

**PREVALENCE AND THE EFFECT OF PARASITES ON HAEMATOLOGICAL
PARAMETERS OF LIVESTOCK IN THE UPPER KERIO VALLEY, KENYA**

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DECLARATION

DECLARATION BY THE CANDIDATE

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DEDICATION

This work is dedicated to livestock farmers in Kerio Valley who struggle each day to ensure that their animals are well fed, in good health and protected despite the harsh environmental conditions and high insecurity including cattle rustling.

ABSTRACT

Livestock remains the mainstay of many communities in Kenya yet its production has been negatively impacted by animal diseases especially parasitic infections. Vector borne haemoparasites and intestinal parasites cause major livestock losses and devastate many livelihoods especially in Africa where majority of people depend on agriculture and livestock farming as source of food and income generation. A cross-sectional study was carried out to obtain baseline information on prevalence of tick-borne haemoparasites, trypanosomes and gastrointestinal parasites and their effects on haematological parameters of livestock in the upper Kerio Valley of Elgeyo-Marakwet County. The study also sought to identify ectoparasites and transmitting vectors of haemoparasites. A total of 468 livestock comprising of cattle, sheep and goats in randomly sampled farms were investigated during the period between May 2016 and July 2017. The impact of the infections was also examined through haematological analysis of the blood samples. Determination of gastrointestinal parasites infections was done by flotation and sedimentation methods. The overall prevalence of helminths in cattle, sheep and goats were 59.9%, 60.1% and 79% respectively. Adult cattle were more likely to be affected by helminth parasites than the young animals. Goats were found to have higher prevalence of Coccidian infections than the sheep and cattle (48%, 32% and 39%) respectively. PCR method identified 29 (32%, N= 90) livestock infected with *Trypanosoma brucei* and *T. congolense*. The results indicated low prevalence of *Theileria parva* (4%), *Anaplasma* (2.8%) and *Babesia* (1.4%). Intestinal nematodes mainly *Strongyloides species* accounted for most infections in livestock however tick-borne parasites showed low prevalence. Nematodes and trematodes infections effect on blood values proved significant at $p < 0.05$. Infection with nematode parasites in sheep significantly ($p < 0.05$) affected WBC, lymphocytes, monocytes, granulocytes, RBC, HGB, MCH, MCHC, platelets and MPV. Although parasite infection affected blood values in cattle and sheep, the changes were not significant. In cattle, WBC (mean 15.10, and MPV (mean 8.9) were significantly higher than normal standard reference values. Age, sex, location and parasitic infections were found to influence blood parameters of livestock in this setting. High prevalence of helminth infections calls for the need to strengthen control strategies and measures that reduce parasite burden for sustainable animal production in Kerio Valley. Haematological parameter ranges in livestock in Kerio Valley established in the study can be utilized in evaluating physiological status of indigenous and mixed breeds in semi-arid areas of Kenya. It is important to determine how other factors such as environment and management systems practiced by livestock owners influence the presence of parasitic infections.

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LIST OF ABBREVIATIONS, ACRONYMS AND SYMBOLS

AAT	African Animal Trypanosomosis
CGEM	County Government of Elgeyo-Marakwet
DNA	Deoxyribonucleic Acid
EDTA	Ethylene-diamine-tetra-acetate
FTA	Flinders Technology Associates
GINs	Gastrointestinal nematodes
GoK	Government of Kenya
Gran	Granulocytes
HCT/PCV	Haematocrit/Packed cell volume
HGB	Haemoglobin
IPCC	Intergovernmental Panel on Climate Change
LYM	Lymphocytes count
MCA	Multiple correspondence analysis
MCH	Mean corpuscular haemoglobin
MCHC	Mean corpuscular haemoglobin concentration
MCV	Mean corpuscular volume
ml	Millilitres
mm	millimetres

MoLD	Ministry of Livestock Development
Mon	Monocytes
MPV	Mean Platelet Volume
NACOSTI	National Commission of Science, Technology and Innovation
PATTEC-KC Council	Pan African Tsetse and Trypanosomiasis Eradication Campaign - Kenya
PCR	Polymerase Chain Reaction
PLT	Platelet
RBC	Red blood cells
RNA	Ribonucleic Acid
WBC	White blood cells
μL	Microlitres

OPERATIONAL DEFINITION OF TERMS

Epidemiology: the science that describes and explains patterns of disease in the host population (i.e. the distribution and determinants of disease).

Final (or definitive) host: a term used to identify the host in which sexual reproduction of the parasite takes place.

Identification: it is a deductive process of determining which taxonomic groups a species belongs to.

Intermediate host: this is a host in which only immature stages grow and develop. Asexual replication may occur (but not sexual reproduction)

Livestock: refers to cattle, goats and sheep

Pathogen: an organism that causes disease

Pathogenesis: the mechanism of the disease process.

Prevalence rate of a problem – Proportion of the population with the problem measured at a point in time

Reservoir host: this depicts a host population that acts as a source of infection for other animals.

Resilience: this is the ability of an animal to tolerate the presence of parasites. The capability of an animal to limit, or to compensate for, the damage caused by the parasites.

Resistance: this is the ability of an animal to defend itself against parasitic attack by means of innate and acquired immune responses. The main methods of enhancing host resistance are by ensuring adequate nutrition and by vaccination.

Stratified sample: A probability sampling design in which the population is first divided into homogenous strata within each of which sampling is conducted (Kothari, 2005)

Subclinical: a term used to describe a degree of parasitism, which interferes with production but is not evident by physical and visual examination of the animal. In practice, it is a state of parasitism usually diagnosed by a positive production response to the administration of an anthelmintic.

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CHAPTER ONE

INTRODUCTION

1.1 Background of the Study

Animal parasitic infections are major threats to livestock production and maintenance in sub-Saharan Africa. Livestock in sub-Saharan Africa are most likely to be infected with vector-borne haemoparasites and gastrointestinal parasites (Bell-Sakyi, 2004) due to favourable conditions for the survival of parasites and poor farm management practices. Trypanosomosis, tick borne diseases and gastrointestinal parasites negatively affect livestock production particularly in East Africa where vectors are endemic and where conditions for intestinal parasites are favourable (Uilenberg, 1995; Minjauw & McLeod, 2003).

In the Kenyan economy, the livestock sector contributes 12% of the Gross Domestic Product (GDP) and 40% of agricultural GDP (Ministry of Agriculture, Livestock, Fisheries and Irrigation-- Kenya--MALFI, 2019). As stated in the Ministry's strategic plan (MoLD, 2010), livestock plays an important role in Kenya's socio-economic development and contributes towards household food and nutritional security especially among the rural communities.

Indigenous and mixed breeds of livestock are important regionally with cattle constituting 77% of the total Kenyan livestock population (Rege, Kahi, Okomo, Mwacharo and Hanotte, 2001). Indigenous breeds are usually preferred by farmers living in semi-arid and arid areas due to their adaptability to heat, water stresses and food scarcity, and they have good disease resistance (Mwacharo & Drucker, 2005).

Among the livestock diseases, trypanosomosis is disease with major health impact, affecting livestock production in 37 countries within the Sub-Saharan region (Thumbi, Jung'a, Mosi, & McOdimba, 2010). It is estimated 45 – 50 million cattle are at risk of infection in the region with an estimated economic loss of approximately US \$ 1.3 billion in cattle production (Kristjanson, Swallow, Rowlands, Kruska, Leeuw, 1999). It is a disease of both humans and animals caused by different species of trypanosomes that are transmitted by various sub-species of tsetse flies. *Trypanosoma* species cause African Human Trypanosomosis (HAT) and African Animal Trypanosomosis (AAT) resulting in public health problem and considerable losses in livestock production and morbidity (MoLD, 2010, and MoLD, 2011).

Trypanosomes infecting animals are genetically diverse including *Trypanosoma vivax*, *T. congolense* and *T. brucei* in the East African region. *Trypanosoma vivax* and *T. congolense* are predominant throughout sub-Saharan African and they are the major causes of AAT (Isaac *et al.*, 2016). *Trypanosoma brucei* constitute two subspecies namely, *T. brucei brucei* and the zoonotic *T. brucei rhodesiense*. They co-circulate between livestock and humans (Bronsvort, 2010), which, if not controlled can become a public health problem.

Tick infestations in livestock on the other hand cause economic losses especially to poor-resource populations, not only through destruction of animal skin and sucking of blood but also transmit a number of pathogenic infections to their hosts (Rehman, Nijhof, Sauater-louis, Shauer, Stauback, & Conraths, 2017). The most important tick borne infections of livestock in Africa include East Coast Fever (ECF), Babesiosis Anaplasmosis and Heart-water (Uilenberg, 1995; Sonenshine & Mather, 1994). These diseases cause losses in terms of lowered production rates, mortalities, decreased

reproduction rates and high expenditures on treatment of animals and control of infections (Van Wyk, Goddard, Bronsvort, Coetzer, Booth, 2013).

Gastrointestinal parasitism in livestock is equally a major impediment in animal production (Van Wyk *et al.*, 2013) due to the high cost of management in terms of treatment of disease and control measures. Gastrointestinal helminths infections are known to cause wide range of immunological responses, affect nutrient absorption and water balance, which then result in low body composition, poor carcass quality, reduced animal productivity and performance (Fox, 1993; Gasbarre, 1997; Government of Kenya, 2010; Bradan, Abuamsha, Aref, Algisi, and Alumor, 2012). Farm animals suffer considerably from heavy infections of gastrointestinal helminths particularly nematodes due to migration of their larval stages and the feeding by the adult worms (Odoi, Gathuma, and Omore, 2007).

Since *parasitic infections are detrimental to the economy of livestock production, effective* control methods for vector-borne infections and parasitic helminths calls for clear understanding of the occurrence, prevalence of vector-borne infections and gastrointestinal among ruminant livestock. In previous works, Masiga, Okech, Wekesa & Ouma (2002), Machila, McDermott, Welburn, Maudlin & Elsler, (2003), Bronsvort, (2010), Thumbi, Jung'a Mosi, McOdimba, (2010), spatial distribution and prevalence of animal trypanosomosis have been determined mostly in western Kenya, the coast region and other areas such as south of Rift Valley but little information is available for the Kerio Valley. Helminth parasites are equally a serious problem in developing world, especially where nutrition and sanitation are poor and environmental conditions favour survival of helminths. Parasites have varied consequences on health of infected animal such as haematological parameters. Reference ranges for haematological parameters are important in livestock production and have

widely been used in management of animal health, understanding disease prognosis and therapeutic monitoring (Al-Bulushi, Swawaf, & Al-Hasani, 2017). Different animal species and breeds are known to have unique standard values of haematological parameters (Shawaf, Hussein, Al-zoubi, Hamaash, & Al-busadah, 2018). This requires knowledge about breed specific blood values for animals in each niche so as to obtain baseline haematological values that can be used in evaluation of animal health and disease under similar climatic and managerial conditions.

This study sought to investigate vector-borne and gastrointestinal infections in livestock and to identify vectors associated with suspected haemoparasites in Kenyan upper Kerio Valley of Elgeyo-Marakwet County. The study furthermore aimed at providing information about the prevalence, characterization of trypanosomes species causing AAT and their co-infections with tick borne and gastrointestinal helminths infections along upper Kerio Valley, Kenya.

1.2 Statement of the Problem

The major threat to livestock production and maintenance especially in sub-Saharan Africa is occurrence of diseases caused by parasites. With the world's increasing population, there is need to expand agricultural production to maintain food security, in terms of meat and milk production. Major animal diseases can have significant economic, social and human health effects in the form of negative effects on the markets, loss of production and farm income, and community losses in terms of social welfare, and some internal parasites are emerging zoonotic infections. The earth is warming up and climate change is happening and this is likely to lead to the emergences and re-emergence of infectious diseases. The tropical climate coupled with poor parasite management strategies at the farm level favour survival and maintenance of various types of pathogens

and parasite vectors. The most important livestock diseases in Africa are helminthosis, intestinal protozoa and vector-borne infections including East Coast Fever, Babesiosis, Anaplasmosis and African Animal Trypanosomosis. These diseases cause animal production losses in terms of reduced production rates, mortality, expenditure in treating and controlling these infections.

Helminth parasites are prevalent and a major limitation to cattle production in many parts of Kenya and some intestinal worms such as *Trichostrongylus* cause impaired productivity. Some parasitic infections such as those caused by trypanosomes and intestinal parasites are confounded in cases where the species co-circulate between animals and human beings, creating public health problems among people living in affected areas..

Tsetse flies are widely distributed in Kenya and are known to correlate with the location of wildlife conservancy where the wild animals act as source of blood meal for tsetse flies and reservoir for trypanosomes and where there is high poverty prevalence. The presence of Rimoi National Reserve in Kerio Valley may contribute to the spread and transmission of trypanosomes since wildlife play a vital role in maintenance of tsetse flies through provision of blood meal and a reservoir for the parasites that can be easily transmitted to domestic animals. A key feature for trypanosomes is variability in types and distribution resulting in the differences between different ecological areas and farming systems and therefore the importance of their characterization so as to develop suitable control programmes.

Ticks are important ectoparasites and vectors to livestock diseases continue to cause significant health threats to livestock by causing direct damage to the skin and

transmitting parasites such as *Theileria*, *Babesia* and *Anaplasma* species. Different species of ticks are adapted to different ranges in temperature and moisture and some are even active in dry climates such as those found in Kerio Valley. Their survival especially in eastern African is contributed by favourable climate, inappropriate control measures and poor management systems.

With regard to livestock health and impact of parasite infections, little is known on the effect of parasitic infections and other factors on haematological parameters, .therefore the information obtained from the haematological tests together with diagnostic methods such as physical examination would be useful in making health decision. The results on the types of *Trypanosoma* species present in Kerio valley indeed are valuable in mitigation and estimating the size of human infective parasites' reservoir among livestock.

The study was aimed to understand the transmission dynamics of livestock parasitic diseases, their prevalence and their effects on haematological parameters. There is lack of information on normal haematological parameter ranges of indigenous livestock breeds kept under free-range management in semi-arid areas, or similar conditions such those found in Kerio Valley. Haematological profiles obtained can be used to evaluate health status of livestock and also act as reference blood values, an important aspect in management of livestock.

1.3 Justification of the Study

Despite great progress in control of livestock diseases such as vector-borne and gastrointestinal infections by the Ministry of Livestock and individual farmers, they remain health threats to animal production. Livestock diseases have increased in the past decade resulting in high mortalities, reduced herd sizes and increase in the cost of

production in terms of treatment and control of animal diseases (Sang *et al.*, 2006; Gachohi, Kitala, Ngumi, Skilton & Bett, 2013). The warm conditions of the Kerio Valley are conducive for the survival of many pathogens and vectors including the ticks that transmit most of the economically important diseases of livestock in sub Saharan Africa. The main land use is livestock keeping with few pockets where farmers practice mixed farming, thus majority of communities living in the valley are pastoralists. Most pastoralists keep indigenous and mixed breeds of cattle, sheep and goats and are usually vulnerable to economic impacts of parasitic infections of livestock.

Vector borne infections such as trypanosomosis, theileriosis, babesiosis among others and gastrointestinal parasites affect majorly livestock that form main economies in rural areas in terms of provision of meat, milk, farm manure, skin/hides and income. In Kerio Valley, there exists a national park, which may be a possible reservoir for trypanosomes and ticks being maintained among the wildlife animals. It has been reported that zoonotic *T. brucei rhodesiense* that is common in East Africa is responsible for human trypanosomosis with cattle and wildlife animals acting as main reservoirs (Majiwa, 1986). Diseases due to vector infestation and helminth infections directly impede livestock production through limiting introduction of highly productive animal breeds in disease prone areas (Perry & Young, 1995).

Early detection, proper identification of animal parasitic infections are critical in the control, limiting extend of pathogenesis due to parasitic infection and also protect potential zoonotic infections in humans. Studies on haematological profiles have been done on indigenous animal in Western Kenya (Van Wyk, *et al.*, 2013) but baseline values of haematological parameters on indigenous animal breeds in semi-arid lands especially in Kerio Valley in Kenya are lacking. Blood data values obtained from this study can be

used to measure animal health status and can be used as reference blood ranges for indigenous breeds reared in their natural environment. Knowledge on haematological parameters of livestock in Kerio valley is also necessary for successful management of infectious diseases with accurate diagnoses and in improving animal production in similar systems.

Data on prevalence and characterization of trypanosome species can be utilized to improve health status among livestock species kept by farmers in Kerio valley so as to improve livelihoods and prevent possible transmission of zoonotic trypanosome species to humans. The information on tick-borne and gastrointestinal parasites on the other hand, is important for policy makers and Ministry of Livestock, Agriculture, Fisheries and Irrigation in the development of clear and effective control measures that will allow the producer/livestock owners to realize farm animal productivity benefits.

In Kenya, most studies on epidemiology of livestock diseases have concentrated in high-potential areas, with specific group of livestock such sheep or goats or cattle (Kanyari 1993; Ng'ang'a, Maingi, Kanyari & Munyua, 2004; Odoi *et al.*, 2007). This calls for similar studies in Kerio Valley where livestock is the mainstay among communities here. The utility of information on epidemiology of livestock parasites is also critical in developing strategies, for management of existing and emerging vector borne and gastrointestinal infections.

1.4 Main Objective

To investigate the occurrence of vector borne parasites, gastrointestinal parasites, ectoparasites, assess trypanosome parasites and evaluate their effects on haematological profiles of livestock kept under semi-arid conditions along the upper Kerio Valley.

1.4.1 Specific Objectives

1. To determine the identity and prevalence of gastrointestinal helminths and protozoan parasites among the livestock in upper Kerio Valley
2. To determine prevalence of haemoparasites affecting livestock in upper Kerio valley
3. To determine the effects of vector borne and gastrointestinal parasites on haematological parameters of infected/infested livestock in the upper Kerio Valley
4. To determine distribution and difference in haematological profiles based on sex, age and location of livestock in upper Kerio Valley

1.5 Research Questions

- 1) What is the prevalence of gastro-intestinal helminth and protozoan infections affecting livestock in upper Kerio Valley?
- 2) What is the prevalence of haemoparasites affecting livestock in upper Kerio valley?
- 3) What are the effects of vector borne haemoparasites and gastrointestinal helminth infections on haematological parameters of the infected animals
- 4) What are the normal haematological parameter ranges in livestock of upper Kerio valley and other factors that affect their values?

CHAPTER TWO

LITERATURE REVIEW

2.1 Socio-economic Importance of Livestock

In many communities in Kenya and many countries across Africa, livestock play important roles in providing income, farm manure and proteins in terms of milk and meat. They are also used as a medium for social exchange in the payment of bride price, fines and gifts to strengthen kinship ties among local communities (MoLD, Kenya, 2010). Livestock are of enormous importance in the Kenyan economy and households with the population estimated to be 60 million comprising of indigenous and exotic breeds, contributing to about 12% of Gross Domestic Product (GDP) and 40% of Agricultural GDP (MoLD, Kenya, 2010; MoLD, Kenya, 2011).

According to Rege *et al.*, (2001), indigenous breeds of cattle constitute 77% of the total Kenyan cattle population. Livestock keepers in areas with adverse environmental conditions prefer indigenous and mixed breeds because of their adaptability to harsh environmental conditions, various vector borne pathogens and helminthosis (Mwacharo and Drucker, 2005). In harsh climatic environments, many livestock farmers and/or pastoralists depend on livestock to sustain livelihoods (Onono, Wieland & Rushton, 2013).

The major challenges to livestock production include parasitic diseases and vectors such as tsetse fly and ticks being the major livestock pests transmitting trypanosome parasites and tick borne pathogens respectively among others (Thumbi *et al.*, 2010, Byaruhanga, Collins, Knobel, Kabasa & Oosthuizen, 2016.). According to the Kenya Ministry of Livestock Development (2011), tsetse fly infests an area covering 138,000 square

kilometres in 38 out of the 47 counties, Elgeyo-Marakwet County included. Consequently, parasitic infections due to trypanosomes, tick-borne and helminthic infections threaten food security and livelihoods in the affected areas causing production losses in terms of mortality, reduction in weight gain, retarded growth, reduced fertility (Thumbi *et al.*, 2013) and high cost incurred in purchase of therapeutic and preventive drugs.

2.2 Livestock Parasitic Diseases

Parasitic diseases are highly prevalent in the tropics and sub-tropics of the world since the animals are highly exposed to infections as a result of favourable environmental conditions, coupled with poor farm practices and unaffordability to appropriate care by majority of the livestock owners (Shah, *et al.*, 2017). There are several parasitic animal diseases both intestinal and haemoparasites. Parasitological diseases such as those caused by nematodes, trematodes, protozoa and ectoparasites have various negative effects on animals in terms of milk, meat and hide production (Fitzpatrick, 2013)

2.3 Trypanosomosis

Trypanosomosis is a disease of both humans and animals depending on taxon. The parasite is majorly transmitted by tsetse flies of *Glossina* genus while others are transmitted mechanically by biting flies (Machila *et al.*, 2003). Animal trypanosomosis is caused by species of flagellate protozoa belonging to the genus *Trypanosoma*, which inhabit the blood plasma and various body tissues and fluids. These parasites affect many animals but they are more pathogenic in mammals, including livestock and man (FAO, 1992). *Trypanosoma* species cause sleeping sickness in human and animal trypanosomosis or ‘nagana’ in various domesticated animals and wildlife animals (Bronsvort, 2010).

In some parts of Kenya, and East Africa, animal trypanosomosis is endemic, causing chronic anaemia (Taylor & Authie', 2004, Bronsvoot, 2010) enlarged lymph nodes, staring coat, weakness and depression, general loss of productivity and overall condition including reduced milk yield and impaired fertility (Connor, 1994; Bowman, 2009).

2.3.1 Pathological effects of trypanosomes infection

Pathogenic trypanosomes that belong to the genus *Trypanosoma* cause disease in all types of domesticated animals and several pathological complications and disorders arise due to trypanosome infection in a susceptible animal (Taylor & Authie' 2004). The resulting disease can occur in two forms, chronic or acute depending on animal's susceptibility status and the strain's virulence (Courtin, 2008). Nagana parasites develop mainly in blood plasma but *T. brucei brucei*, develops additionally in the tissue fluids. Trypanosome species that affect livestock depend on the strain, the number of transmitted trypanosomes, ecological factors, transmitting tsetse flies *sp* and genetically determined susceptibility of a particular animal breed (Gibson, 2009). The age and the conditions of the animal are important as well as the vicinity of game animals (Seifert, 1996).

