to a recent report (Githaka *et al.*, 2022; Nagagi *et al.*, 2020; Vudriko *et al.*, 2016) which confirmed resistance of OP (chlorfenvinphos) in different parts of Tanzania. The effectiveness of Chlorfenvinphos in study areas of Morogoro and Chalinze districts may be due to the high cost that limits their availability in the free market hence less used as also described by livestock keepers during sample collection.

### **3.5 Conclusion**

This study found six tick species namely *Rhipicephalus (Boophilus) microplus*, *R (Bo) decoloratus*, *R. evertsi*, *R. appendiculatus*, *Amblyomma variegatum* and *Hyalomma rufipes* that are responsible for the transmission of TBD pathogens. However, the predominant tick species in the Chalinze district was *A. variegatum* while *Rh. (Bo). microplus* was the overall predominant species in the study area. Nevertheless, the susceptibility status of four tick larvae species that are *A. variegatum*, *R (Bo) microplus*, *Rhipicephalus spp*, and *H. rufipes* against eight commonly used acaricides has been documented such that the acaricides with reduced effectiveness particularly synthetic pyrethroids; Cypermethrin 15g/l, Alphacypermethrin 100g/l,

Alphacypermethrin 10g/l and (Alphacypermethrin100g/l + Chlorpyrifos 100g/l) highlights the need for careful acaricide selection, rotation strategies, combining acaricide classes and including revalidation of

manufacturer-recommended doses may enhance acaricide effectiveness while acaricide with ingredients Chlorpyrifos 500g/l + Cypermethrin 50g/l and Chlorfenvinphos 300g/l are encouraged to be used in the study area due to their excellent effectiveness.

### **Recommendations**

Regular monitoring of tick resistance development against different acaricides plays an important role in slowing down resistance selection. Therefore the responsible authority should make sure that the livestock owners and workers are educated on acaricide application and tick control practices in general. Cost-effective control strategies should be implemented to reduce losses caused by ticks and TBDs. On the other hand, the government should encourage good land use and pasture management and implement a good policy on the traditional movement system of animals as well as subsidize acaricides to lower their cost allowing the farmers to have an equal chance of buying any acaricides he /she want. Finally. Since experimental study results exhibit the presence of acaricide resistance, the authors recommend further study that will cover a larger area for confirmation of the problem and enhancing specific measures to control it.

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**Conflict of interest**

The authors declare that they have no conflict on the matter of publication and this document is their original work done in the Eastern zone of Tanzania and no part of it has been submitted somewhere else for publication or conference presentation.

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## CHAPTER FOUR

### 4.0 SUMMARIZING DISCUSSION, CONCLUSION, AND RECOMMENDATIONS

#### 4.1 Summarizing Discussion

The study shows general awareness of livestock farmers, distribution and resistance status of ticks and TBDs affecting cattle, and the control methods practiced in the eastern part of Tanzania. The findings show that fenced shelter and free-range grazing were the main housing and feeding mode practiced by the farmers respectively however, few were using houses in line with supplements to their animals where this finding is similar to that reported by (Baltussen *et al.*, 2020; Kavana *et al.*, 2005). The study revealed different constraints to farmers such as the presence of counterfeit and high-cost acaricides, tick infestation, and lack of dipping sites that collectively account for an increase in vectors for transmission of disease pathogens to cattle as also reported by (Jaime Betancur Hurtado & Giraldo-Ríos, 2019; Nagagi *et al.*, 2020). Similarly, the farmers had moderate knowledge about ticks whereby, some of the respondents used key features of ticks to identify them for example 27.9% of respondents described *Rhipicephalus (Boophilus)* spp as big and blue, while red and small in size representing *Rhipicephalus* genera by (29.5%) of respondents and *Amblyomma* spp described as colored tick by 28.7% of respondents. These findings were confirmed by laboratory findings whereby six tick species were identified namely *R. (Bo) microplus*, *R. evertsi*, *R. appendiculatus*, *R. (Bo) decoloratus*, *H. rufipes*, and *A. variegatum*. These findings are consistence with the findings by (Walker *et al.*, 2014; and Nagagi *et al.*, 2020) who reported the presence of all these tick species from different parts of Tanzania. Nevertheless, the availability of these tick species conforms to the types of TBDs listed by respondents as also reported by (Kerario *et al.*, 2017; Isack *et al.* 2017). Generally, the livestock