Pathogenic trypanosomes cause debilitating diseases in livestock and it is manifested in form of anaemia, weight loss, and reproductive complications with consequent disruptions in the secretions and plasma concentration of hormones necessary for normal reproductive processes in both sexes and death may result (Sekoni, 1994). Oxidative stress can arise due to trypanosome infection as a result of imbalance between the production of reactive oxygen and the inability to readily detoxify the reactive intermediates resulting in tissue damage such as cell components like proteins and lipids (Murray, 2003).

2.3.2 Symptoms of trypanosomosis

Clinical trypanosomosis have symptoms that are similar in animals that are tolerant and those that are non-tolerant and these include parasitaemia with fever, anorexia, dullness and anaemia. While some may die in the acute phase, others may enter a chronic phase (FAO, 1992). Trypanosomosis is also characterized by relapsing parasitaemia, anaemia, loss of condition, abortion, alterations in the ovarian cycle leading to infertility and, if left untreated it results in a high mortality (Murray, 2003). Trypanosomosis is known to cause immunosuppression further complicating animal's morbidity by being more prone to infection by other diseases and in other cases it may cause severe weight loss and haemorrhages (MoLD, 2011).

2.3.3 *Trypanosoma* species

Trypanosoma vivax, *T. congolense* and *T. brucei* are the three most important species of trypanosomes, responsible for considerable losses in production and livestock morbidity where they occur (Kristjanson, 1999). These parasites are transmitted by tsetse flies in the genus *Glossina*, in which they have obligate life cycle stages (Bronsvooort, 2010). A trypanosome is an elongated, spindle shaped single celled protozoa with a single nucleus lying near the middle of its length and a single flagellum that arises near large mitochondria with a kinetoplast and passes out through the anterior (Bowman, 2009).

The principal species of pathogenic trypanosomes infecting particularly African cattle include; *Trypanosoma congolense*, *T. brucei*, and *T. vivax* (FAO, 1992). *Trypanosoma congolense* is found in most domestic mammals such as cattle, sheep, goats, horses, pigs, camels and dogs; and many wild animals and consists of three sub-groups namely Savannah, Kilifi and Forest clades (Majiwa, 1986). *Trypanosma brucei* has three morphologically identical subspecies; *T. b. brucei*, *T. b. gambiense* and zoonotic species;

T. b. rhodesiense, which causes human sleeping sickness and can be found practically in all domestic and wild animals (FAO, 1992, Gibson, 2009, Bronsvort, 2010).

Infection levels of trypanosomes vary depending on livestock types, whether cattle, sheep or goats and further dependent on the season, the density of tsetse flies and overall animal resistance (Gibson, 2009).

2.3.4 Trypanosomes life cycle

Salivarian trypanosomes have a complex life cycle involving more than one species i.e. mammal and tsetse fly. They also have different morphological forms in their life cycles (Gunn & Pitt, 2012). Several subspecies of tsetse flies serve as their vectors and each has unique ecological requirements that define their habitats and geographical distribution (MoLD, 2011). The *Glossina* fly gets infected when it takes a blood meal from parasitaemic mammal host in which the trypanosomes undergo a cycle of development. The trypanosomes then undergo multiplication in the digestive tract of the fly until the infective metacyclic trypanosomes (metatrypanosomes) are produced and move to the salivary glands (FAO, 1983). The period from ingestion of infected blood to the appearance of infective forms in the salivary glands varies from one to three weeks and once infective metatrypanosomes are present, the fly remains infective for the remainder of its life (Gunn & Pitt, 2012). During the act of feeding the fly penetrates the skin with its proboscis and the rupture of small blood vessels in intermediate host forms a pool of blood in the tissues in which the fly injects saliva to prevent coagulation. The infective metacyclic trypanosomes injected with the saliva proliferate in the blood and lymph during the haemolymphatic stage in chancre (Courtin, 2008) (Figure 2.1).

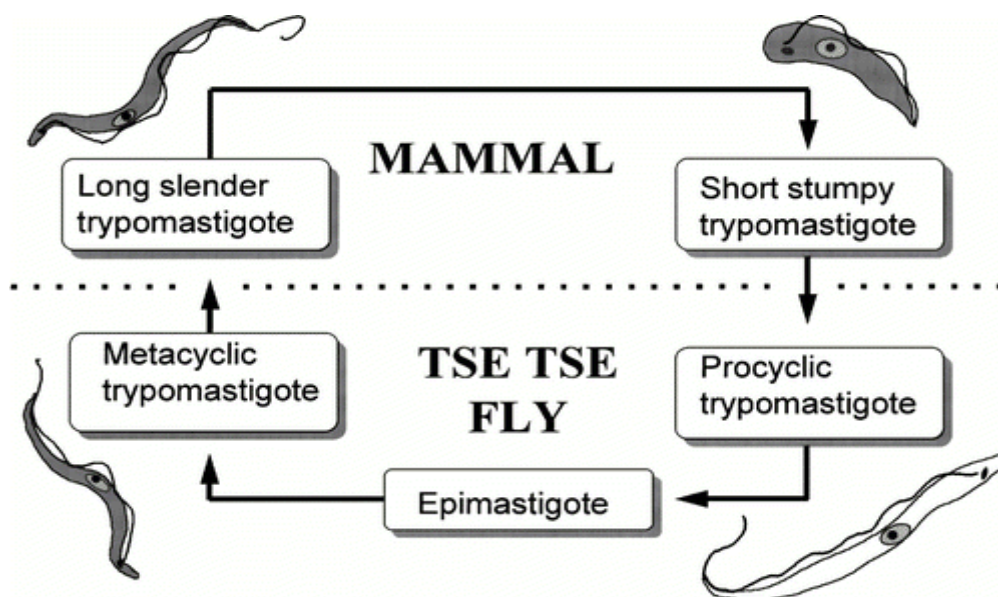


Plate 0:1: General life cycle of trypanosomes in mammal and the vector

(Source: Borst and Fairlamb, 1998)

The parasites then enter the lymphatic system and pass into the bloodstream. Inside the host, they transform into bloodstream trypomastigotes, which are carried to other sites throughout the body and reach other blood fluids (e.g., lymph, spinal fluid), and continue the replication by binary fission (Gunn & Pitt, 2012). The entire life cycle of African trypanosomes is represented by extracellular stages.

2.3.5 Survival of trypanosomes in the vertebrate host

Indigenous breeds of livestock are usually adapted to harsh conditions and possess disease resistance compared to exotic breeds. Tolerance to trypanosomes infections requires the production of IFN-gamma in the early stages of the infection that triggers the development of classically activated macrophages, which then destroy the parasites (Borst & Fairlamb, 1998; Magona, *et al.*, 2011). The survival of parasitic organisms is dependent on being able to evade and escape the immune defences of the host.

Trypanosomes can survive and develop in the host aided by receptors and transporters that are extracted from their hosts (Borst & Fairlamb, 1998). Exposure to different conditions in an insect and vertebrate host causes the trypanosomes to adjust to specific assortment of nutrients and immune defences that must be evaded. This is achieved by evading immune mechanisms through hiding their variant receptors and undergoing major genetic transformation so as to achieve variation on the surface antigens (Borst & Fairlamb, 1998; Gunn and Pitt, 2012)

2.3.6 Antigenic variation of trypanosomes and molecular diagnosis

Trypanosomes have potential of variation in which they exhibit remarkable antigenic variation of the surface glycoproteins (VSG) where hundreds of antigenic types are found (Levinson, 1998). This is where one antigenic type will coat the surface of the parasites for approximately 10 days followed by other types in sequence in the new progeny following immune response when the host make B-cell responses to the parasite immunodominant VSG (Borst & Fairlamb, 1998). Consequently, trypanosomes have the ability to undergo transcriptional switching of VSG genes, thereby displaying antigenically distinct VSG surface coats to the host immune system and escape immune elimination. Mansfield (1990) states that this interplay leads to the fluctuation of parasitaemias associated with chronic African trypanosomosis. This allows the parasite to continually evade the host immune defences in which the trypanosome parasites uses this strategy to allow the host immune system to regulate their own numbers but maintain active infection by directly evading the lethal effects of the immune response (Gunn & Pitt, 2012).

Molecular techniques offer sensitive and specific tools for the diagnosis of blood-borne infectious diseases and rely primarily on the detection of the causative agent in the blood

sample (Ahmed, *et al.*, 2011). Genetic markers have been used to rapidly identify genes that characterize different species of trypanosomes thus enabling identification of different types of trypanosome species, sub-species, strains and different repertoire antigenic levels (Majiwa, 1986).

Polymerase Chain Reaction (PCR) is a molecular diagnostic method, which has been shown to detect the presence of parasite's DNA equivalent to one trypanosome in 10ml of host blood thus reducing the limit at which trypanosome parasites can be, detected (Masake, 2002). Studies by various researchers (Desquesnes *et al.*, 2001; Thumbi *et al.*, 2008) utilized multiple species identification using a single primer sets based on ribosomal RNA gene sequences. According to Bronsvort *et al.* (2010), field applications of PCR have been used to estimate trypanosome prevalence for the monitoring of control programmes and for detecting very low parasitaemias.

2.4 Tick-borne Pathogens

2.4.1 Tick-borne heamoparasites

Among the economically important diseases of livestock in sub-Saharan Africa transmitted by ticks, include East Coast Fever (ECF) caused by *Theileria*, Babesiosis caused by *Babesia sp.*, and Anaplasmosis caused by *Anaplasma* (Uilenberg, 1995 and Minjauw & McLeod, 2003). Out of all these pathogens, East Coast Fever caused by *Theileria sp.* is most important, causing more cattle deaths than the other heamoparasites (Byaruhanga *et al.*, 2016). Animals living in free range have high chance of being infected with several parasitic infections and many may suffer concurrent parasitic infections, which heavily impact negatively on the animal general health (Jacobs *et al.*, 2016).

Haemoparasites are infectious agents that live within the bloodstream of their hosts and manifest themselves as intracellular parasites. Some of the haemoparasites only become evident when the animal responds to infection and some can easily be observed in peripheral blood in apparently healthy animals (Bell-Sakyi, 2004). Due to development of immunity to some of the parasitic infection, the animals may serve as carriers and therefore source of infection to the vectors, perpetuating constant infection and reinfection of livestock (Byaruhanga *et al.*, 2016).

East coast fever is principally a disease of cattle, caused by protozoan *Theileria parva* and transmitted by various ticks including *Rhipicephalus appendiculatus* (red ear tick) and *Hyalomma* species (Shah, *et al.*, 2017). *Theileria* parasites invade host lymphocytes and induce them to divide continuously and after 2-3 weeks, attacking cytotoxic T-cells initiate a massive destruction of parasitized lymphoblasts, all the while, the parasites invade erythrocytes and differentiate into the life cycle stages which are infective to the ticks (Jacobs *et al.*, 2016). The symptoms include anorexia, hepatomegally, splenomegally, swelling of the lymph nodes and petechial haemorrhage (Gunn & Pitt, 2012).

Babesiosis is caused by several species of *Babesia*; an intra-erythrocytic parasite. Although human infections may occur, and may be fatal, babesiosis is primarily a disease of economic importance of cattle, sheep and other domesticated animals (Gunn & Pitt, 2012). Important species of *Babesia* include *Babesia bigemina* (Fig. 2.3), *B. bovis* and *B. divergens*. Clinical signs of babesiosis include febrile condition, anaemia and haemoglobinuria as a result from the loss of function and destruction of red blood cells by the replicating parasites (Shah, *et al.*, 2017). Cattle normally acquire infection from recovered animals, which continue to harbour sub-clinical infection and adult cattle tend

to be more severely affected than the young animals (Gunn & Pitt, 2012; Shah, *et al.*, 2017).

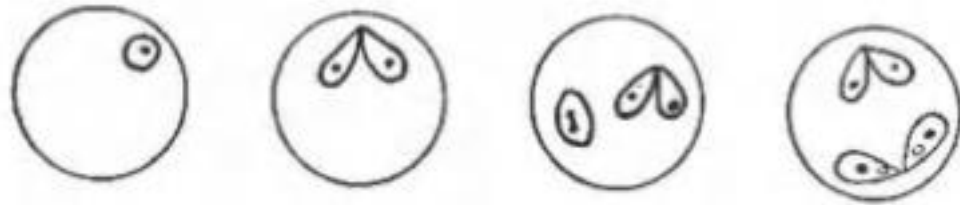


Plate 0:2: Diagrammatic presentation of *Babesia bigemia*

(Source: Gunn & Pitt, 2012)

2.4.2 Transmission dynamics of tick-borne pathogens

Factors that influence transmission of tick-borne pathogens include the number of times a vector feeds and the vertebrate host that are fed on (Sonenshine & Mather, 1994). A three-host tick has higher potential of transmitting disease to more vertebrate hosts than a one-host tick especially if transmission of the pathogen is also possible transovarially (Jacobs *et al.*, 2016). The efficiency of pathogen transmission by the ticks according to Jacobs *et al.*, (2016) is contributed by the fact that many tick species feed on more than one animal during their life cycle, remain attached for longer periods while feeding during which pathogens may be deposited and many tick borne pathogens also multiply within the tick.

2.5 Helminths and Protozoa

Helminth and protozoan parasites are prevalent in sub-Saharan African due favourable environmental conditions and poor management practices (Bell-Sakyi, 2004). Helminth

and protozoan especially coccidial infections have been found to be widespread in Kenya, and stongylid species have been reported to be most prevalent and highly pathogenic in livestock (Waruiru, *et al.*, 2000). Gastrointestinal parasitism in livestock is threat to animal production due to the high cost of management in terms of treatment of disease and control measures (Van Wyk *et al.*, 2013). They cause clinical and subclinical diseases contributing further to economic losses.

2.5.1 Gastrointestinal nematodes

Nematodes are helminths with remarkably constant body form with relatively large body cavity (pseudocoelom) and the males are relatively smaller than the females (Bowman, 2009). Gastrointestinal nematode infections affect productive performance of livestock for both large and small-scale farmers. Important nematode species affecting ruminant animals in the tropics include *Haemochus species*, stomach worms; blood sucking *Trichostrongylus*, *Bunostomum Oesophagostomum spp*, *Ostertagia* species among others (Bowman, 2009). The nematode infections can be detrimental to the health of an animal if intensity is high especially if the animals are not well fed (KARI, 1999). The generalized life cycle of nematodes are represented by four stages; the adult, preinfective, infective and pre-adult separated by contamination, development, infection and maturation (Bowman, 2009) as illustrated by *Strongyloides sp* life cycle in Figure 2.3. The distribution of gastrointestinal nematodes is dependent on environmental conditions and management systems.

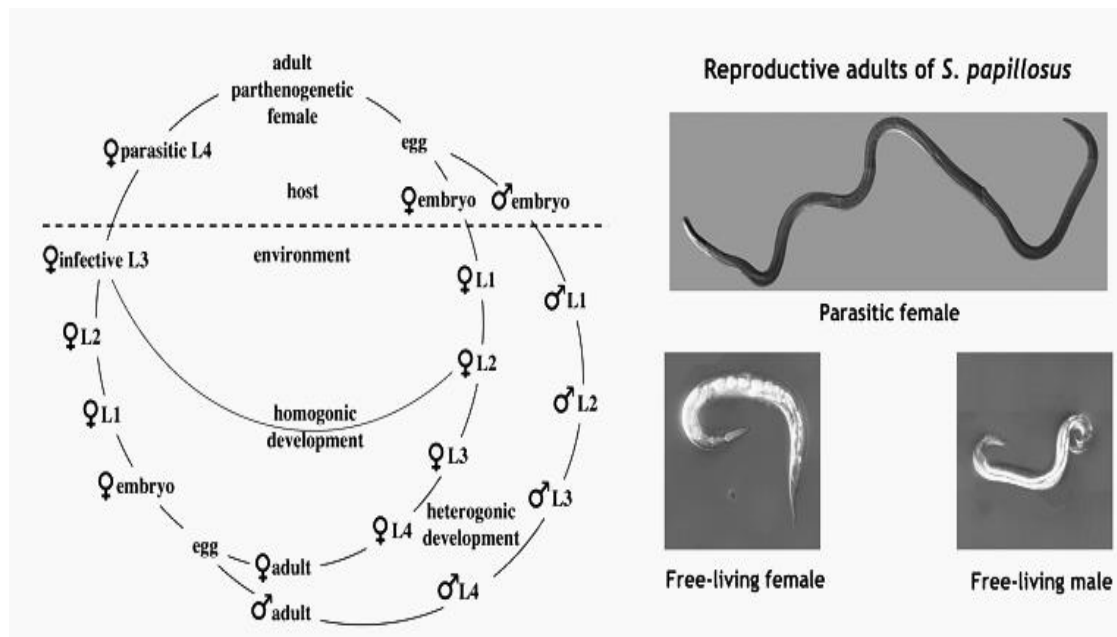


Plate 0:3: Life cycle of *Strongyloides* sp

(Adopted from Nametschke *et al.*, 2010)

2.5.2 Cestodes

Cestodes belong to class Cestoda (tapeworms). Parasitic tapeworms use vertebrates as definitive hosts and vertebrates or invertebrates as intermediate hosts depending on species (Taylor, 2007). The definitive host harbours the adult or sexual mature forms of the parasites and most adult tapeworms reside in the small intestines of their hosts. Tapeworms cannot be transmitted directly from one animal to another because they must undergo development in the intermediate host (Jacobs *et al.*, 2016).

Animals get infected when they ingest cestode eggs and hatch in the small intestines (Taylor, 2007). The hatched oncospheres then penetrate the gut, enter the circulation and are swept around the body where they ultimately, penetrate muscle fibres in which they develop into cysticerci. The heart, tongue, masseter muscles and intercostal muscles are

common sites of infection although non-muscular tissues such as the liver and kidneys may also be parasitized (Gunn & Pitt, 2012). The common parasitic tapeworms in animals include *Moniezia sp*, *Diphylidium* and *Echinococcus* among others. Infections with tapeworms are rarely pathogenic except in cases in which there is an actual impaction of the gut or when the infection is very heavy. Lambs infected with *Camurus cerebralis* - the larval form of *Taenia multiceps* for instance is usually fatal (Taylor, 2007).

2.5.3 Trematodes

Trematodes are flukes with those that invade blood being referred to as blood flukes; those found mainly in the intestines are intestinal flukes and those that invade liver and biliary ducts are known as liver flukes (Gunn & Pitt, 2012). The life cycles of trematodes involve an intermediate host; usually a fresh water snails, and definitive host being warm blooded animals (Kagenda & Angwech, 2018).

Flukes that are a problem in the tropics include *Fasciola sp*, *Schistosoma sp* and *Dicrocoelium*. Fasciolosis is a well-established infection of cattle, and sheep caused by either *Fasciola hepatica* or *F. gigantica* (Egbu, 2013) and predominantly cause disease in ruminants and wildlife. *Fasciola* causes production losses in ruminants and usually lethal when it affects sheep (Jacobs, *et al.*, 2016). *Fasciola* species when mature feed on blood thus affecting haematological profiles of affected animal that result to anaemia, apart from causing tissue damage due to migratory habits leading to liver condemnation (Egbu, 2013).

2.5.4 Gastrointestinal protozoa

A number of enteric protozoa that cause disease are considered to have zoonotic potential in which clinical impact varies and mostly poorly defined (Thomson & Smith, 2011;

Geurden *et al.*, 2014). Intestinal protozoa including coccidian, *Giardia*, *Balantidium spp* are linked with clinical and sub-clinical symptoms in livestock and many of them though neglected, are known to have zoonotic potential (Geurden *et al.*, 2014). Coccidiosis caused by several species in the class Coccidia, affects livestock especially small ruminants and calves (Nganga, *et al.*, 2004). *Eimeria* species specifically are associated with both acute and chronic disease in affected animals (Waruiru *et al.*, 2000).

2.6 Vectors

Many parasites have complex life cycles involving two or more species of host within which different developmental stages occur. Majority of the intermediate hosts are arthropods but molluscs can be vectors (Gunn & Pitt, 2012). Some parasites used intermediate hosts to physically transmit them between hosts, playing a role in parasite epidemiology. Climate change may have direct consequences in geographical ranges, survival and transmission dynamic of the vector-borne infections by vectors (Short, Caminade & Thomas, 2017). According to Gunn & Pitt, (2002), the effectiveness of a vector as a transmitting agent is heavily dependent on its feeding behaviour and host preference hence controlling of vectors plays a great role combatting vector-borne parasites.

2.6.1 Transmission of African animal trypanosomosis

Apart from *T. equiperdum* that affects majorly horses, and is transmitted through coitus, the rest of the trypanosomes are transmitted by insects' bites majorly the *Glossina* (tsetse) flies. The tsetse fly has strong and long proboscis for sucking blood, both male and females are blood sucking and they have strong wings which sustain long flights (Fig 2.4) (Gunn & Pitt, 2002). Tsetse flies are the chief vectors of African trypanosomes in which they serve as intermediate hosts that enable the parasites to multiply actively (FAO,

1992). Infections with *T. vivax* on the other hand are transmitted both by tsetse flies and mechanically by biting flies such as *Tabanid* flies (Gutierrez, *et al.*, 2006).

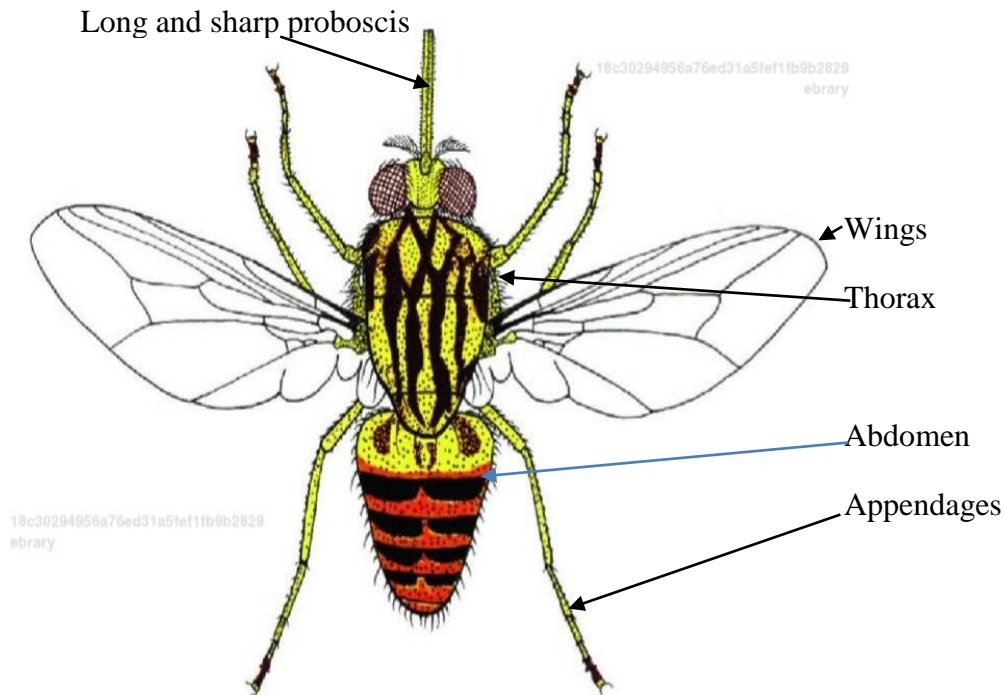


Plate 0:4: *Glossina* fly (tsetse)

(Adopted from Principles of Veterinary Parasitology, Taylor et al., 2007)

Tsetse flies are widely distributed in many parts of Kenya and are known to correlate with the location of wildlife conservancy and high prevalence of poverty (Ministry of Livestock Development, 2011). Both male and female flies are blood-sucking, with the aid of long and sharp proboscis, active during day especially morning and evening, they possess strong wings which enable the tsetse to fly long distances, and occur all year round though they are more numerous during rainy season (Jacobs, *et al.*, 2016) and therefore they all have high potential of transmitting pathogens to the vertebrate hosts

throughout the year. According to survey made by MoLD (2011), the tsetse flies most likely to be distributed along Kerio valley from Bogoria region are *Glossina pallidepes* (spatially distributed in all tsetse fly belts in Kenya) and *G. morsitans* found mostly in tsetse fly belt on Kenya-Uganda border north of Mt Elgon.

2.6.2 Tick Vectors

Ticks are important ecto-parasites of livestock as well as vectors to many pathogens because of their voracious blood-feeding activities (Bell-Sakyi, 2004). Ticks comprise of various genres but practically it is difficult to identify ticks to species level because ticks are capable of interbreeding to produce individuals that are fertile (Walker, *et al.*, 2003). According to Walker *et al.* (2003), all the feeding ticks at each life cycle stage are parasitic and are therefore capable of transmitting a pathogen if they are infected. Ticks are ectoparasites on many species of farm animals and continue to cause significant health threats to livestock, particularly cattle. Ticks transmit pathogens passively or may serve as obligatory intermediate host for certain protozoan parasites such as *Theileria*, *Babesia* and *Anaplasma* species (Hendrix & Robinson, 2006).

Ticks may be one or two or three host ticks depending on the number of animal hosts they feed on during their life cycle. The life cycle of ticks involves laying of eggs on the soil and after a few days depending on the species, hatch to larvae, which crawl to nearby vegetation as they wait for their appropriate hosts (Walker, *et al.*, 2003). Moulting occurs between different life cycle stages from larvae to nymph and finally to the adult and this may occur on the host or on the ground depending on the species (Sonenshine & Mather, 1994). The fully engorged larvae, nymph or female adult drop from the host, while the males may remain on host for days or months mating with numerous females.

Apart from transmitting prokaryotic and eukaryotic parasites, ticks are parasitic in their own rights as ectoparasites affecting the quality of skin and sucking of blood from their hosts (Walker *et al.*, 2003). This has implication to the economic loss in terms of poor quality of hides and general poor condition of the animal and expenditures in tick control measures (Bell-Sakyi, 2004). Different species of tick vectors are known to be adapted to different ranges in temperature and moisture: some occur only in warm regions with a fair degree of humidity, while others are mostly active in dry climates (Randolph, 2005).

Ecological parameters that affect tick-borne infections according to Sonenshine & Mather, (1994), include and not limited to vector competency, population dynamics of the vector species, seasonality of tick vector activity, host specificity and other host-related factors, zoogeographic range of vectors and habitat requirements. Understanding these factors helps in the control of vector borne diseases of animals and humans. While ticks play a role in acquiring and transmitting infections from one vertebrate host to the other, the host on the other hand are capable of acquiring, maintaining and donating infectious agents to the next tick vector (Byaruhanga, *et al.*, 2016) hence transmission to other hosts hence maintenance of parasites.

2.6.3 Snail Vectors

Flukes have aquatic snails as their intermediate hosts, implying that they undergo indirect life cycles and therefore prevalent where the snail vectors and infective stages can survive (Choubisa and Jaroli, 2013). Parasites such Schistosomes and *Fasciola* species have fresh water snails as vectors in which they undergo a period of development. Majority of the trematode parasites are obligate parasites of vertebrates during their adult stages while the asexual stages develop in snail intermediate hosts. *Fasciola sp* penetrate *Lymnae sp* in which they develop, while for Schistosomes' immature stages invade appropriate snail

vectors including *Biomphalaria sp* and *Bulinu sp* (Gunn and Pitt, 2012). The distribution of trematodes follows the distribution of snail vectors whose prevalence depends on the presences of water bodies which provide favourable ecological conditions for the growth of the snails and development of trematode larval stages (Nzalawahe *et al.*, 2014). Hence, livestock can pick up infections from water sources or swamps containing infective snails and infective stages; cercariae or metacercariae.

2.7 The Impact of Climate Change on Parasitic Infections

The earth is warming up and therefore climate change is happening, largely spurred by human activities, consequently having many serious and potentially damaging effects in the long run. According to the Intergovernmental Panel on Climate Change (Intergovernmental Panel on Climate Change: (IPCC), 2007), climate change currently contributes to the global burden of disease and premature deaths since animals, humans included are exposed to climate change through changing weather patterns and these climate change exposures are likely to affect the health status of millions of biological systems. The rise in temperature could in effect extend the ecosystems range of pests and pathogens. Some of the animal diseases are expected to spread to areas that initially were not inhabited by its vectors as temperature rises and precipitation patterns change (WHO, 2009).

Climate change is likely to lead to the emergence and re-emergence of certain wildlife diseases, infectious diseases especially vector-borne diseases, which are dependent on environmental and climatic conditions e.g. babesiosis, trypanosomosis, and a host of internal and external parasites (World Conservation Society, 2008 and Singh, 2012). With the health of wild animals directly linked to their ecosystems and environment, even

minor disturbances can have far reaching consequences on diseases they might encounter and transmit to other animals as the climate changes.

2.8 Haematological Parameters and Parasitic Infections

Haematological parameters are components of blood and refer to the numbers and morphology of individual cellular elements of blood (Etim *et al.*, 2014). Etim *et al.*, (2014), described blood as a connective tissue that act as a pathological reflector of the status of the health of the animals exposed to toxicity or infections.

Haematological parameters have been demonstrated to have health implications that are important in diagnosis, prognosis and treatment of animal diseases in animals under different management systems (Onasanya, *et al.*, 2015, Zvinorova *et al.*, 2016). Cellular components of blood which include Leucocytes (white blood cells), Erythrocytes (red blood cells) and Thrombocytes (platelets), are also used in diagnosis, monitoring diseases and understanding general physiological state of animal (Merck Manual 2012, Egbu, 2013, and Etim *et al.*, 2014). Red blood cells are involved in transport of oxygen and carbon dioxide in an animal body (Isaac, 2013) while white blood cells and differential components play important role in the fight of infections and defend the body against infectious organisms by transporting or producing antibodies against pathogens (Etim *et al.*, 2014 and Kubkomawa *et al.*, 2015).

The erythrocytes (RBC) measurements include packed cell volume (PCV/HCT), which indicates the proportion of whole blood volume occupied by red blood cells, haemoglobin (HGB) that shows concentration of red blood cells per unit volume of whole blood, mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC). Changes in leucocytes are measured through white blood cell counts (WBC), Lymphocytes, and granulocytes. Thrombocyte parameters that are involved in

aggregation and clot formation are measured by values such as mean platelet volume (MPV), platelet distribution width (PDW) and plateletcrit (PCT) (Egbu, 2013; Onasanya, *et al.*, 2015). Reference ranges of haematological values used in determining physiological status of animal may vary depends depending on the environment, nutrition and disease (Shawaf, *et al.*, 2018). Standard reference values widely used for disease prognosis, diagnosis and therapeutic monitoring in livestock are shown in Table 2.1 (Al-Bulushi, *et al.*, 2017).

Parasitic infections have varied consequences on haematological parameters of infected livestock. In case of any parasitic infections, the most affected cell types are red blood cells, white blood cells and thrombocytes, however, haematological changes varies with levels of immunity, endemicity of diseases, nutritional status and demographic factors (Etim *et al.*, 2014). Packed cells volume also known as haematocrit has been used previously as an indicator of parasitic infestation especially helminthiasis (Zvinorova *et al.*, 2016). Trypanosomes for instance, cause disruption of erythrocytes' membrane through direct effects or indirectly through their products. Haemoparasites including tick borne and trypanosomes cause decrease in haemoglobin concentration and haematocrit (Ng'wena, *et al.*, 2011). Fatal haemolytic anaemia has been associated with dairy herd with mostly vector-borne infections such *Anaplasma marginel*, *Babesia*, and *Theileria* species (Riond, 2008).

Table 0:1: Normal Haematological Ranges of Livestock

	COW	SHEEP	GOAT
WBC mm ³	4 – 12	4 - 12	6 – 16
RBC in 1mm ³	5 – 10	8 – 16	12 – 20
HGB gm/100ml	8 – 14	8 – 16	8 – 14
PCV/HCT %	24 – 48	24 – 50	24 – 48
MCV mu ³	40 – 60	23 – 48	18 – 24
MCH	11 – 17	9 – 12	8 – 12
MCHC	26 – 34	29 – 35	35 – 42
NEUT %	15 – 45	10 – 50	30 – 48
LYMPH %	45 – 75	40 – 75	50 – 70
MONO %	2 – 7	1 – 6	1 – 4
EOS %	2 – 20	1 – 10	3 – 8
BASO %	0 – 2	0 – 3	0 – 2

Key

WBC- White blood cell count; RBC- Red blood cell count; LYMPH% - Lymphocyte %; MONO%- Monocytes; EOS% - Eosonophils, BASO%;- Basophils, NEU % - Neutrophils, HGB- Haemoglobin concentration (g/dl); HCT- Packed cell volume/heamatocrit; MCV – Mean cell volume; MCH – Mean corpuscular volume Hb;

MCHC – Mean corpuscular Hb concentration, (Ministry of Livestock: Veterinary laboratory Techniques)

Liver flukes such as *Fasciola hepatica* and *F. gigantica* migrate through liver tissue damaging it and when matures it feeds on blood tissue at rate of 0.2ml to 0.5ml per day per fluke (Wiedosari *et al.*, 2006) interfering with haematological profiles of the infected animal. Previous studies have proven that infection with *F. hepatica* in sheep and cattle causes significant decrease in PCV (HCT), haemoglobin concentration, RBC (Egbu, 2013). Furthermore, physiological characteristics including haematological parameters are also affected by genetic and non-genetic factors (geographical, time of the day, life habit, physiological status) of an animal (Etim *et al.*, 2014).

Haematological changes have been reported to provide a clinical picture of the host animal and the extend of blood cell damage, thus supports the fact that haematological parameters are good measures that can be used to asses health status of the animal (Khan & Zafar, 2005; Olafedehan, 2010). Isaac (2013) established that animals with good and normal blood composition are likely to show good production performance. Haematological profiles are also linked to the environment and therefore they can be used in selection of animals that are genetically resistant to certain diseases and prevailing environmental conditions (Isaac, 2013, Etim *et al.*, 2014, Kubkomawa, 2015).

2.9 Control Strategies against Livestock Diseases

The most common livestock diseases in Kenya's agricultural areas include all tick borne diseases, trypanosomosis and helminth parasites most of which are constantly present in production systems (Waruiru, *et al.*, 2000, Nganga, *et al.*, 2004; Thumbi, *et al.*, 2008). Therefore, livestock owners spend much of their income on purchasing dewormers, acaracides and treatment of endemic animal diseases. The current control measures

against common helminth and tick-borne parasitic infection in livestock include regular deworming and spraying/dipping the animals using various brands of acaricides (MoLD, 2010). Trypanocidal drugs on the other hand are commonly used method for control of bovine trypanosomosis in Sub Saharan Africa (MoLD, 2011). However, the preferred control measures for vector-borne infections are usually geared towards vector control rather than chemotherapy for vector-borne infection. According to MoLD (2011), target and trap techniques have been used with great success in reducing tsetse populations in endemic areas.

The use of parasite resistant animals has been adopted by communities in parasite prone areas. For instance, use of tryptolerant livestock in tsetse fly prone areas allows keeping of livestock rearing in tsetse fly invested areas. As stated by MoLD report (2011), animal populations, which have been subjected to natural selection by continuous exposure to disease pressure over many generations, develop traits that make them resistant or tolerant to animal parasites.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study Area

The study was carried out along the upper part of Kerio Valley of Elgeyo-Marakwet County (Fig 3.1). The County is located in the North Rift region of Kenya and it borders the counties of West Pokot to the north, Baringo County to the east, southeast and south, Uasin-Gishu to the southwest and west, and Trans Nzoia to the northwest (County Government of Elgeyo-Marakwet (CGEM, 2019).

The valley lies between Latitude $0^{\circ} 38' 24''$ (0.6772°) N and $35^{\circ}36'31''$ E (35.6086° E). The valley floor lies at 1200m above sea level (CGEM, 2019). It sits at an elevation of 1,000 meters in the Great Rift Valley; the isolated Kerio Valley is situated in a narrow, long strip that is approximately 80 km by 10 km wide at its broadest, through which the Kerio River flows. It has semi-tropical vegetation on the slopes, while the floor of the valley is covered by dry thorn bush. Annual mean temperatures of 24°C is experienced and the rainfall ranges between 1000mm to 1400mm. Due to low rainfall, the communities in the valley have practiced irrigation systems utilizing irrigation furrows since time immemorial (National Museums of Kenya, 2010).

It is inhabited mostly by the Keiyo and Marakwet, the sub-tribes of the Kalenjin community. Most people in this county are peasant farmers practicing small-scale farming and livestock rearing while some are purely pastoralists. Elgeyo-Marakwet County has a large population of livestock, which are adapted to semi-arid of Kerio Valley. The livestock kept in Kerio Valley are mostly short-horned East African Zebu breed (*Bos indicus*) (Rege *et al.*, 2001), small East African goats, Red Masai sheep that

are grazed extensively on communal land except in some sections in Keiyo South County where animals are grazed on individual farms. Supporting infrastructure includes cattle dips, which are mainly managed by the community, and livestock keepers pay per animal head some set amount of money per a dipping session.

In the vicinity is a Rimoi National Reserve in Kerio valley with good vegetation cover, which provides a stable microclimate with suitable temperature and humidity where tsetse flies can thrive well. The game reserve consists of large mammalian animals such as elephants, waterbuck, buffalos, bushbuck, and baboons among others (MoA, 2005). The presence of wildlife also provides parasite reservoir for livestock infestation and provide an important source of food for the survival of parasite vectors. Due to the nature of farm practices, livestock are highly exposed to gastrointestinal helminths and flukes, ectoparasites such as ticks and vector-borne parasites. Environmental conditions coupled with poor sanitation in some farms also favour survival of free-living forms of nematodes, all year round creating constant source of infection to livestock and humans.

The harsh climatic conditions experienced in the Kerio valley results in loss of livestock due to drought and sometimes banditry may occur resulting in loss of animals and human life. This forces the inhabitants in the area to move remaining animals away into the forests and sometimes to the escarpment for security as well as to find food for them. The main system of grazing in Kerio Valley is free range and at night, the animals are kept in an open animal enclosure close to the homesteads. Animals reared in the valley are naturally exposed to pathogen challenge in their environment.

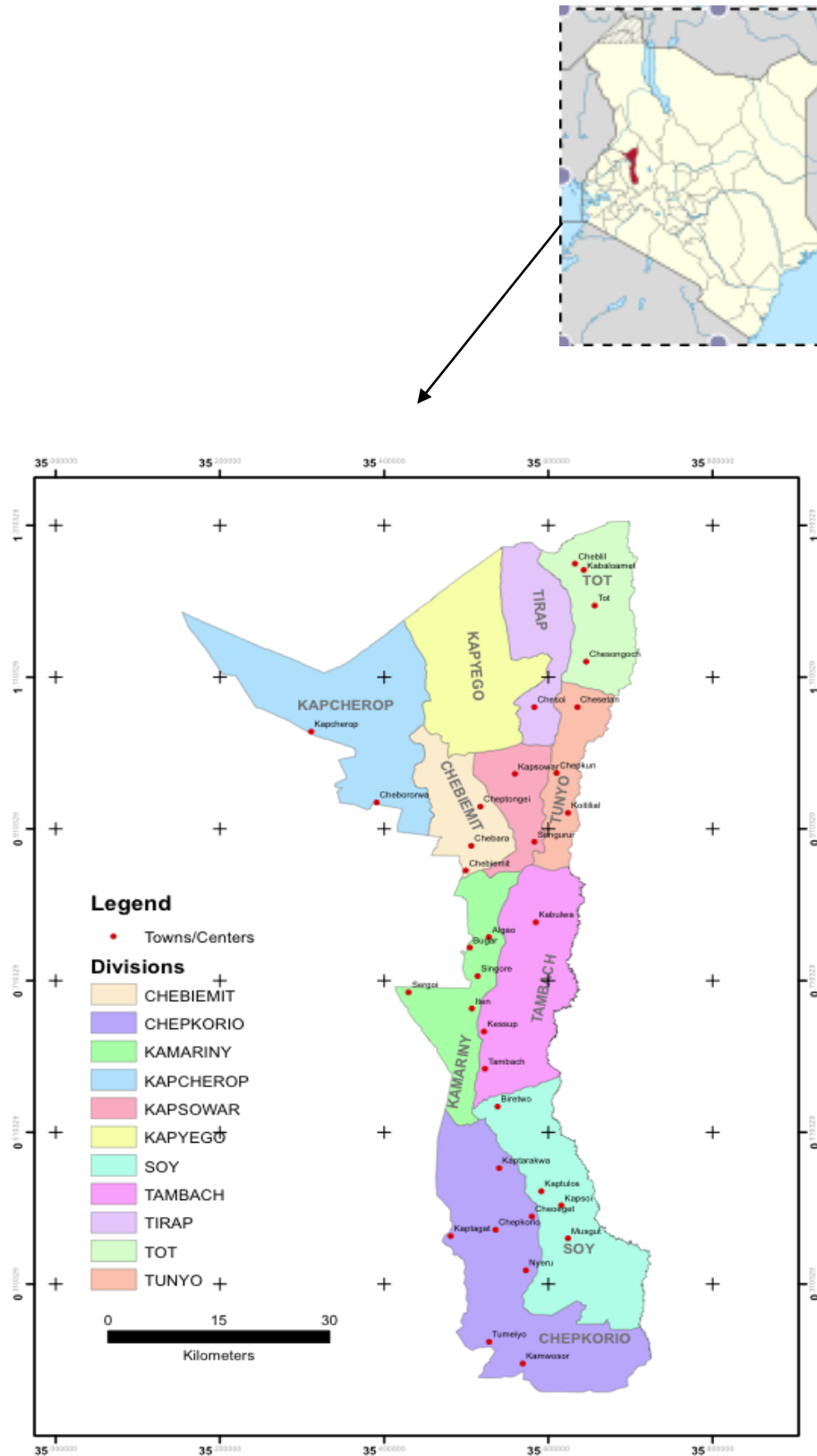


Figure 0:1: Map showing administrative divisions of Elgeyo-Marakwet County, Kenya (Source: Author, 2018)

The study area comprised of four administrative regions of Elgeyo-Marakwet County lying in the valley that included Tot, Tunyo, Tambach and Soy Fig (3.1). Pastoralism is more pronounced in the lower Kerio Valley towards Marakwet sub-county bordering West Pokot than the Keiyo sub-county.

3.2 Study Livestock

The target population used for the study of gastro-intestinal infections, haemoparasites, ectoparasite infestations and characterization of trypanosomes were cattle, sheep and goats.

Each animal was classified based on sex and age defined as yearlings, weaners and adults. The age of the study animals were determined from information given by the owner and the accompanying veterinary officer. The animals with no infections at the end of the parasite detection process acted as the control in order to assess parasitic effect on haematological indices as well as determine the normal blood parameter ranges under natural conditions. In total, 285 cattle, 98 goats and 84 sheep were evaluated and most of them apparently looked healthy (Table 3.1).

Table 0:1: Study population

Animal types	Cattle	Sheep	Goats	Total
2016	212	66	98	276
2017	73	18	0	91
Total animals sampled	285 (61.03%)	84 (19.78%)	98 (20.98%)	467

A total of 467 animals were sampled from 4 strata along the upper Kerio Valley of Elgeyo-Marakwet County

3.3 Research Design

Cross-sectional study design was used which involved multistage stratified sampling method to select the study clusters. In each study area, farms were randomly selected and random sampling of the animals was applied. Since there was no geographical overlap among sampling sites, the selections also provided geographical stratification as well. The study employed qualitative and quantitative methods to collect species and prevalence data on the parasites, haematological parameters and molecular characterization of the trypanosomes (Kothari, 2005).