farmers had a poor understanding of TBDs with an exceptional few respondents who were familiar with *Anaplasmosis* by 8 (6.2%), ECF by 22 (17.1%) and *Babesiosis* by 9 (7.0%) whereby similar results were reported by (Laisser *et al.*, 2014; Jacob *et al.*, 2017). This points out that even though respondents were able to mention different diseases including TBDs affecting their cattle, they were not aware of which diseases are transmitted by ticks. The persistent use of acaricides is the main reason that hastens resistance selection in ticks (Abbas *et al.*, 2014). It has been reported that there is a relationship between either overdosing or dosing on acaricide and the degree of resistance as revealed by Vudriko *et al.* (2018) and Lees *et al.* (2013) that a species controlled by a recommended dose of acaricide may show resistance when an under dose of the same acaricide is applied. The commonly used acaricide application method was hand spray however is less efficient as most farmers did not have special structures for immobilizing the animals during the acaricide application as well as awareness of acaricide composition, rotation and combination technique between different chemical classes making it difficult to achieve thorough acaricide application (Githaka *et al.*, 2022).

The susceptibility test observed that acaricide classes OPs (acaricide G = Chlorfenvinfos 300g/l), amidine (acaricide A = Amitraz 12.5% and acaricide H = Amitraz 125g/l), as well as a combination between SP and OP (acaricide F = (Chlorfenvinfos 500g/l + Cypermethrin 50g/l)), performed better (above 96% mortality rate) compared to SP chemical. These results point out that these groups of acaricide classes are effective in controlling ticks and TBDs in their respective areas. Although amidine class (Amitraz) was mentioned by the respondents as a less effective and widely used acaricide however, the study of De La Canal *et al.* (2021) suggested that the susceptibility status of amitraz in *R. microplus* strain resulted from multiple factors including cytochrome P450 detoxification and mutation in the octopamine

receptors while *Li et al.* (2004) suggested target site insensitivity as mechanism responsible for amitraz resistance instead of detoxification mechanism. On the contrary, acaricide A (Amitraz 12.5%) and H (Amitraz 125g/l) showed low mean mortality rates against *R. (Bo) microplus* species from the Mikese ward in Morogoro district this may be due to amitraz persistently and frequently used in Mikese ward compared to other acaricide classes as also reported by *Abbas et al.*, (2014) that frequent acaricide application is a risk factor for the emergence of resistant tick strains. This corresponds with *Obaid et al.* (2022) who stated that the continual use of a single acaricide class is usually responsible for the selection of emergence and development of acaricide resistance. In general, acaricide with class SPs (acaricide D = (Alphacypermethrin. 100g/l), acaricide C = (Cypermethrin 15g/l), acaricide B = (Alphacypermethrin100g/l + Chlorpyrifos 100g/l) and acaricide E =(Alphacypermethrin 10g/l) showed low mortality rate to almost all subjected tick species, having percentage mortality of 85.7%, 84.6%, 93.9% and 91.3% respectively.

However, this study was in disagreement with work reported by *Hezron et al.* (2012) that a cypermethrin with 10% w/v was 99% effective in killing multi-host ticks after treatment. The resistance of these tick species against SP (Alphacypermethrin and Cypermethrin) may be due to the spread of tick-resistant strains through livestock movement for searching pasture, water, and trading purposes as well as the use of the same SP acaricide and coformulation (CoF) for a prolonged period and acaricide misuse adapted by livestock farmers. However, *Shyma et al.* (2015) suggested that, for those acaricides becoming ineffective the manufacturer-recommended dose should be revalidated.

Generally, acaricide G (Chlorfenvinphos 300g/l) performed best however this is contrary to recent reports (*Githaka et al.*, 2022; *Nagagi et al.*, 2020; *Vudriko et al.*, 2016) confirmed resistance of OP

(chlorfenvinphos) in different parts of Tanzania. The effectiveness of Chlorfenvinphos in the study area may be due to high costs that limit their availability in the free market hence less used as also described by livestock keepers during sample collection. Finally, acaricide F (combination of Chlorpyrophos 500g/l + Cypermethrin 50g/l) was successfully applied to control all tick species under study as also reported by (Obaid *et al.*, 2022; Thullner *et al.*, 2007) that the combination of OPs (Chlorpyrophos) with SPs (cypermethrin) has been successfully applied for the control of *R. microplus* in different parts of the world.