The samples were collected from animals in six randomly selected study sites namely; Kabulwo (Tambach in Keiyo North) Tot, Chesongoch (Tot division in Marakwet West), Chesetah (Tunyo division Marakwet East), Chemoibon and Kaptomonger (Soy in Keiyo South) areas within upper Kerio Valley of Elkeyo-Marakwet County (Fig 3.1).

Samples (blood and stool) were taken from every second animal from the order in which they align in a crush. Ticks were picked if present from each animal sampled for identification on tick species infesting the animals. The animal health officer recorded the general health conditions, age group and sex of sampled animals and recorded against identification code. The veterinary health officer dewormed all animals restrained in the crush and treated the sick animals using appropriate drugs.

3.4 Ethical Issues

3.4.1 Ethical clearance

Permission to collect samples from the livestock was obtained from the National Commission of Science and Technology and Innovation (NACOSTI) and from the Research Ethics Committee, University of Eastern Africa, Baraton (Appendix 2 & 3). The study informed and collaborated throughout the sample collection with Veterinary and Animal Health County office of Elkeyo-Marakwet County.

3.4.2 Confidentiality

Confidentiality was maintained throughout the study by keeping names of the farmers and farms confidential, instead, special codes were assigned for use during data analysis.

3.4.3 Benefits of the research

Infected livestock were treated with current recommended drugs and livestock in selected farms were dewormed by accompanying veterinary officer using appropriate dewormers. Each farmer was also provided with free acaracides. Research findings would be published in reviewed journal for public consumption. Research findings and recommendations will be shared with Elkeyio-Marakwet County Veterinary and Animal Health Department in form of online publication so that they can be advised on how to improve parasite control among livestock farmers.

3.4.4 Risks

The research process ensured that there were correct collection procedures of blood sample collection in accordance with established standard operating procedures.

3.5 Sampling and Data Collection

Multi-stage stratified sampling was used where study population were randomly sampled from the individual strata. The sampling units were villages forming a cluster from which samples were collected from animals from the selected clusters.

Compound sample size calculation was obtained using Thrusfield (2005) formula

$$n = (Z^2 \times P_{\text{exp}} (1 - P_{\text{exp}})) / e^2$$

Where:

- Z = value from standard normal distribution corresponding to desired confidence level (Z=1.96 for 95% CI)
- P_{exp} is expected true proportion
- e is desired precision (half desired CI width)

Fifty percent (50%) prevalence rate was assumed due to the fact that there was no prior information on prevalence on individual parasites being investigated.

$$n = (1.96^2 \times 0.05(1-0.5))/0.05^2$$

$$n = 384$$

Three hundred and eighty four (384) samples were required but more animals were examined so as to reduce the sampling error. The number of animals of different ages and sex were sampled depended on their availability in each study site.

3.5.1 Collection of blood samples

The animals were restrained in a crush pen during sample collection (Fig 3.2) and also during deworming and treatment. The blood samples were collected from the jugular vein

of the animals, in which the area of the puncture was first cleansed using alcohol swap. Animals of all ages, both males and females were bled using disposable syringes and needles and blood evacuated into collection tubes.



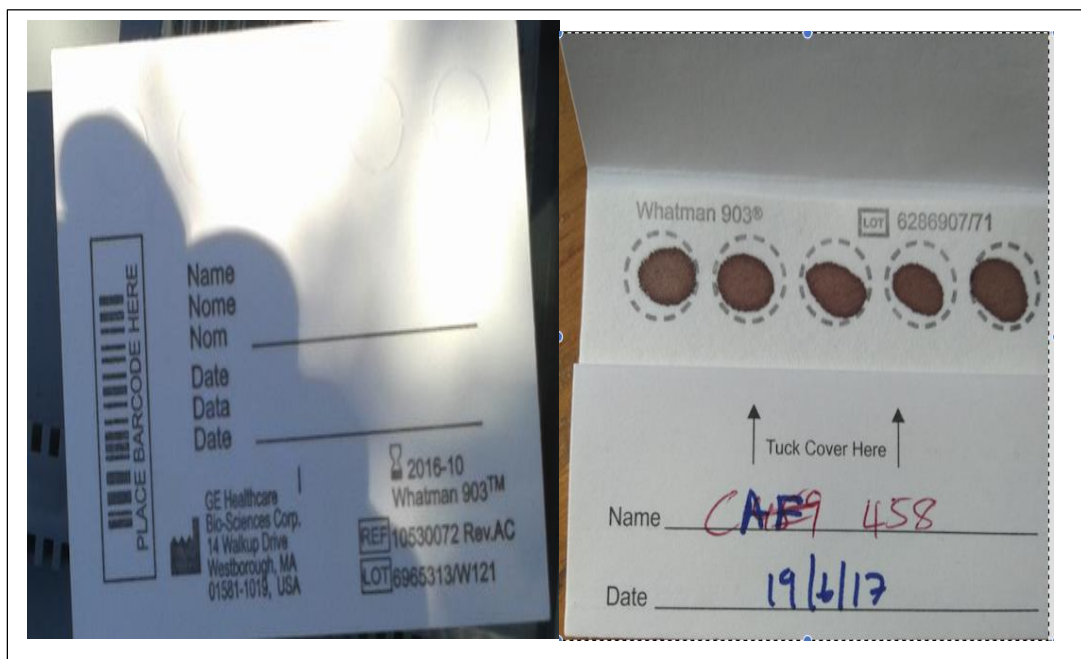
Plate 0:1: Cattle restrained in a crush

(Source: Author, 2016)

About 12ml of blood samples drawn from jugular vein of animals after locating the vein using fingers and hypodermic syringe inserted by accompanying animal health

officer/veterinary doctor. Five millilitres (5ml) of blood were collected into Ethylene Diamine Tetra-acetic Acid (EDTA) for haematological parameter analysis. The remainder of the blood was used immediately to prepare thin and thick blood smear in well-labelled slides and blood spots on the labelled Flinders Technology Associates cards (FTA-4 holes- Maidstone, Kent UK) (Fig 3.3).

The blood smears were air-dried and stored well in slide boxes before transporting to the laboratory. The blood samples (~20µl) collected, were directly applied to FTA Cards (Whatman, Maidstone, Kent, UK) circles and allowed to air dry prior to storage at room temperature using an established method of preservation for sensitive detection of trypanosome infections by PCR using published protocols (Picozzi, 2002).



(a)

(b)

Figure 0:2: FTA cards without blood spots (a) and (b) blood spotted card

(Source: Author, 2016)

3.5.2 Characterization of Trypanosomes using PCR

The technique of blood samples in Flinders (FTA) is a method in which the blood collected is put in FTA cards where all other organelles are lysed but nucleic acid remains entrapped and preserved at room temperature until data processing. FTA cards are designed with protein denaturants that cause lyse cells, denature proteins and protect nucleic acids from nucleases, oxidative and UV damage, this make them to suitable in the field conditions (Bronsvort, *et al.*, 2010).

The sample analysis depended on multiple species identification using single primer sets based on ribosomal RNA gene sequences as demonstrated by Dlugosz & Wisniewski (2006) and Desquesnes *et al.*, 2001). After DNA extraction was done, PCR was used to amplify the DNA. The list of PCR primers used in the study is shown in Table 3.2.

Table 0:2: DNA Sequence of Primers used in trypanosome identification

Primer ID	Sequence	Species	Product Size	Cycles No.
ITS1 CF	CCG GAA GTT CAC CGA TAT TG	<i>Trypanozoon;</i> <i>T.</i> <i>congolense savannah,</i> <i>T. congolense kilifi;</i> <i>T.</i> <i>congolense forest, T. vivax</i>	480; 700; 620; 700; 250	
ITS1 BR	TTG CTG CGT TCT TCA ACG AA			
ITS11	GAT TAC GTC CCT GCC ATT TG	<i>T. congolense forest;</i> <i>T.</i> <i>congolense kilifi</i>	1513; 1422	Two rounds, 35 cycles, each, the second round PCR was seeded with 1µl of the first round
ITS12	TG TTC GCT ATC GGT CTT CC	<i>T. congolense savanna</i>	1413	
ITS13	GGA AGC AAA AGT CGT AAC AAG	<i>T. brucei s.l.</i>	1207 – 1224	95° C for 7 min; 94° C for 1
ITS14	TGT TTT CTT TTC CTC CGC TG	<i>T. theileri;</i> <i>T. simiae tsavo;</i> <i>T. simiae; T. vivax</i>	988; 954; 850; 611	min, 55° C for 1 min, 72 °C for 2 min

Characterization of trypanosome parasites was dependent on genetic analysis of bands formed by different isolates compared to known profiles as described below. Those that presented identical profile were considered to belong to the same genotype.

For each PCR reaction, one 2mm punch was cut from the samples on the Whatman FTA Card and prepared according to the manufacturers' instructions. The discs were then washed twice in Whatman FTA purification reagent to remove PCR inhibitors from the sample, followed by two washes with 1xTE buffer (10mM Tris, 0.1mM EDTA) to remove residual FTA purification reagent.

Polymerase Chain Reaction

Standard PCR cycling was carried out in 29.98ml reaction mixture containing 2.4µl of 2.5mM dNTPs in a reaction tube in which 3µl of 10x buffer was added. 1.08µl of MgCl₂ was then added into the mixture, then 1.5µl of Primer 1 (forward) and primer 2 (reverse) respectively was added into the reaction tube. 17.02µl of PCR water was pipetted and added into the mixture. Further, 0.5µl of Tag pol (enzyme) was added and the master mix was vortexed to have a uniform mixture. 30µl of the master mix was measured and was put in labelled amplification tubes. 3µl of the DNA extract was added into the amplification tubes and then loaded into pre-set thermal cycler and allowed to run under the following conditions;

- a. 95°C for 7 min (initial denaturing step)
- b. 35 cycles of;
 - i. Denaturing steps at 94° C for 1 min,
 - ii. Annealing at 55° C for 1 min,
 - iii. Extension at 72° C for 2 min
- c. Elongation at 72°C for 7min

d. 4°C until used

Gel Electrophoresis of DNA samples

Prepared 1X TBE buffer and 2% agarose was heated in microwave until it started to boil and until no specks were seen in the melted agar. The flask containing the agar was set to cool until it was comfortable to touch (55°C) then 5µl of Ethidium bromide was added to agar and mixed thoroughly during cooling to mix properly. The comb was then set in the groove at the black end of the gel tray and mixture was poured on a gel tray. The melted agar was then added into the tray until it raised up to about a little less than 1/3 the length of a comb tooth. The gel was then allowed to polymerise for 30 mins. When cooled, a small amount of buffer was poured into the gel as a lubricant after which the comb was removed to leave tiny wells.

The buffer was then poured in the box until it just barely covered the gel. Five microlitres (5µl) of the DNA loading dye was measured into each DNA sample and 10µl of each of the DNA sample was loaded in each well in the gel using micropipette. A 5µl ladder was pipetted in the first well to act as control. Electrode posts were lined in the back of the lid. The power was turned on and voltage adjusted into 130 volts for 45mins. One positive control (genomic DNA) and one negative control (blank FTA disc) were run with each set of reactions. The migration was from the positive electrode to the negative electrode. Polymerase chain reaction products were then separated by electrophoresis and visualized by ultraviolet light.

3.5.3 Blood parasitaemia

Both thick and thin blood smears were prepared within a few minutes after collection of the blood samples from the animals, these were air-dried and stored in slide boxes awaiting fixing and staining in the laboratory.

Parasitaemia of haemoparasites were determined by microscopy-based techniques using direct observation of thick and thin blood smears obtained from jugular puncture. After air-drying, smears were routinely stained with Giemsa stain for 30 minutes. Only thin smear was fixed using ethanol. They were examined using light microscope at x 200 and x 400 to detect blood parasites. The thick blood smear was used to determine the presence of the blood parasites in the host, while the thin film was used in full identification of parasite species if present.

3.5.4 Haematological indices

Haematological parameters of the sampled animals were analysed to determine the effects of haemoparasites, ectoparasites and gastrointestinal helminth infections on animal health in a natural setting.

Haematological indices investigated included red blood cells (erythrocytes), white blood cells (leucocytes) and platelets (thrombocytes) cell counts are important in understanding monitoring health of the animal. The erythrocytes measurements include packed cell volume (PCV) which indicates the proportion of whole blood volume occupied by red blood cells, haemoglobin (HGB) that shows concentration of red blood cells per unit volume of whole blood, mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC).

3.5.5 Gastrointestinal helminths

(i) *Faecal collection*

Faecal samples were collected from the animal rectum using sterile gloves per animal into clean polypots to prevent cross contamination. The containers were sealed and then labelled with proper identification. They were immediately transported to the laboratory and preserved using 10% Formal saline awaiting examination and identification of eggs, oocysts and cysts.

(ii) *Faecal sample processing*

Formol-ether concentration method, a technique that is capable of identifying a variety of helminths, protozoa was used to determine gastrointestinal helminths and in isolation of cysts, oocysts and eggs of other intestinal parasites. The McMaster technique was used to determine the intensity of parasite infection by counting the number of eggs or larvae per gram of faeces.

The procedure involved weighing 3.0g or 3 teaspoonful of faeces and put in 42 ml of water then sieved using mesh sieve. The filtrate was collected in a 15ml test tube and centrifuged at 2000rpm for 2 minutes. The supernatant was then poured off and the sediment agitated before filling to the same level with flotation solution. The fluid was then removed using a pipette and both chambers of master slide were filled. The numbers of eggs in 3 chambers were counted and value from each chamber multiplied by 100 to arrive at the number of eggs per gram of faeces. The egg counts in excess of 1000 was generally considered as heavy infection and those over 500 of moderate infection (Taylor *et al.*, 2007).

Faecal smears were also made on glass slides and examined for the presence of moving oocysts and cysts of protozoan's parasites. Intensity of the intestinal parasites were determined using (MoLD, 1985 and Kanyari *et al.*, 2009). Identification of larval stages of gastrointestinal nematodes was also examined using identification keys using established protocols.

3.5.6 Ectoparasites

Tick ectoparasites were collected from the body of the animals into plastic and sealable bags containing 70% ethanol and transported to the laboratory for identification. The samples were labelled with the origin of the samples. Identification was done based on morphological and structural differences in the species at different instars based on Walker *et al.*, (2003) procedures.

Animals with dermatoses were evaluated by examining skin scrapings. The scraped material was placed in sealable bags with identification of the animal and transported into the lab. A drop of 10% Sodium peroxide was added on mounted scrapings and scanned under low magnification (Merck's Manual, 2012). To identify mite species, 10% Potassium Hydroxide (KOH) was added into the skin scrapings that were ground using a mortar and pestle. The mixture was heated in a Bunsen burner flame then centrifuged at 2000rpm for 3min., the supernatant was poured off and sediment was placed on glass slide and observed under x10 objective lenses.

3.6 Data analyses

The data was entered into Excel spreadsheet and exported to STATA 12.0 (Stata Corp, 2012) for analyses. Prevalence rates were determined and compared using appropriate statistics at confidence of 95% (Ahmed *et al.*, 2011). Descriptive analyses were carried

out to compare the prevalence and co-infections of animal infections. Multiple correspondence analyses (MCA) were conducted to determine the relationship between sex, age, and location of livestock with the presence or absence of parasites. Multivariate analysis ANOVA on the other hand was utilized in determining variations between the trypanosome infection, tick infestations and gastrointestinal helminths.

One-way analysis of variance was used in determining various haematological variables of animals among infected and non-infected animals. Haematological values were expressed as means \pm standard deviations and compared among different groups. Characterization of trypanosome parasites were dependent on genetic analysis of bands formed by different isolates compared to known profiles. Those that presented identical profiles were considered to belong to the same genotype. The significance limit throughout the data analyses was $p \leq 0.05$.

CHAPTER FOUR

RESULTS

4.1 Study Population Statistics

The population statistics for each livestock examined for parasites under the study in Kerio Valley are summarized in table 4.1- 4.3.

Table 0:1: Summary of Livestock by sub-County

	Cattle	Sheep	Goats	Total
Marakwet Sub County	176	70	53	299
Keiyo Sub-County	109	14	45	168
Total animals sampled	285	84	98	467

A total of 467 animals were sampled from 4 strata i.e. Keiyo North, Keiyo South, Marakwet East and Marakwet West) along the upper Kerio Valley of Elgeyo-Marakwet County.

Table 0:2: Frequencies of animals by age group and sex

	<i>Sex of the animal</i>		<i>Age of the animal</i>			<i>TOTAL</i>
	Female	Male	Yearling	Weaner	Adults	
Cattle	221 (77.5%)	64 (22.5%)	23 (8.1%)	29 (10.2%)	213 (74.7%)	285
Sheep	58 (69%)	26 (30.95%)	13 (15.5%)	8 (9.5%)	63 (75%)	84
Goats	66 (67%)	32 (33%)	23 (23.5%)	10 (10.2%)	65 (66.3%)	98

Most livestock were female; 77.5%, 69% and 67% for cattle, sheep and goats respectively. In terms of age group, majority animals under the study were adults (Table 4.2).

The number of blood and stool samples collected from specific group of livestock are shown in Table 4:3. Less number of blood samples collected were less because, some blood samples clotted before storing in EDTA tubes.

Table 0:3: The number of animals from whom specimen were collected

Animal types	Cattle	Sheep	Goats	Total
Total blood samples	236	84	77	381
Total stool samples	285	84	96	465

Most of the animals under the study were observed to be apparently in good health conditions as confirmed by the animal health officer and livestock owners.

4.2 Prevalence of Intestinal Parasites in Livestock

4.2.1 Prevalence of intestinal parasites in cattle

Parasites found in faecal samples of cattle included protozoa and helminths as shown in Plate 4:1, 4:2 and 4:3. The larval stages found in the faecal sample were determined to belong to *Strongyloides* species, which hatch immediately the eggs were released. Figure 4.1 presents the prevalence of the parasites evaluated.



(a)



(b)

Plate 0:1: *Strongyloide* egg f and *Trychostrongylus* sp

(Source: Author, 2016)



Trichuris sp egg x200



Strongyloides sp larva x 200

Plate 0:2: *Trichuris* egg and *Strongyloides* larva

(Source: Author, 2016)



Plate 0:3 Protozoan oocyst x 200

(Source: Author, 2016)

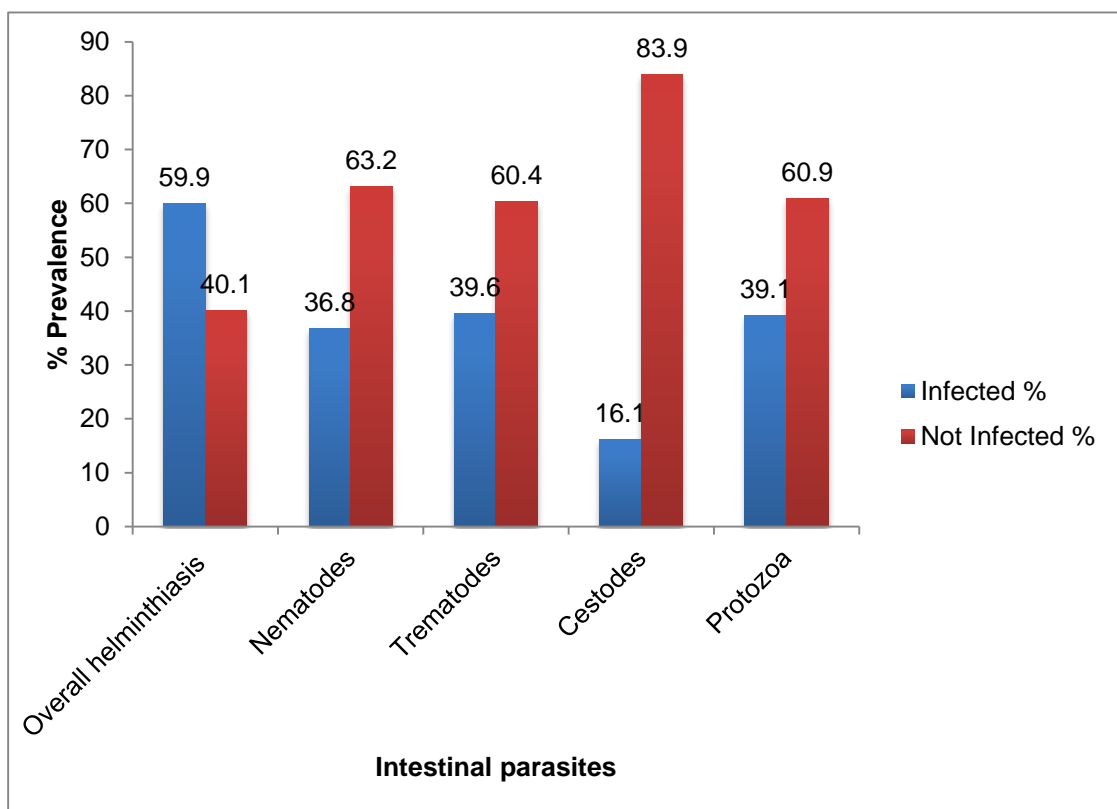


Figure 0:1: Prevalence of intestinal parasites in cattle

A multiple correspondence analysis (MCA) was conducted to determine the relationship between sex, age, and location of cattle with the presence or absence of helminth parasites. The model could explain about 51% of the variance in the original variables (inertia=0.505), with dimension one and two accounting for 52% and 49% of the variability, respectively while mean Cronbach's Alpha was 0.53. This suggested that the model fitted the data. The joint plot of category points is presented in Figure 4.2.

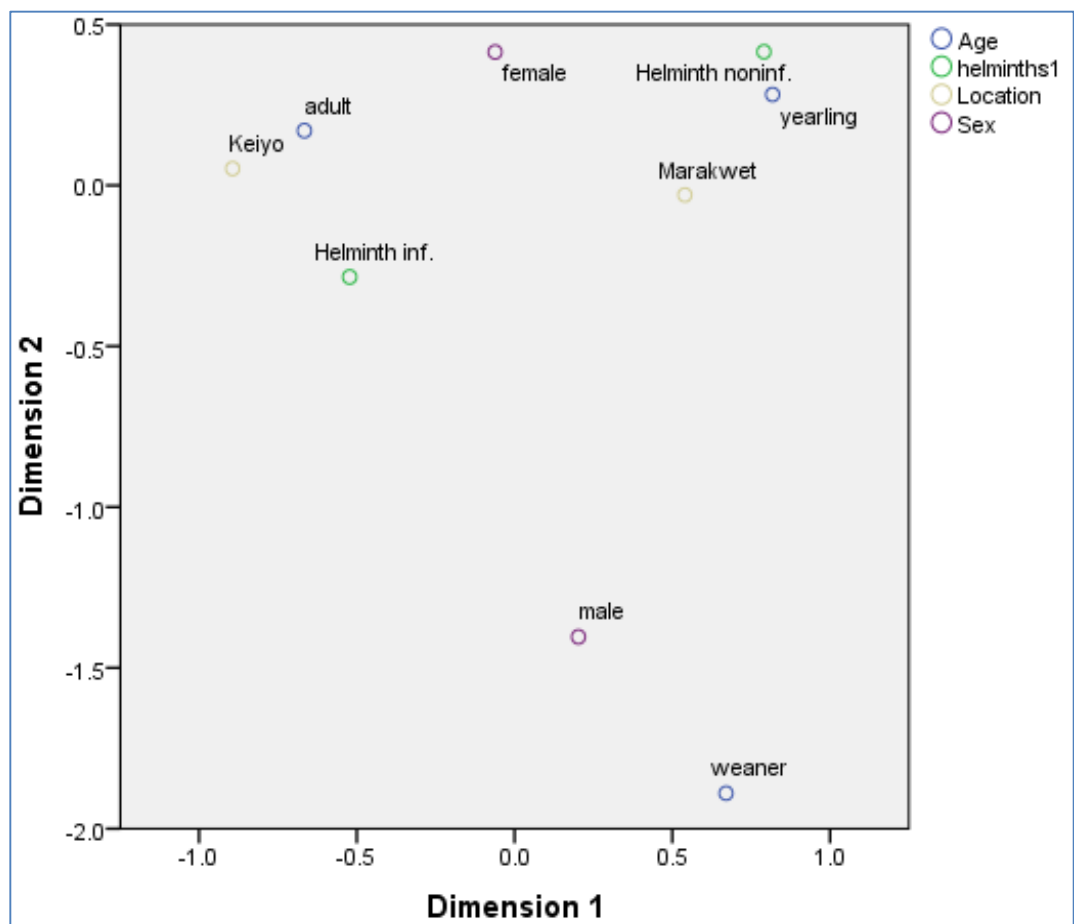


Figure 0:2: Relationship between age, sex, location and helminth infection in cattle

Key: noninf. = not infected; inf. = infected

'Helminth inf.' (infected) occurred near 'Keiyo' and 'adult' while 'Helminth noninf' (non-infected) aggregated near 'yearling' and 'Marakwet'. This showed that cattle

infected with helminthes were likely to be adults rather than yearlings. In addition, more infected cattle were found in Keiyo rather than in Marakwet. However, the sex of cattle did not appear to influence helminth prevalence as 'female' and 'male' were roughly equidistant between 'Helminth inf' and 'Helminth noninf'.

A MCA was run to determine the relationship between the cattle's sex, age, and location with the presence or absence of protozoa parasites. The model could explain about 56% of the variance in the original variables, with dimension one and two accounting for 57% and 56% of the variability, respectively while mean Cronbach's Alpha was 0.60. This suggested that the model fitted the data. The joint plot of category points is presented in Figure 4.3.

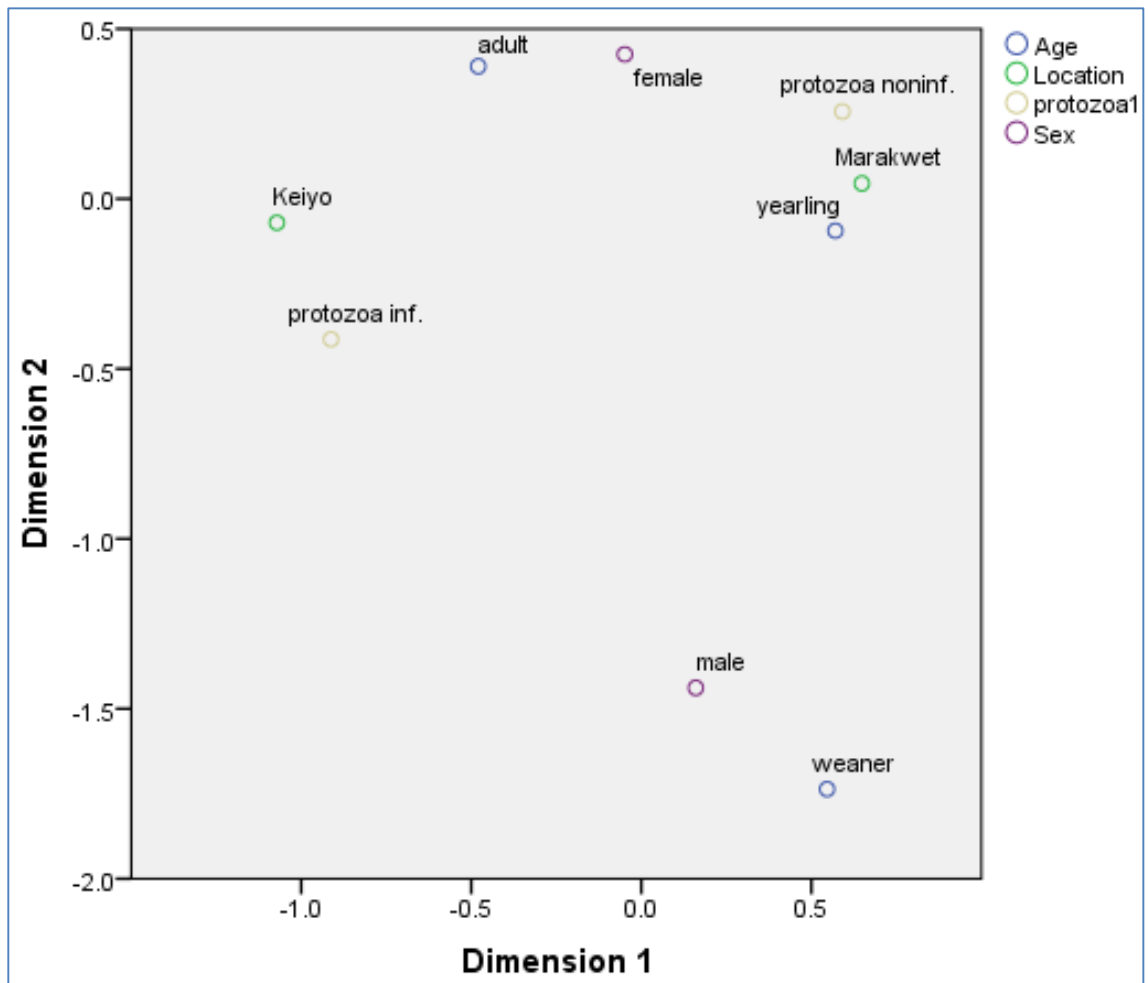


Figure 0:3: Relationship between age, sex, and location and protozoa infection in cattle

Key: noninf. = non infected; inf. = infected

‘Protozoa inf.’ (infected) occurred near ‘Keiyo’ while ‘protozoa noninf. (non-infected) was found near ‘Marakwet’, showing that, like for helminthes, protozoan parasites have a higher prevalence in cattle found in Keiyo compared to those in Marakwet sub county. However, cattle sex and age did not significantly affect Protozoa prevalence since ‘female’ and ‘male’ and ‘adult’ and ‘weaner’ were roughly equidistant from ‘protozoa inf.’ and ‘protozoa noninf.’ points.

Intestinal parasites were categorized into classes thus: nematodes, trematodes, cestodes and protozoa. Investigation of intestinal parasite categories in cattle showed 21% were

infected with co-infections with all possible parasite combinations under the study while 6% had triple combinations of intestinal parasite infections (Fig. 4.4)

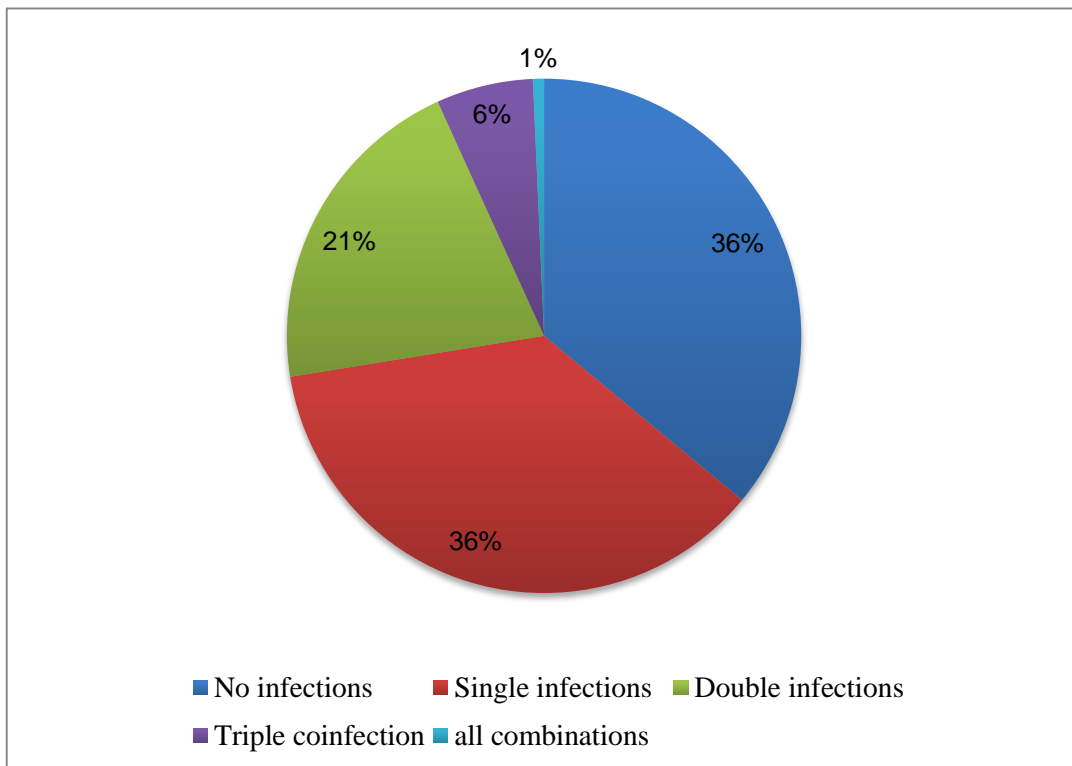


Figure 0:4: Co-infections of intestinal parasitic categories in cattle

4.2.2 Parasitism in goats

Ninety-eight goats constituting mostly small East African goats were sampled comprising of all age groups, both ewes and bucks. The prevalence of parasites in goats are presented in Fig 4.5 (n=98).

Intestinal helminths accounted the highest prevalence of 77% compared to protozoa (48%). Among the helminths, nematodes had the highest prevalence (75%) compared to trematodes and cestodes (both 18%). Overall parasite prevalence (whether the goat was infected with helminth or protozoa) was 85%.

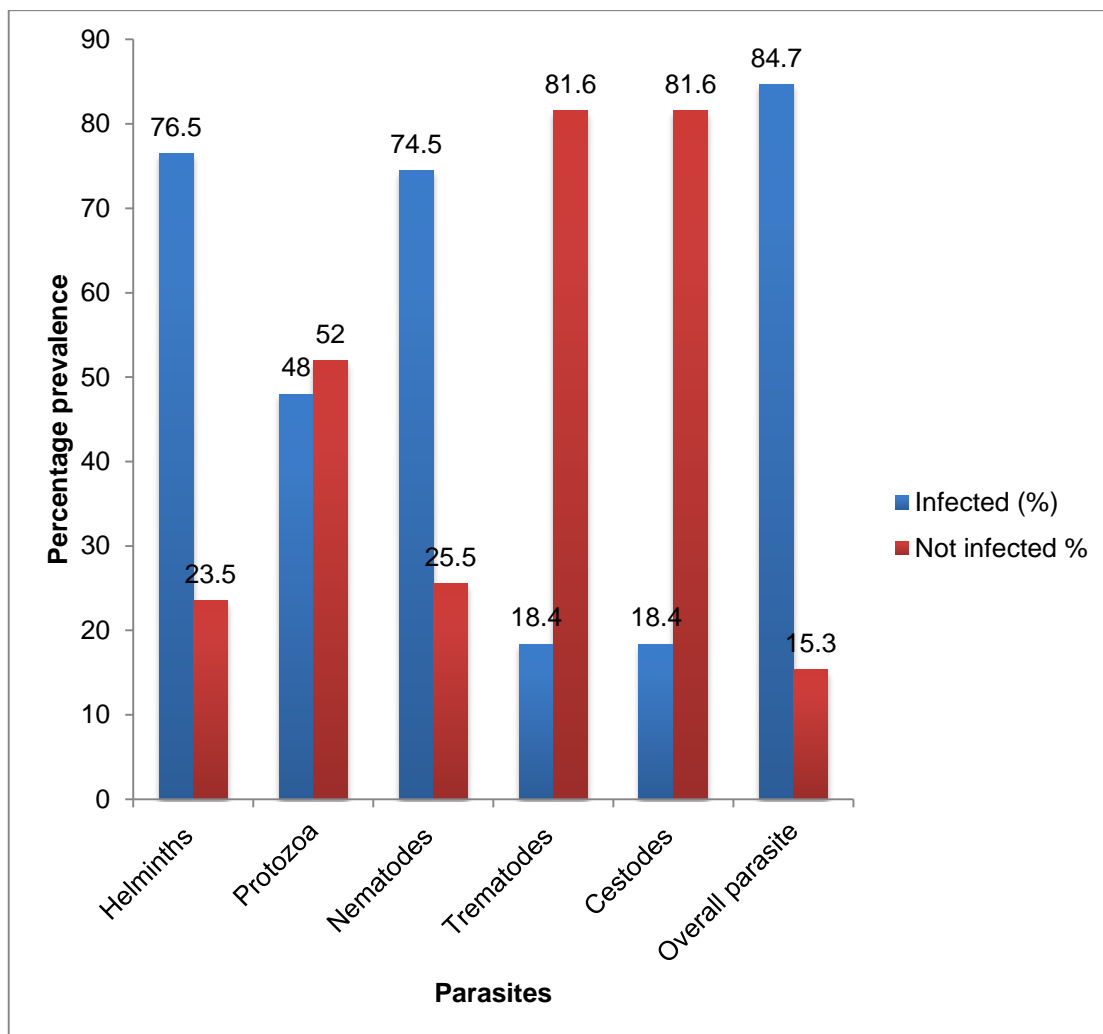


Figure 0:5: Prevalence of intestinal parasites in goats

A MCA was ran to determine the relationship between the cattle's sex, age, and location and the presence or absence of protozoan and helminth parasites. The model could explain about 55% of the variance in the original variables, with dimension one and two accounting for 56% and 54% of the variability, respectively while mean Cronbach's Alpha was 0.58. This suggested that the model fitted the data. The joint plot of category points is presented in Figure 4.6.

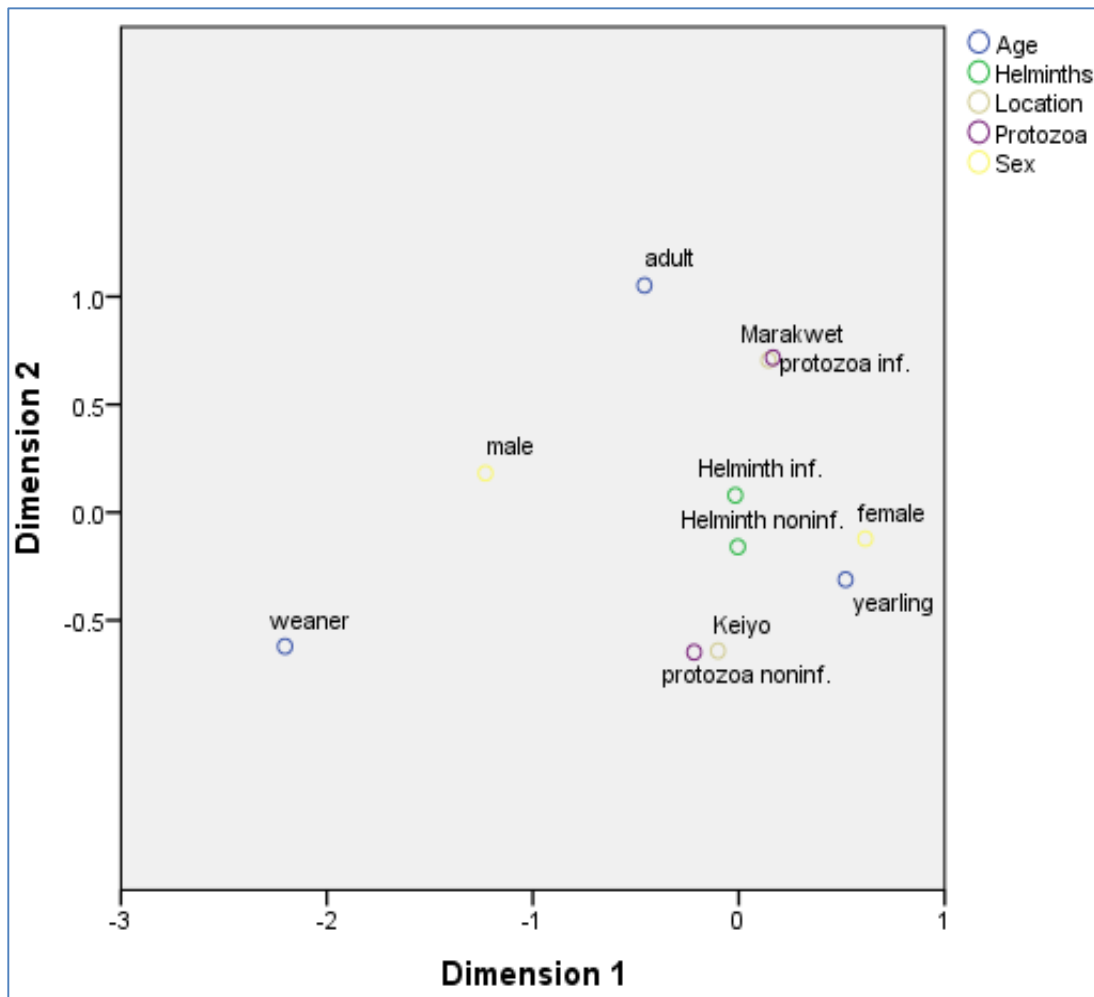


Figure 0:6: Relationship between goats' age, sex, location and parasite infection

Key: noninf. = non infected; inf. = infected

“Protozoa inf.” occurred near Marakwet while “protozoa noninf.” aggregated near Keiyo. This showed that the location of the goat affected the occurrence of the protozoan parasites, with goats in Marakwet having a higher prevalence compared to goats from Keiyo. Protozoan parasites are also likely to be more in adult goats (“adult” is nearer “protozoa inf.”) than in either yearlings or weaners. However, goats’ sex appeared to have no influence on the prevalence of protozoan parasites, as those infected with protozoa and non-infected were equidistant from both male and female animals.

“Helminth inf.” and “Helminth noninf.” occurred near each other and appeared to be in the middle of the covariates (Marakwet and keiyo, male and female, and adult, weaner and yearling). This suggested that the goats’ location, sex and age had no bias on helminth prevalence.

On investigation of concurrent infections, the proportion of double infections with all possible combinations of intestinal parasites i.e. nematodes, trematodes, cestodes and protozoa showed 35.7% prevalence, and only 4% of goats were infected with all groups of intestinal parasites (Fig. 4.7)

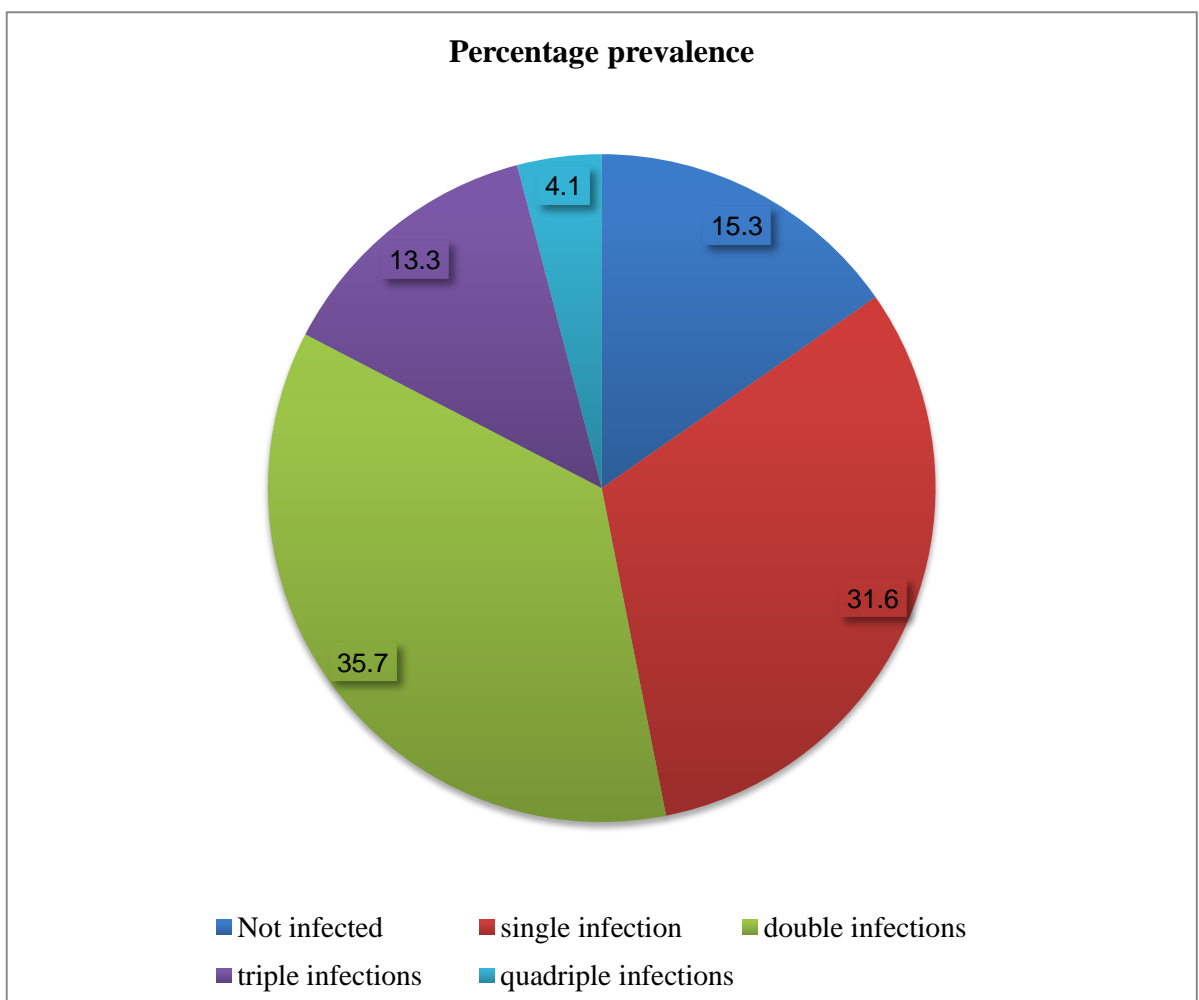


Figure 0:7: Co-infections in goats

4.2.3 Parasitism in Sheep

The prevalence of intestinal parasites in sheep are presented in Fig. 4.8 (n= 84). Overall parasite prevalence (whether the sheep was infected with helminth or protozoa) was 63%. Intestinal helminths had the highest prevalence (61%) compared to protozoa (32%). Among the helminths, nematodes had the highest prevalence (58%), followed by trematodes (14%) while cestodes were low (2%).