#### **4.2 Conclusion**

The present study shows that ticks and TBDs are the most common livestock production constraints identified by farmers where East Coast fever (ECF) was ranked second after CBPP followed by tick infestation. On the other hand, the farmers had little knowledge of tick species and TBDs showing that most of the respondents were not aware of the causes of TBDs however, the study found six tick species namely *Rhipicephalus (Boophilus) microplus*, *R. evertsi*, *R. appendiculatus*, *R. (Bo) decoloratus*, *Amblyomma variegatum*, and *Hyalomma rufipes* though, *A. variegatum* was predominant tick species in Chalinze district while the overall predominant tick species in the study area was *R. microplus* and that all species are responsible for the transmission of TBD pathogens.

The most common tick control method used by farmers was acaricide application through hand spray whereby sixteen acaricides were used also the study revealed malpractices such as persistent use of acaricide at once-per-week intervals, overdosing as well as lack of dipping sites may increase the risk for emerging and development of acaricide resistant tick species. Indeed the livestock keepers listed some acaricides that they thought to be resisted by ticks whereby

acaricide D (Alphacypermethrin 100g/l) ranked high followed by acaricide E (Alphacypermethrin 10g/l). Likewise, the experimental results concurred with farmer's doubt whereby, the susceptibility status of four tick larvae species has been documented to determine which acaricides are effective to be used and those to be considered out of use as a result, the tick control program will be successfully managed in Tanzania.

### **4.3 Recommendation**

This study has revealed that there are malpractices in acaricide usage and low knowledge of TBDs. Regular monitoring of tick resistance development against different acaricides plays an important role in slowing down resistance selection. Therefore the responsible authority should make sure that the pastoralists, agro-pastoralists, and workers are educated on acaricide application and tick control practice in general. as well as constructing dipping sites and cost-effective control strategies should be implemented to reduce losses caused by ticks and TBDs. On the other hand, the authority should implement a good policy on the animal movement system, encourage good land use, and pasture management, and as well as subsidize acaricides to lower their cost so the farmers have an equal chance of buying any acaricides he /she want. Lastly, more studies should be conducted to confirm the tick resistance problem and come up with specific measures to be taken.

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## APPENDICES

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### **Kuhusu Tasnifu Hii**

Kupe, ni wadudu wanaohusika na kueneza magonjwa kwa ng'ombe, huleta changamoto katika kudhibiti licha ya njia inayopendekezwa kuwa viuatilifu (acaricides). Utafiti huu ulilenga kutathmini uelewa wa wafugaji kuhusu magonjwa yaenezwayo na kupe (TBDs) na ukinzani wa viuatilifu Mashariki mwa Tanzania. Wafugaji wapatao 129 walihojiwa na jumla ya kupe 952 zilikusanywa na kujaribiwa dhidi ya viuatilifu vinane vilivyoonekaka kutumika sana katika eneo la utafiti huu.. Matokeo yalionyesha uelewa mdogo wa magojwa yanayoenezwa na kupe miongoni mwa wakulima. Kunyunyizia kwa mikono (hand sprayer) ndiyo njia iliyopendekezwa na wafugaji kwa ajili ya udhibiti wa kupe na magonjwa yanayoenezwa na kupe kwa asilimia 89.1. Miongoni mwa aina za kupe waliopatikana, *R. (Bo) microplus* ilipatikana kwa wingi (57.9%) ikifuatiwa na *Amblyomma variegatum* kwa asilimia 25.9. Uwezo wa kupe kuathiriwa na viuatilifu ulitofautiana kutoka kiuatilifu kimoja na kingine, ikiangazia makosa yanayochangia kushindwa kwa viuatilifu hivyo ni pamoja na uchanganyaji mbaya wa kiwango cha kiuatilifu husika, kutumia kiuatilifu cha aina moja kwa mda mrefu, mchanganyiko wa dawa kushindwa kufika sehemu zote za mwili wa mnyama. Matokeo haya yanatoa nafasi muhimu kwa ajili ya kuandaa programu zinazolengwa kama vile kuelimisha wakulima kupitia maafisa mifugo ili kukabiliana na magonjwa yanayoenezwa na kupe na kudhibiti ukinzani wa viuatilifu katika jamii za wafugaji.