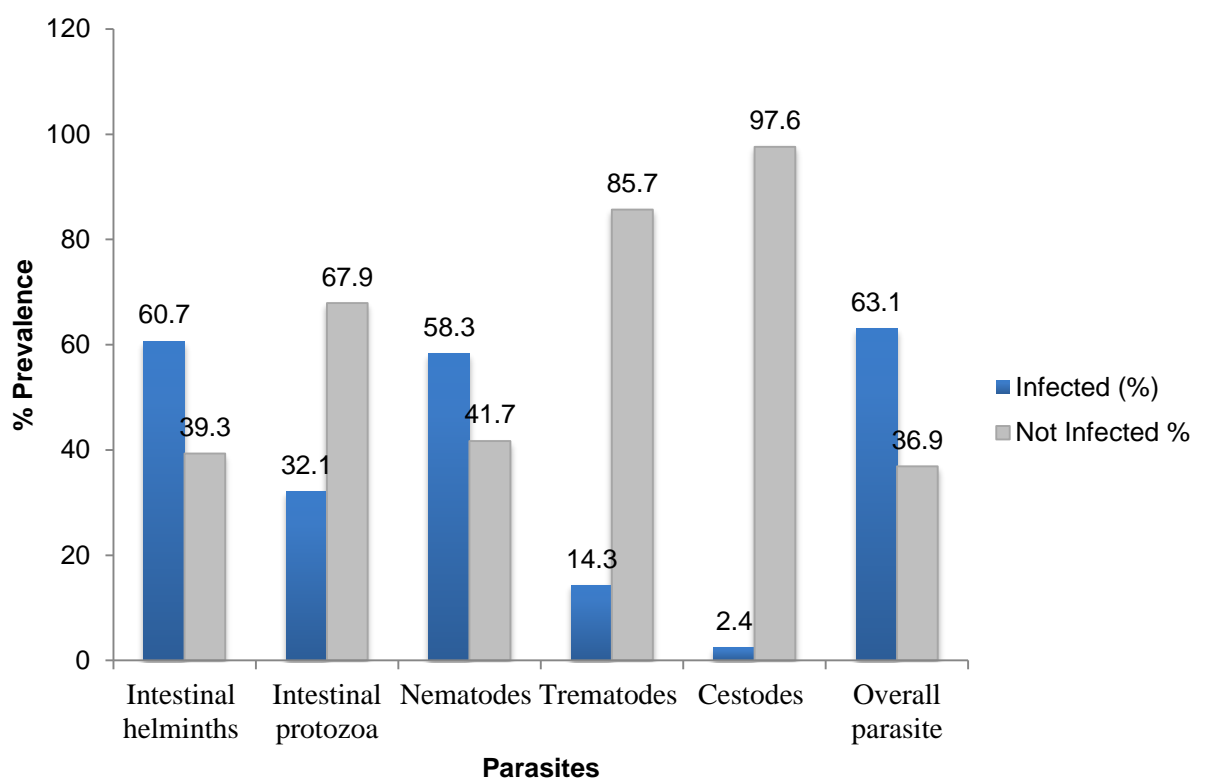


Figure 0:8: Prevalence of intestinal parasites in sheep

A MCA (multiple correspondence analyses) was conducted to determine the relationship between the sheep's sex, age, and location with the presence or absence of helminth parasites. The model could explain about 55% of the variance in the original variables (inertia=0.55), with dimension one and two accounting for 56% and 54% of the

variability, respectively while mean Cronbach's Alpha was 0.60. This showed that the model fitted the data. Figure 4.9 presents the joint plot of category points.

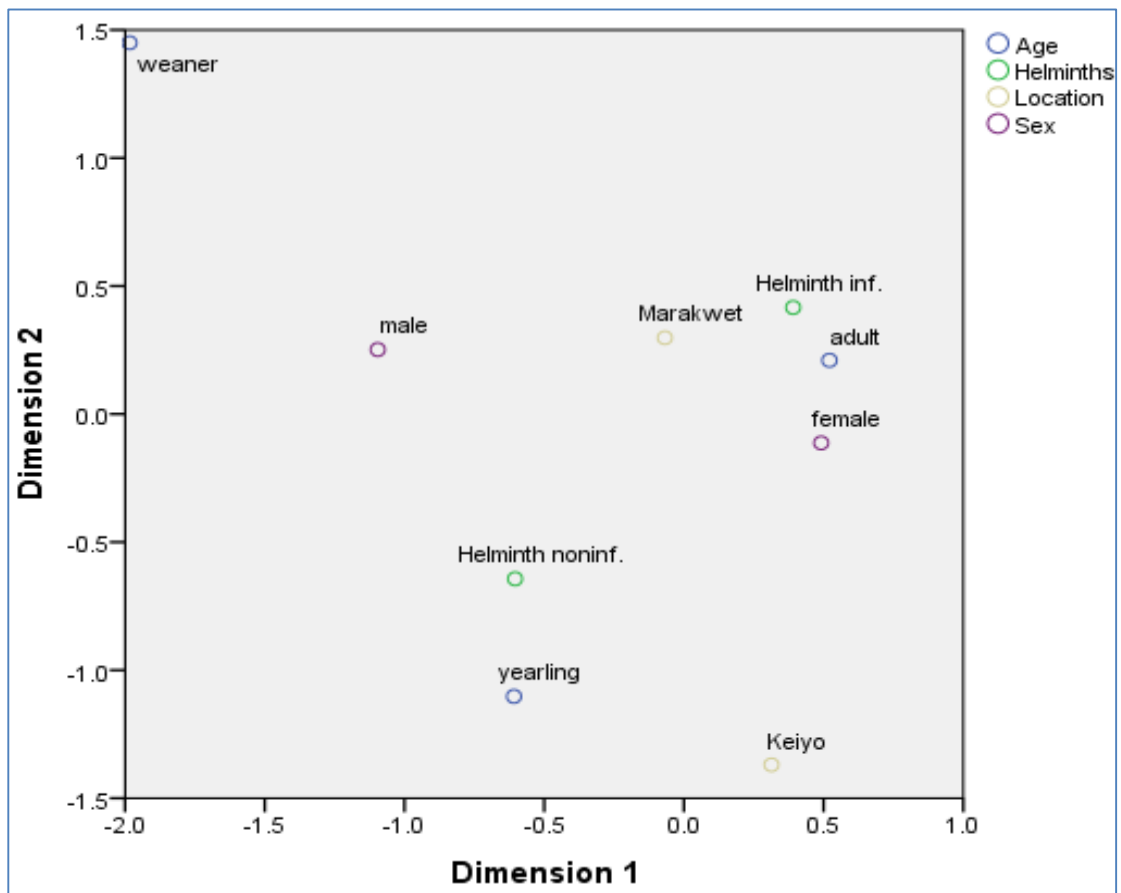


Figure 0:9: Relationship helminth infections and age, sex and location of sheep

Key: noninf. = non infected; inf. = infected

'Helminth inf.' (infected) was found near 'Marakwet', 'adult' and 'female' while 'Helminth noninf.' (non-infected) aggregated near 'yearling'. The results suggested that sheep were likely to be infected with helminths than those from Marakwet, adults and female as opposed to yearlings or sheep from Keiyo.

A MCA was conducted to determine the relationship between the sheep's sex, age, and location with the presence or absence of protozoa parasites. The model explained 52% of the variation in the original variables, with dimension one and two accounting for 53%

and 51% of the variability, respectively while mean Cronbach's Alpha was 0.56. The results indicated that the model fitted the data. The joint plot of category points is presented in Figure 4.10

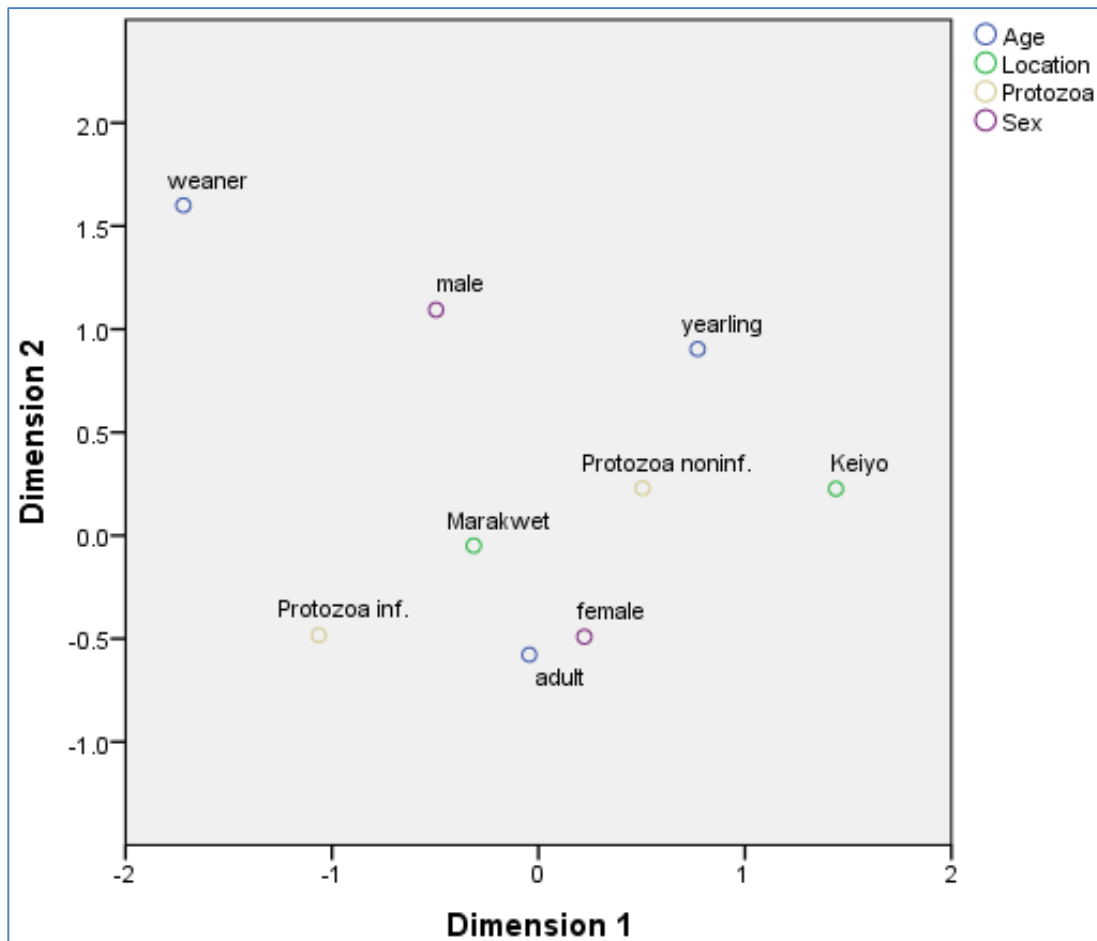


Figure 0:10: Relationship between protozoan infection and age, sex, and location of sheep

Key: noninf. = non infected; inf. = infected

'Protozoa inf.' (infected) occurred near 'Marakwet' and 'adult' while 'protozoa noninf.' (non-infected) was found near 'Keiyo' and 'yearling' showing that, like for helminths, protozoan parasites have a higher prevalence in adult sheep located in Marakwet compared to yearlings found in Keiyo. However, sheep's sex appeared not to significantly affect Protozoa prevalence since 'female' and 'male' were roughly equidistant from 'protozoa infected' and 'protozoa non-infected'.

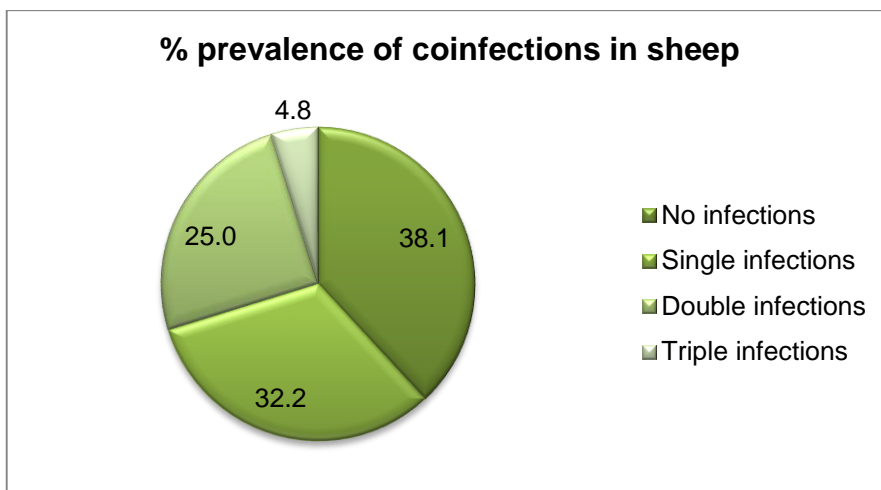


Figure 0:11: Proportion of intestinal parasites in sheep

Only 29.8% of sheep investigated showed infection with more than one category of intestinal parasites, while 32% had one single infection (Fig 4.11)

4.3 Prevalence of various nematode infections

The eggs of nematode species detected were Trichostrongyles, which comprises of several *genera* including *Bunostomum*, *Haemonchus*, *Strongyloides* and *Trichostrongylus* species. Strongyloide and Bunostomum as shown in Plate 4:4.

Embryonated Strongyloide egg from goat (x200)

Bunostomum egg from cattle (x200)

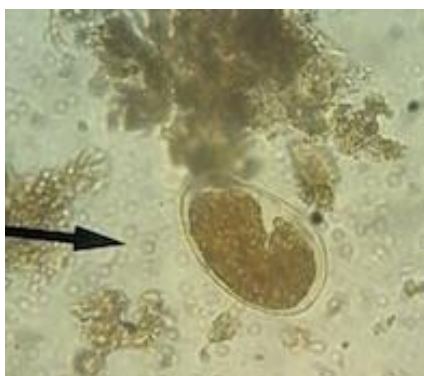


Plate 0:4: Picture showing observed nematode eggs

Source: Author, 2017

Table 4:4 shows various nematodes diagnosed and their prevalence.

Table 0:4: Prevalence of specific nematode infections in livestock

<i>Nematodes (genera)</i>	<i>Sheep</i>	<i>Goats (N = 93)</i>	<i>Cattle</i>
	Infected (N) (%)	Infected (N) (%)	Infected (N) (%)
<i>Trichostrongylous</i>	20 (83) 24.1%	31 (33.3%)	16 (284) 5.63%
<i>Strongyloides</i>	33 (83) 39.8%	42 (45.2%)	52 (284) 18.3%
<i>Ascaris</i>	12 (66) 18.2%	18 (19.35%)	30 (211) 14.2%
<i>Bunostomum</i>	28 (66) 42.4%	34 (36.6%)	24 (284) 8.45%
<i>Trichuris</i>	1 (66) 1.5%	3 (3.2%)	1 (73) 1.37%
<i>Heamonchus</i>	-	-	4 (284) 1.4%
<i>Enterobius</i>	-	6 (6.45%)	5 (211) (2.37
<i>Avitellina</i>	1 (17) 5.9%	-	6 (73) (8.2%)
<i>Toxoscaris</i>	-	-	1 (73) 1.4%
<i>Ostergia</i>	-	-	1 (73) 1.4%

Other nematode infections observed included *Acutellina centripunctata*, *Cooperia pectinita*, *Chabertia ovina*, *Oesophagostomum columbianum*, *neoascaris vitulorum*, *Skijahinema ovis*, *Gongylomena pulchrum*, and *Physaloptera canis*

4.4 Prevalence of platyhelminthes

Trematodes diagnosed from faecal samples included *Schistooma*, *Fasciola* and *D. dentriticum* the common one being *Fasciola hepatica*. *Schistosoma* species identified using microscopy includes *Schistosoma bovis*, *S. indicum* and *S. japonicum* suggesting the presence of their vectors; fresh water snails in Kerio River, which the animals take water from. The study also demonstrated the presence of *Eurytrema pancreatum*, eggs in the faecal samples (Table 4:5).

Table: 0:5: Prevalence of platyhelminthes

	Sheep	Goats	Cattle
	Infected (N) (%)	Infected (N) (%)	Infected (N) (%)
<i>Taenia</i>	1 (66) 1.52%	7 (93) 7.53%	31 (211) 14.7%
<i>Fasciola</i>	8 (66) 12.1%	18 (93) 19.4%	44 (284) 15.5%
<i>Schistosoma</i>	5 (83) 6%	-	62 (284) 21.8%
<i>Dicrocoelium</i>	-	-	22 (284) 7.75%
<i>Diphylidium</i>	-	-	15 (211) (7.1%)
<i>Haemochus</i>	-	12 (93) 12.9%	18 (211) 8.5%

The prevalence of cestodes was relatively lower than other parasites in all types of livestock, *Taenia* and *Hymenolepis* being the only cestodes found. Among the infected animals, *Hymenolepis* species was more prevalent cestode that infected 18 cattle (8.5%), and 12 goats (12.9%).

4.5 Prevalence of Haemoparasites

The results regarding the prevalence of haemoparasites by microscopy are presented in the Table 4:6 below;

Table 0:6: Bovine haemoparasites

	Infected (N=72)	% Prevalence
<i>Theileria parva</i>	3	4.2
<i>Babesia bigemina</i>	1	1.4
<i>Trypanosomes</i>	2	2.8
<i>Anaplasma marginale</i>	2	2.8
<i>Theileria + Anaplasma</i>	1	1.4

Haemoparasites were only observed in cattle blood using microscopy but none were observed in sheep or goats. *Theileria parva* in cattle was the commonest with 100% incidence occurring in adults.

4.6 Identification of trypanosomes using PCR

A total of 90 blood samples in FTA cards were analysed for the presence of trypanosomes. Trypanosome positive samples were detected by PCR using a 4 panel of primers that identify them to species level. The analysis showed 29 positive samples for *T. brucei* and *T. congolense* as indicated in Fig. 4.12.

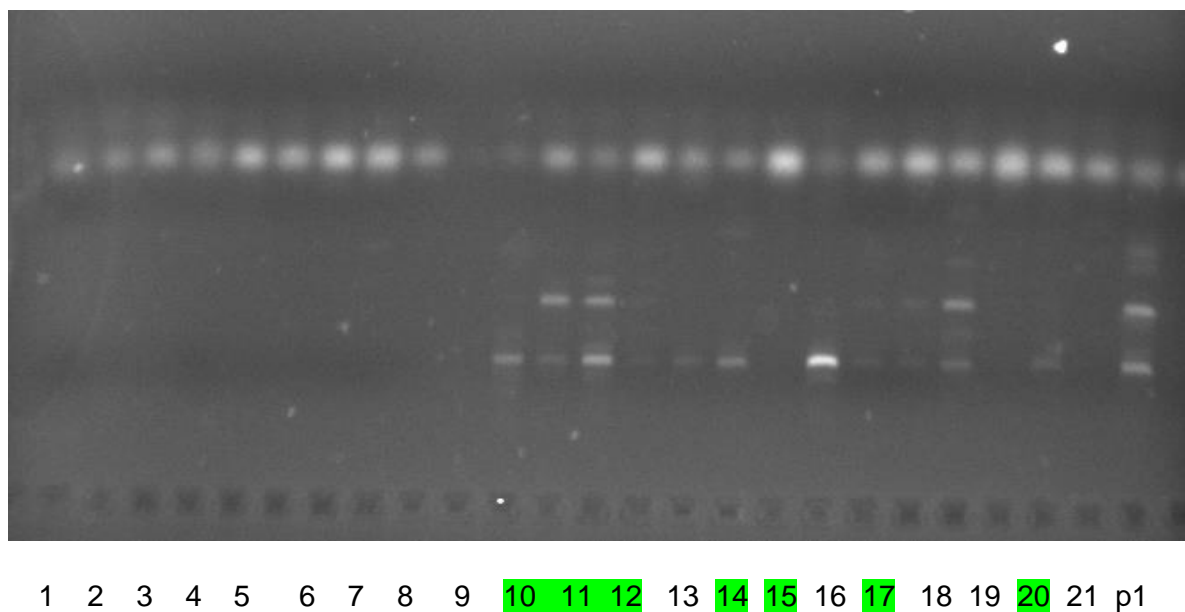


Figure 0:12: Trypanosome DNA bands

Trypanosome species identified through PCR were *Trypanosoma congolense* and *T. brucei*. The results of trypanosomes causing AAT identified are presented in the following table (Table 4.7).

Table 0:7: Prevalence of trypanosome by microscopy and PCR

Method	T congolense	T brucei	Total
Positive by PCR	10 (11.1%)	19 (22.2%)	29 (32.2%) N=90
Positive by microscopy			2 (2.2%) N =90

PCR analysis of trypanosome infection using species-specific primers showed positive identification of 29 (32.2%). Trypanosomes with higher prevalence was *T. brucei* with equally high presence *T. congolense* showing remarkable results considering that this in an area that trypanosomosis has not been previously reported.

Trypanosome infection in cattle and sheep are represented in table 4.8

Table 0:8: Prevalence of *Trypanosoma sp* in cattle and sheep

	<i>T. congolense</i>	<i>T. brucei</i>	Total prevalence rate (N=90)
Cattle	12	14	26 (28.9%)
Sheep	-	3	3 (3.3%)

Cattle were more likely to be infected with trypanosomes than the sheep (3.3%). *Trypanosoma brucei* was the most prevalent trypanosome found in livestock. No co-infection of trypanosome infections was identified.

4.7 Other Parasites

Intestinal protozoan parasites were also observed. These included coccidians, amoebas, and ciliate. Ectoparasites included mites and ticks.

Table 0:9: Table showing the prevalence of various parasites in the stool samples

	Coccidia		Amoebas		Ciliates	
Cattle	19 (N= 284)	6.7%	89 (N= 211)	42.2%	5 (N=211)	2.4%
Sheep	7 (N= 83)	8.4%	22 (N = 66)	33.3%	7 (N= 66)	10.7%
Goats	11 (N = 93)	11.8%	41 (N = 93)	44.1%	9 (N = 93)	9.7%

Intestinal amoebas were frequently found in livestock followed by coccidian species. Coccidian species included *Eimeria* species and *Isospora belli*.

There were four different species of ticks identified in cattle in the study area. The ticks frequently observed included red ear tick (*Rhipicephalus appendiculatus*). Other species identified during the sampling periods included *Hyalomma sp*, *Boophilus* and *Amblyomma variegatum* (Plate 4:5 and Plate 4:6). Mites identified from skin scrapings were *Chorioptic mange* and *Psoroptes mange* mites of visibly infected animals. The mites' infestations were found in Tot study site.

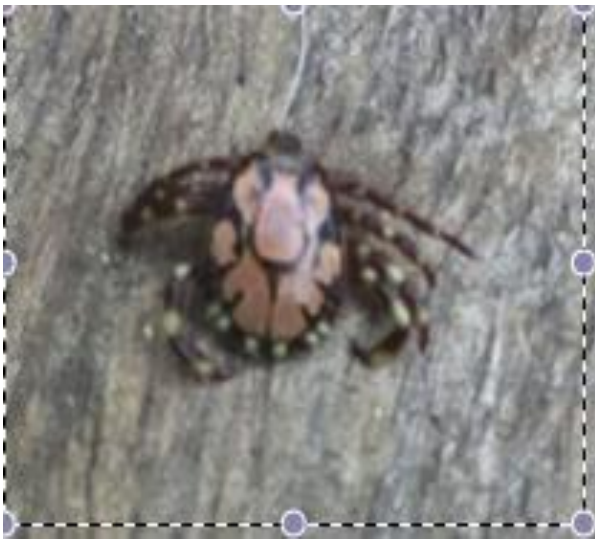


Plate 0:5: *Amblyomma variegatum*

(Source: Author, 2016)



Plate 0:6: Female and Male *Amblyomma variegatum* sp

4.8 Haematological Parameters

The tables 4.10 through to 4.25 below presents the overall haematological parameters in livestock in the study, the standard error of means. The tables shows the haematological parameters segregated according to whether the animal was infected (any helminth or protozoa) or non-infected, and the standard reference values for the blood parameters as obtained from Merck's Manual (2012) and Ministry of Health (MoLD, 2011).

4.8.1 Haematological parameters in cattle and factors that affect their values

(i) Haematological parameters for parasite infected and non-infected cattle

Table 4.10 represents the overall haematological parameters in cattle in the study area, the standard error of means in comparison with standard reference ranges between the infected and non-infected with parasites. Parameter levels in infected and non-infected cattle were compared using independent samples T tests while one - sample *T*-tests were

used to analyse differences between parameter levels (for infected and non-infected cattle in each case and reference values cited by Merck's Manual (2012).

Table 0:10: Haematological parameters of cattle in comparison with infection and reference values

<i>Parameter</i>	<i>Overall</i>	<i>Infected</i>	<i>Non-infected</i>	<i>Reference</i>
<i>(n=217)</i>	Mean±SEM	Mean ± SEM	Mean ± SEM	<i>range</i>
WBC (10e3/ul)	15.10±0.80	15.15±.97 ^a	14.9± 1.39 ^a	4-12 ^b
Lymp (%)	55.31±1.5	56.59±1.88 ^a	53.37±2.55 ^a	45-75 ^a
Monocytes (%)	9.56±1.3	8.20±1.50 ^a	11.58±2.42 ^a	0-8 ^a
RBC (10e3/ul)	6.09±0.08	6.06±0.11 ^a	6.12±0.14 ^a	5-10 ^a
HGB (g/dL)	9.72±0.11	9.66±0.14 ^a	9.79±0.19 ^a	8-15 ^a
HCT (%)	27.50±0.57	27.68±0.83 ^a	27.08±0.63 ^a	24-46 ^a
MCV (fl)	44.37±0.34	44.31±0.42 ^a	44.44±0.49 ^a	40-60 ^a
MCH (pg)	16.61±0.40	16.32±0.30 ^a	17.07±0.83 ^a	11-17 ^a
PLT (10e3/uL)	245.73±17.2	238.55±21.73 ^a	254.69±24.89 ^a	100-800 ^a
MPV (fL)	8.9±0.40	9.31±0.54 ^a	8.42±0.26 ^a	3.5-6.5 ^b

Key. Means with similar letters in a row are not significantly different by *T* tests

The results indicated that all parameter levels did not differ significantly ($p < 0.05$) in infected and non-infected cattle. Exceptionally, only two parameters, White blood cell counts (mean 15.10) and Mean Platelet Volume (mean 8.9) were significantly higher in cattle in the study compared with standard reference values (4-12 and 3.5-6.5, respectively). All the other measured parameters were not significantly different ($p < 0.05$) from the reference values.

(ii) *Haematological parameters according cattle sex*

Since Section 4.8.1 did not establish significant differences in the parameters between the infected and non-infected animals, the factors were presented (Table 4:11) without correcting for whether the animals were infected or not.

Table 0:11 Haematological factors of cattle by sex

<i>Parameter</i>	<i>Female (n=168)</i>	<i>Male (n=49)</i>	<i>p value</i>
(n=217)	Mean±SEM	Mean ± SEM	
WBC (10e3/ul)	14.60±0.94	16.81±1.45	0.25
Lymp (%)	53.98±1.80	59.68±2.77	0.12
Monocytes (%)	10.79±1.67	5.52±1.12	0.01
Gran (%)	15.06±3.06	9.09±1.85	0.29
RBC (10e3/ul)	6.07±0.09	6.16±0.18	0.63
HGB (g/dL)	9.76±0.13	9.59±0.20	0.52
HCT (%)	27.97±0.71	25.89±0.73	0.04
MCV (fl)	44.99±0.37	42.27±0.56	P<.0001
MCH (pg)	16.72±0.41	16.20±0.78	0.56
MCHC	36.67±0.56	38.71±1.82	0.15
PLT (10e3/uL)	232.13±17.6	292.08±41.76	0.19
MPV (fL)	8.88±0.44	9.33±0.43	0.59

Key: The *p* values in bold are significant

Independent samples T tests were conducted to determine on whether the haematological parameters differed significantly in male and female cattle. The tests showed that three factors, Monocytes %: $t(215) = 2.61, p=0.01$; HCT: $t(215) = 2.04, p=0.04$; and MCV: $t(215) = 3.59, p<0.0001$) were found to be significantly higher in female than in male cattle.

However, the rest of the parameters did not differ (at $p < 0.05$) significantly between the two sexes.

(iii) *Haematological parameters according to cattle age*

Haematological parameters in cattle of different ages are shown in Table 4.12. There were no outliers in the data, as assessed by inspection of a boxplot. Scores for each level of age for all parameters were normally distributed, as assessed by Shapiro-Wilks test ($p > 0.05$).

Table 0:12 Haematological factors according to cattle age

	Yearling (n=84)	Weaner (n=28)	Adult (n= 105)
Parameter (n=217)	Mean\pmSEM	Mean \pm SEM	Mean \pm SEM
WBC (10e3/ul)	11.79 \pm 0.95 ^a	16.06 \pm 2.51 ^b	17.49 \pm 1.27 ^b
Lymphocytes (%)	48.58 \pm 2.70 ^a	58.94 \pm 3.63 ^b	60.89 \pm 1.76 ^b
Monocytes (%)	16.83 \pm 2.70 ^a	7.94 \pm 2.72 ^b	2.91 \pm 0.51 ^b
Granulocytes (%)	25.25 \pm 5.22 ^a	9.13 \pm 2.09 ^b	3.61 \pm 0.25 ^b
RBC (10e3/ul)	6.27 \pm 0.10 ^a	6.16 \pm 0.31 ^a	5.93 \pm 0.13 ^a
HGB (g/dL)	9.94 \pm 0.15 ^a	9.17 \pm 0.43 ^a	9.69 \pm 0.16 ^a
HCT (%)	28.35 \pm 0.50 ^a	25.22 \pm 1.38 ^a	27.43 \pm 1.04 ^a
MCV (fl)	45.43 \pm 0.43 ^a	40.93 \pm 0.98 ^b	44.46 \pm 0.46 ^a
MCH (pg)	16.67 \pm 0.74 ^a	15.16 \pm 0.37 ^a	16.94 \pm 0.45 ^a
MCHC	35.26 \pm 0.27 ^a	37.36 \pm 0.98 ^a	38.56 \pm 1.17 ^b
PLT (10e3/uL)	247.73 \pm 22.1 ^a	410.18 \pm 77.99 ^b	199.84 \pm 18.9 ^a
MPV (fL)	8.20 \pm 0.21 ^a	9.83 \pm 0.64 ^a	9.39 \pm 0.70 ^a

Key. Means with similar letters in a row are not significantly different Tukey's HSD or Games Howell tests

The Levene's Test indicated error variance to be equal across the groups for all parameters (all $p > 0.05$), except lymphocyte % ($F=8.40, p<0.0001$), monocyte % ($F=24.72, p<0.0001$), granulocyte % ($F=8.63, p<0.0001$), RBC ($F =4.13, p =0.02$), and PLT ($F = 13.65, p<0.0001$)

Consequently, post-hoc analysis was conducted with Tukey's Honestly Significant Difference (HSD) test when the assumption of equality of variance was tenable and with Games-Howell test when it was violated.

Analysis of variance showed significant differences in age groups of cattle for WBC, $F(2, 214) = 5.71, p=0.004$, lymphocyte %, $F(2, 214) = 8.04, p<0.0001$, monocyte %, $F(2, 214) = 13.94, p<0.0001$, granulocyte %, $F(2, 214) = 10.09, p<0.0001$, and MCV, $F(2, 214) = 9.97, p<0.0001$. Others were MCHC, $F(2, 214) = 3.32, p=0.04$ and PLT, $F(2, 214) = 8.78, p<0.0001$. However, the tests revealed no significant differences ($p<0.05$) in cattle age groups for RBC, HGB, HCT, MCH, and MPV.

Post hoc analysis showed that WBC in general and lymphocytes were significantly higher in both weaners and adults compared to yearlings. On the other hand, monocytes and granulocytes were significantly higher in yearlings than in both weaners and adults. While MCV was lower in weaners compared to both yearlings and adults MCHC was higher in adults compared to other age groups. However, PLT were significantly higher in weaners relative to the other age groups.

(iv) Cattle haematological parameters according to location

Table 4.13 presents haematological parameters in cattle presented according to location. There were no outliers in the data, as assessed by inspection of a boxplot. Scores for each level of age for all parameters were normally distributed, as assessed by Shapiro-Wilks

test ($p > 0.05$). The Levene's Test indicated error variance to be equal across the groups for all parameters (all $p > 0.05$), except WBC ($F=4.63$, $p=0.03$), monocyte % ($F=22.78$, $p<0.0001$), granulocyte % ($F=7.85$, $p=0.006$), RBC ($F =6.44$, $p =0.01$), and PLT ($F = 18.93$, $p<0.0001$). Consequently, for variables that violated the assumption of equality of variance, t – values with adjusted degrees of freedom were reported.

Table 0:13: Haematological factors according to location of cattle

<i>Parameter</i>	<i>Keiyo (n=55)</i>	<i>Marakwet (n=162)</i>	
<i>(n=217)</i>	Mean+SEM	Mean + SEM	p value
WBC (10e3/ul)	17.26+1.12	14.37+1.00	0.06
Lymphocytes (%)	56.07+2.90	55.06+1.80	0.77
Monocytes (%)	2.37+0.19	11.94+1.71	P<.0001
Granulocytes (%)	4.34+0.38	16.76+3.15	P<.0001
RBC (10e3/ul)	6.14+0.12	6.07+0.11	0.67
HGB (g/dL)	9.96+0.19	9.64+0.13	0.17
HCT (%)	26.82+1.33	27.74+0.62	0.49
MCV (fl)	41.82+0.51	45.24+0.37	P<.0001
MCH (pg)	17.30+1.11	16.37+0.31	0.27
MCHC	39.24+0.35	36.42+0.78	0.04
PLT (10e3/uL)	118.58+10.66	289.16+20.94	P<.0001
MPV (fL)	8.32+1.27	9.21+0.21	0.28

The values in bold showed significant difference

Independent samples t tests were conducted to determine whether the haematological parameters differed significantly in Keiyo and Marakwet cattle. The tests indicated that four parameters, Monocytes %: $t(144.53) = -5.55$, $p=0.002$; granulocytes: $t(145.17) = -$

3.91, $p=0.02$; MCV: $t(116.24) = -5.35$, $p<0.0001$), and PLT: $t(211.59) = -7.26$, $p<0.0001$) were found to be significantly higher in cattle in Marakwet compared to those in Keiyo. However, MCHC, $t(215) = 2.06$, $p<0.04$) was found to be significantly higher in Keiyo cattle (39.24) relative to cattle from Marakwet (36.42). However, the rest of the parameters did not differ (at $p<0.05$) significantly between cattle in the two locations.

4.8.2 Haematological parameters in goats and factors that affect their values

(i) Haematological parameters for parasite infected and non-infected goats

Table 4.14 presents the overall haematological parameters in goats in the study, the standard error of means, and 95% confidence intervals for the means. The table also shows the parameters segregated according to whether the animal was infected (by any parasite, helminth or protozoa) or non-infected, and the standard reference values for the parameters. There were no outliers in the data, as assessed by inspection of a boxplot. Parameter scores were normally distributed, as assessed by Shapiro-Wilks test (all had $p > 0.05$). In addition, the values for skew and kurtosis for all the factors were within the benchmark ± 2.0 (Field, 2005). Levene's Tests indicated error variances to be equal across the groups of infected and non-infected goats (all $p > 0.05$). Parameter levels in infected and non-infected goats were compared using independent samples T tests while one - sample T-tests were used to analyse differences between parameter levels (for infected and non-infected cattle in each case) and reference values.

Table 0:14: Overall haematological parameters in goats

<i>Parameter</i>	<i>Overall</i>	<i>Infected</i>	<i>Non-infected</i>	<i>Reference</i>
<i>(n=217)</i>	Mean\pmSEM	Mean \pmSEM	Mean \pmSEM	<i>range</i>
WBC (10e3/ul)	21.99 \pm 1.50	22.81 \pm 1.78 ^a	19.06 \pm 2.30 ^a	4-13 ^b
Lymp (%)	65.94 \pm 2.52	65.67 \pm 2.84 ^a	67.16 \pm 7.27 ^a	50-70 ^a
Monocytes (%)	2.62 \pm 0.23	2.63 \pm 0.28 ^a	2.58 \pm 0.61 ^a	0-4 ^a
Gran (%)	3.55 \pm 0.57	3.77 \pm 0.70 ^a	2.90 \pm 0.95 ^a	
RBC (10e3/ul)	1.82 \pm 0.15	1.76 \pm 0.15 ^a	2.17 \pm 0.59 ^a	8-18 ^b
HGB (g/dL)	7.99 \pm 0.19	7.91 \pm 0.22 ^a	8.42 \pm 0.46 ^a	8-12 ^a
HCT (%)	6.53 \pm 0.68	6.29 \pm 0.72 ^a	7.97 \pm 2.54 ^a	
MCV (fl)	33.36 \pm 0.49	33.34 \pm 0.56 ^a	33.42 \pm 1.27 ^a	16-25 ^b
MCH (pg)	69.53 \pm 5.95	70.21 \pm 6.58 ^a	73.27 \pm 18.1 ^a	5.2-8 ^b
MCHC	216.54 \pm 21.5	217.02 \pm 23.89 ^a	238.92 \pm 62.76 ^a	30-36 ^b
RDWCV	11.37 \pm 0.33	11.32 \pm 0.37 ^a	11.46 \pm 0.96 ^a	
RDWSD	15.14 \pm 1.66	15.09 \pm 1.97 ^a	15.31 \pm 3.07 ^a	
PLT (10e3/uL)	2025.5 \pm 642	2056.7 \pm 692.7 ^a	1921.7 \pm 1854.7	300-600 ^b
MPV (fL)	10.03 \pm 1.11	10.13 \pm 1.25 ^a	9.70 \pm 2.90 ^a	
PCT	2.82 \pm 0.96	2.78 \pm 1.01 ^a	2.94 \pm 2.89 ^a	22-38 ^b
PDW	13.62 \pm 1.02	13.74 \pm 1.28 ^a	13.20 \pm 1.56 ^a	

Key. Means with similar letters in a row are not significantly different by T tests

The results indicated that all blood parameter levels did not differ significantly (at $p < 0.05$) in infected and non-infected goats. One- sample T-tests indicated that seven parameters were significantly different in goats in the study compared with reference values. Four of

them, WBC, MCV, MCH, and MCHC were significantly higher in goats (both infected and non-infected) compared with the standard values whereas RBC, PLT, and PCT were significantly lower in goats (both infected and non-infected) compared with reference values.

(ii) *Haematological parameters according to sex of goat*

Table 4.15 presents haematological parameters in goats presented according to sex. The parameters are presented without correcting for whether the animals were infected or not since Section 4.3.2 did not establish significant differences in the parameters between the infected and non-infected animals, the factors were presented without correcting for whether the animals were infected or not.

Scores for each level of sex for all parameters were normally distributed, as assessed by Shapiro-Wilks test ($p > 0.05$). The Levene's Test indicated error variance to be equal across the groups for all parameters (all $p > 0.05$), except lymphocytes % ($F=23.42$, $p=0.008$), HGB ($F=4.65$, $p<0.03$), MCH ($F=22.09$, $p<0.0001$), and MCHC ($F=4.13$, $p<0.0001$).

Independent samples *T*-tests were conducted to determine whether the haematological parameters differed significantly in male and female goats. The tests indicated that four parameters, Monocytes %: $t(74) = -2.81$, $p=0.007$; granulocytes %: $t(74) = -2.08$, $p=0.04$; MCH: $t(32.17) = -2.57$, $p=0.02$; and MCHC: $t(32.35) = -2.52$, $p = 0.01$ were found to be significantly higher in male compared with female goats. For MCH and MCHC, p values for t values with corrected degrees of freedom were reported as the equality of variance assumption had been violated. However, the rest of the parameters did not differ (at $p<0.05$) significantly between the two sexes.

Table 0:15: Haematological factors according to goat sex

Parameter (n=76)	Female (n=50)	Male (n=26)	p value
	Mean±SEM	Mean ± SEM	
WBC (10e3/ul)	20.280±1.61	25.31±3.04	0.11
Lymphocytes (%)	66.55±2.56	64.11±5.69	0.70
Monocytes (%)	2.18±0.21	3.50±0.51	0.007
Granulocytes (%)	2.74±0.39	5.16±1.42	0.04
RBC (10e3/ul)	1.95±0.19	1.56±0.23	0.22
HGB (g/dL)	7.88±0.27	8.19±0.24	0.45
HCT (%)	7.06±0.89	5.52±1.04	0.29
MCV (fl)	33.71±0.61	32.68±0.80	0.32
MCH (pg)	56.94±5.02	93.74±13.43	0.02
MCHC	171.96±18.2	302.25±48.27	0.01
RDWCV	11.79±0.41	10.56±0.54	0.08
RDWSD	16.61±2.42	13.16±2.13	0.31
PLT (10e3/uL)	1897.8±761.8	2313.0±1357.1	0.78
MPV (fL)	10.13±1.40	9.80±2.04	0.89
PCT	2.70±1.13	3.06±2.04	0.87
PDW	12.63±0.97	15.82±2.36	0.16

Values in bold show significance

(iii) *Haematological parameters according to age of goats*

Haematological parameters in goats of different ages are shown in Table 4.16. There were no outliers in the data, as assessed by inspection of a boxplot. Scores for each level of age for all parameters were normally distributed, as assessed by Shapiro-Wilks test ($p > 0.05$).

The Levene's Test indicated error variance to be equal across the groups for all parameters (all $p > 0.05$), except MCH ($F=3.35$, $p =0.04$) and MCHC ($F=4.94$, $p =0.01$). Consequently, post-hoc analysis was conducted with Tukey's Honest Significant Difference (HSD) test when the assumption of equality of variance was tenable and with Games-Howell test when it was violated. Analysis of variance showed no significant differences (all $p>0.05$) in parameters between the different age groups for all the parameters under investigation.

Table 0:16: Haematological factors according to goat's age

Parameter (n=76)	Yearling (n=49)	Weaner (n=9)	Adult (n=18)
	Mean±SEM	Mean ±SEM	Mean ±SEM
WBC (10e3/ul)	22.17±1.93 ^a	22.21±5.43 ^a	21.42± 2.25 ^a
Lymp (%)	64.82±3.28 ^a	76.32±4.00 ^a	62.56±5.61 ^a
Monocytes (%)	2.34±0.23 ^a	7.94±2.72 ^a	2.91±0.51 ^a
Gran (%)	3.50±0.72 ^a	2.48±0.78 ^a	4.26±1.36 ^a
RBC (10e3/ul)	1.99±0.20 ^a	1.36±0.15 ^a	1.59±0.29 ^a
HGB (g/dL)	7.84±0.26 ^a	8.80±0.33 ^a	8.00±0.36 ^a
HCT (%)	7.24±0.92 ^a	4.30±0.50 ^a	5.74±1.39 ^a
MCV (fl)	33.88±0.62 ^a	31.57±0.36 ^a	32.86±1.10 ^a
MCH (pg)	62.35±7.4 ^a	70.61±8.33 ^a	88.54±13.9 ^a
MCHC	194.1±26.1 ^a	226.5±29.2 ^a	272.6±53.2 ^a
RDWCV	11.87±0.42 ^a	9.85±0.31 ^a	10.72±0.69 ^a
RDWSD	16.20±2.30 ^a	11.05±1.35 ^a	14.02±2.82 ^a
PLT (10e3/uL)	2166.3±750 ^a	-	1555.3±1460 ^a
MPV (fL)	10.03±1.21 ^a	-	10.03±3.15 ^a
PCT	2.92±1.09 ^a	-	2.47±2.41 ^a

PDW	13.59±1.24 ^a	-	13.70±2.05 ^a
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Key. Means with similar letters in a row are not significantly different Tukey's HSD or Games Howell tests

(iv) *Goat haematological parameters according to location*

Table 4.17 presents haematological parameters in goats shown according to location. There were no outliers in the data, as assessed by inspection of a boxplot. Scores for each level of age for all parameters were normally distributed, as assessed by Shapiro-Wilks test ($p > 0.05$). The Levene's Test indicated error variance to be unequal across the groups for all parameters (all $p < 0.05$), except WBC ($F=3.85$, $p=0.053$). Consequently, except for WBC, p values for t values with adjusted degrees of freedom were reported for all parameters.

Independent samples T-tests were conducted to determine whether the haematological parameters differed significantly in Keiyo and Marakwet goats. The tests indicated that four factors, lymphocytes %: $t(17.04) = 3.03$, $p=0.008$; PLT: $t(14.93) = 7.44$, $p=0.001$; MPV: $t(73.97) = 7.47$, $p<0.0001$; and PCT: $t(28.58) = 5.98$, $p<0.002$) were significantly higher in goats located in Keiyo compared to those in Marakwet. However, granulocyte %: $t(14.94) = -2.96$, $p = 0.01$; HGB: $t(73.61) = -5.44$, $p<0.0001$; MCH: $t(28.70) = -5.09$, $p<0.0001$) and MCHC: $t(28.58) = -4.95$, $p<0.0001$) were found to be significantly higher in goats in Marakwet relative to those in Keiyo.

Table 0:17: Haematological factors according to location of goats

<i>Parameter (n=96)</i>	<i>Keiyo (n=48)</i>	<i>Marakwet (n=28)</i>	<i>p value</i>
	Mean±SEM	Mean ±SEM	
WBC (10e3/ul)	22.27±2.18	21.53±1.64	0.81
Lymph (%)	71.52±1.19	53.3±5.80	0.008
Monocytes (%)	2.42±0.19	3.074±0.62	0.34
Granu (%)	2.18±0.26	6.58±1.46	0.01
RBC (10e3/ul)	1.80±0.08	1.86±0.38	0.87
HGB (g/dL)	7.35±0.25	9.07±0.19	P<.0001
HCT (%)	5.95±0.29	7.54±1.80	0.39
MCV (fl)	32.85±0.22	34.24±1.26	0.29
MCH (pg)	45.74±2.20	110.32±12.47	P<.0001
MCHC	132.35±7.76	360.85±45.49	P<.0001
RDWCV	10.94±0.21	12.09±0.79	0.17
PLT (10e3/uL)	4249.8±553.9	119.06±29.60	0.001
MPV (fL)	13.88±0.94	6.73±0.20	P<.0001
PCT	6.02±0.99	0.07±0.01	.002
PDW	11.68±2.00	15.27±0.21	0.13

However, the rest of the parameters did not differ (at $p < 0.05$) significantly between cattle in the two locations.

The mean values of WBC, LMP, MCH, MCHC, PLT and granulocytes were higher in helminth-infected goats than non- infected but HGB were lower.

4.8.3 Haematological parameters of sheep and factors affecting their values

The results of the haematological values obtained from sheep are here represented.

(ii) Overall haematological parameters in sheep

Table 4.18 presents the overall haematological parameters for sheep in the study, their standard error of means (SEM), and 95% confidence intervals for the means. The table also shows the parameters segregated according to whether the animal was infected by any parasite (helminth or protozoa) or non-infected, and the standard reference values for the parameters. There were no outliers in the data, as assessed by inspection of a boxplot. Parameter scores were normally distributed, as assessed by Shapiro-Wilks test (all had $p > 0.05$). In addition, the values for skeweness and kurtosis for all the factors were within the benchmark ± 2.0 (Field, 2005).

Levene's Tests indicated error variances to be equal across the groups of infected and non-infected sheep (all $p > 0.05$) except for RBC ($F=10.23$, $p=0.002$) and PLT ($F=5.86$, $p=0.018$). Parameter levels in infected and non-infected sheep were compared using independent samples T -tests while one - sample T -tests were used to analyse differences between parameter levels (for infected and non-infected sheep in each case) and reference values. The T -values with adjusted degrees of freedom were reported in the independent samples tests for RBC and PLT.

Independent sample T -tests showed that ten parameters were significantly different in infected and non-infected sheep (at $p < 0.05$). Six factors; WBC, $t(80) = -3.32$, $p=.001$; lymphocytes, $t(80) = -3.09$, $p=.003$; MCH, $t(80) = -2.23$, $p=.029$; MCHC, $t(80) = -2.42$, $p=.018$; PLT, $t(66.89) = -3.35$, $p=.001$; and MPV, $t(80) = -2.12$, $p=.038$ were significantly higher in infected sheep relative to the non-infected animals. On the other hand,

monocyte, $t(80) = 2.39$, $p = .021$; granulocyte, $t(80) = 3.04$, $p = .004$; RBC, $t(47.55) = 2.41$, $p = .018$; and RDW, $t(80) = 2.92$, $p = .005$ were significantly greater in non-infected sheep compared with infected ones.

One-sample *T*-tests indicated differences in haematological parameters both in infected or non-infected sheep compared with standard reference values. White blood cell count (WBC), MCH, and MCHC were significantly higher in sheep of the present study compared to the normal standard blood values. Lymphocytes, PLT, and MPV were significantly higher in infected sheep relative to reference values but these parameters were not significantly different between non-infected sheep and the reference values. Monocytes were significantly higher in non-infected sheep compared with either infected or standard values.

Table 0:18: Overall haematological parameters in sheep

<i>Parameter (n=84)</i>	<i>Overall</i>	<i>Infected</i>	<i>Non-infected</i>	<i>Reference range</i>
	Mean±SEM	Mean ± SEM	Mean ± SEM	
WBC (10e3/ul)	69.56±5.71	83.15±6.62 ^a	46.01±9.24 ^b	4-8 ^c
Lymp (%)	72.07±3.83	80.76±4.20 ^a	58.34±6.24 ^b	40-55 ^b
Monocytes (%)	14.65±2.10	10.81±2.47 ^a	20.70±3.40 ^b	0-6 ^a
Gran (%)	7.56±0.89	5.54±0.93 ^a	10.73±1.55 ^b	
RBC (10e3/ul)	7.57±0.35	6.93±0.38 ^a	8.68±0.68 ^b	9-15 ^b
HGB (g/dL)	10.45±0.23	10.19±0.24 ^a	10.91±0.47 ^a	9-15 ^a
HCT (%)	26.91±1.49	25.08±1.90 ^a	30.07±2.33 ^a	
MCV (fl)	34.34±0.21	34.09±0.21 ^a	34.77±0.43 ^a	28-40 ^a
MCH (pg)	15.053±0.42	15.75±0.52 ^a	13.82±0.71 ^b	8-12 ^c
MCHC	44.08±1.33	46.47±1.63 ^a	39.93±2.15 ^b	31-34 ^c
RDW	11.80±0.33	11.16±0.26 ^a	12.92±0.65 ^b	

PLT (10e3/uL)	1600.1 \pm 209.	2101.8 \pm 302.9 ^a	873.6 \pm 207.2 ^b	800-1100 ^b
MPV (fL)	12.74 \pm 1.05	14.55 \pm 1.64 ^a	10.13 \pm 0.74 ^b	

Key. Means with similar letters in a row are not significantly different by T tests

(iii) *Haematological parameters according to sex of sheep*

Table 4.19 presents haematological parameters in sheep according to sex.

Table 0:19: Haematological factors according to sheep sex

<i>Parameter (n=82)</i>	<i>Female (n=56)</i>	<i>Male (n=26)</i>	p value
	Mean±SEM	Mean ± SEM	
WBC (10e3/ul)	74.41±7.06	59.11±9.55	0.22
Lymphocytes (%)	74.79±4.39	65.24±7.65	0.26
Monocytes (%)	13.28±2.36	18.12±4.40	0.30
Granulocytes (%)	7.08±1.10	8.75±1.53	0.41
RBC (10e3/ul)	7.66±0.44	7.37±0.62	0.71
HGB (g/dL)	10.58±0.28	10.18±0.37	0.42
HCT (%)	27.65±1.95	25.30±2.12	0.46
MCV (fl)	34.35±0.25	34.33±0.35	0.97
MCH (pg)	14.98±0.49	15.20±0.85	0.81
MCHC	43.85±1.52	44.57±2.69	0.80
RDW	11.87±0.41	11.67±0.33	0.76
PLT (10e3/uL)	1771.8±281.9	1218.0±242.3	0.14
MPV (fL)	13.41±1.45	11.24±0.92	0.34

Scores for each level of sex for all parameters were normally distributed, as assessed by Shapiro-Wilks test ($p > 0.05$). The Levene's Test indicated error variance to be equal across the groups for all parameters (all $p > 0.05$), except PLT ($F=5.56$, $p = 0.02$). Independent sample T -tests were conducted to determine whether the haematological parameters differed significantly in male and female sheep and were all found not to be significant (at $p < 0.05$).

(iv) *Haematological parameters according to the age of sheep*

Haematological parameters in sheep of different ages are shown in Table 4.20. There were no outliers in the data, as assessed by inspection of a boxplot. Scores for each level of age for all parameters were normally distributed, as assessed by Shapiro-Wilks test ($p > 0.05$).

Table 0:20: Haematological factors according to the age of sheep

Parameter	Yearling ($n=49$)	Weaner ($n=9$)	Adult ($n=18$)
	($n=76$)		
	Mean \pm SEM	Mean \pm SEM	Mean \pm SEM
WBC (10e3/ul)	33.40 \pm 9.60 ^a	71.76 \pm 19.43 ^b	82.62 \pm 6.62 ^b
Lymp (%)	54.25 \pm 6.65 ^a	48.80 \pm 18.98 ^a	85.10 \pm 3.61 ^b
Monocytes (%)	23.90 \pm 3.90 ^a	26.43 \pm 11.25 ^a	7.92 \pm 1..51 ^b
Gran (%)	11.41 \pm 1.70 ^a	11.19 \pm 2.74 ^a	4.93 \pm 0.87 ^b
RBC (10e3/ul)	9.81 \pm 0.83 ^a	8.06 \pm 1.16 ^a	6.67 \pm 0.35 ^b
HGB (g/dL)	11.63 \pm 0.26 ^a	10.38 \pm 0.63 ^{ab}	10.02 \pm 0.26 ^b
HCT (%)	37.69 \pm 4.01 ^a	27.70 \pm 3.97 ^{ab}	22.79 \pm 1.29 ^b
MCV (fl)	35.10 \pm 0.62 ^a	34.41 \pm 0.42 ^a	34.05 \pm 1.10 ^a
MCH (pg)	13.15 \pm 0.93 ^a	13.80 \pm 0.99 ^{ab}	15.93 \pm 0.49 ^b
MCHC	37.57 \pm 2.77 ^a	40.26 \pm 3.11 ^{ab}	47.05 \pm 1.56 ^b
RDW	13.00 \pm 0.32 ^a	11.54 \pm 0.51 ^a	11.40 \pm 0.49 ^a
PLT (10e3/uL)	300.06 \pm 138 ^a	2127.29 \pm 664 ^b	2028.3 \pm 272.4 ^b
MPV (fL)	8.11 \pm 0.52 ^a	12.68 \pm 1.59 ^{ab}	14.56 \pm 1.51 ^b

Key. Means with similar letters in a row are not significantly different Tukey's HSD test

The Levene's Test indicated error variance to be equal across the groups for all parameters (all $p > 0.05$), except MCH ($F=3.35, p=0.04$) and MCHC ($F=4.94, p=0.01$). Consequently, post-hoc analysis was conducted with Tukey's Honestly Significant Difference (HSD) test when the assumption of equality of variance was tenable and with Games-Howell test when it was violated.

Analysis of variance showed that WBC and PLT were significantly lower in yearlings relative to both weaner and adults whereas lymphocytes, MCH, and MCHC were lower in both yearling and weaner compared to adults. Monocytes, RBC, granulocytes, HGB, and HCT were generally greater in both yearling and weaner compared to adults. On the other hand, there were no significant differences in the levels of MCV and RDW in sheep of all age groups.

(v) Sheep haematological parameters according to location

Table 4.21 presents haematological parameters in sheep according to location. There were no outliers in the data, as assessed by inspection of a boxplot. Scores for each level of location for all parameters were normally distributed, as assessed by Shapiro-Wilks test ($p > 0.05$). The Levene's Test indicated error variance to be unequal across the groups for the parameters WBC, lymphocytes, HGB, RBC, MCV, RDW, PLT, and MPV and therefore, t values with adjusted degrees of freedom were reported for these factors.

Table 0:21: Haematological factors according to location of sheep

<i>Parameter (n=96)</i>	<i>Keiyo (n=15)</i>	<i>Marakwet (n=67)</i>	<i>p value</i>
	Mean±SEM	Mean ±SEM	
WBC (10e3/ul)	40.93±6.11	75.97±6.62	P<.0001
Lymp (%)	75.08±5.30	71.72±4.23	0.63
Monocytes (%)	11.42±2.93	15.01±2.32	0.61
Gran (%)	13.50±2.61	6.88±0.91	0.02
RBC (10e3/ul)	6.15±0.22	7.89±0.43	0.001
HGB (g/dL)	10.77±0.25	10.38±0.27	.31
HCT (%)	26.51±4.90	26.99±1.48	0.90
MCV (fl)	35.41±0.79	34.10±0.17	0.13
MCH (pg)	17.75±0.72	14.44±0.47	.002
MCHC	51.01±2.82	42.52±1.45	0.013
RDW	14.23±1.22	11.26±0.20	0.03
PLT (10e3/uL)	150.33±26.94	1988.52±240.8	P<.0001
MPV (fL)	11.53±4.65	13.07±0.53	0.75

The values in bold are were significantly different between Marakwet and Keiyo sub counties

Independent samples t tests were conducted to determine whether the haematological parameters differed significantly in Keiyo and Marakwet sheep. The tests indicated that granulocytes %: $t(80) = 2.32, p=0.02$; MCH: $t(80) = 3.14, p=0.002$; MCHC: $t(80) = 2.53, p = 0.013$; and RDW: $t(14.79) = 2.38, p=0.03$ were significantly higher in sheep located in Keiyo compared to those in Marakwet. However, WBC: $t(51.26) = -3.44, p < 0.001$; RBC: $t(78.70) = -3.59, p=0.001$; and PLT: $t(56.35) = -7.59, p < 0.0001$ were

found to be significantly higher in sheep in Marakwet relative to those in Keiyo. However, the rest of the parameters did not differ (at $p < 0.05$) significantly between sheep in the two locations.

4.8.4 Relationship between parasite categories and haematological values

Intestinal parasites were grouped into categories based on their classes i.e. coccidia, nematodes, trematodes and cestodes.

Table 0:22: Effect of nematode infection on haematological parameters of sheep

Parameter (n=82)	Not infected (n=34)	Infected (n=48)	p value
	Mean±SEM	Mean ± SEM	
WBC (10e3/ul)	51.73±8.83	82.19±7.02	0.008
Lymp (%)	60.14±6.18	80.29±4.33	0.008
Monocytes (%)	19.93±3.34	11,01±2.54	0.036
Gran (%)	9.88±1.49	5.96±1.04	0.031
RBC (10e3/ul)	8.46±0.61	6.94±0.41	0.17
HGB (g/dL)	10.83±0.42	10.18±0.25	0.04
HCT (%)	29.42±2.10	25.13±2.05	0.000
MCV (fl)	34.85±0.39	34.00±0.20	0.16
MCH (pg)	14.04±0.72	15.76±0.51	0.04
MCHC	40.54±2.18	46.598±1.61	0.03
PLT (10e3/uL)	912.1±196.6	2164.8±321.17	0.001
MPV (fL)	10.41±0.72	14.66±1.77	0.04

On determination of relationship between different categories of parasitic infections and blood values, it was found that nematodes (Table 4:22) and cestodes infections significantly affected PLT and MPV of goats. The tests found PLT ($p=0.02$, $p=0.1$) and MPV ($P=0.1$, $P=0.1$) for nematode and cestode infections respectively. Infection with nematodes in sheep elevated WBC, lymphocytes, MCH, MCHC, PLT and MPV while Monocytes, Granulocytes, RBC, HGB, HCT were lowered ($p<0.005$). However, only a few were affected in goats (PLT ($p=0.02$) and MPV ($p=0.03$) and in cattle only monocytes ($p=0.008$) were lower in infected animals.

Table 0:23: Effect of trematode infection on haematological parameters of cattle

Parameter (n=216)	Not infected (n=147)	Infected (n=69)	014(2)
	Mean±SEM	Mean ± SEM	p value
WBC (10e3/ul)	13.75±0.91	18.05±1.57	0.01
Lymp (%)	53.10±1.94	59.95±2.37	0.04
Monocytes (%)	11.89±1.87	4.59±0.76	0.000
Gran (%)	16.62±3.42	7.43±1.46	0.02
RBC (10e3/ul)	6.16±0.11	5.97±0.11	0.25
HGB (g/dL)	9.785±0.15	9.48±0.16	0.13
HCT (%)	28.35±0.80	25.80±0.50	0.04
MCV (fl)	44.79±0.40	43.54±0.56	0.08
MCH (pg)	16.90±0.53	15.99±0.17	0.24
MCHC	37.16±0.87	37.07±0.39	0.95
PLT (10e3/uL)	255.3±19.3	227.3±32.23	0.43
MPV (fL)	8.58±0.20	9.82±1.04	0.24

Trematode infection on the other hand only lowered monocytes ($p=0.02$) in sheep but in cattle WBC and lymphocytes were higher and monocytes, granulocytes, HCT were lowered in infected cattle than those not infected (Tab 4:23). Trematode infection in goats, showed no significant difference ($p<0.05$) in haematological parameters of infected and non-infected goats

Intestinal protozoan infection did not make any difference in infected or non-infected cattle and goats but had significant effects on some of blood values in sheep as shown in the Table 4:24 below;

Table 0:24: Haematological values in sheep infected or not infected with protozoa

Parameter	Not infected (n=56)	Infected (n=26)	
(n = 82)	Mean\pmSEM	Mean\pmSEM	p value
WBC (10e3/ul)	59.05 \pm 7.03	92.20 \pm 8.34	0.004
Lymp (%)	66.16 \pm 4.63	90.27 \pm 2.50	0
Monocytes (%)	17.48 \pm 2.58	5.91 \pm 1.60	0
Granu (%)	8.95 \pm 1.09	3.26 \pm 0.44	0.005
RBC (10e3/ul)	8.29 \pm 0.48	6.02 \pm 0.26	0
HGB (g/dL)	10.92 \pm 0.30	9.44 \pm 0.26	0.002
HCT (%)	29.92 \pm 2.02	20.43 \pm 0.98	0
MCV (fl)	34.60 \pm 0.28	33.80 \pm 0.21	0.07
MCH (pg)	14.49 \pm 0.51	16.23 \pm 0.74	0.06
MCHC	42.10 \pm 1.59	48.33 \pm 2.32	0.03
PLT (10e3/uL)	1229.1 \pm 238	2615.6 \pm 350	0.003
MPV (fL)	11.84 \pm 1.41	15.23 \pm 0.42	0.024

Significant values are shown in bold.

CHAPTER FIVE

DISCUSSION

5.1 Parasite Prevalence

Indigenous livestock which are the local breeds of animals kept under communal and free range in Kerio valley, Kenya are infected with a variety of helminth and protozoan parasites found in both blood and gastrointestinal tract. The occurrence of parasites in livestock was found in co-infective states in majority of the animals than a single infection. This was possibly due to a favourable climate for survival of parasites, vectors, transmission of parasites and cross contamination of environment from animal faeces and availability of hosts that maintain arthropod vectors. Climate change is also a threat to livestock production because of its impact among others water availability, meat and milk production and livestock diseases (Rojas-Downing, et al., 2017). This change could have favoured survival of disease agents or affected the susceptibility of livestock to diseases.

Despite the presence of gastrointestinal or blood parasites in livestock kept in Kerio Valley, there were no obvious observable clinical symptoms, suggesting that most parasitic infection in indigenous of Kerio valley are generally subclinical and not life threatening. However, non-clinical symptoms such as reduced meat and milk production, and diarrhoea, have been established by previous studies (Fitzpatrick, 2013), and the presence of infections in herd might also hinder growth especially in young animals (Huang, 2014), which cannot be ruled out in this animal population. Although the animals were apparently healthy, the asymptomatic state consequently

signified the contribution of the affected livestock role in transmission of infections to susceptible animals and maintenance of disease agents in the environment.

5.1.1 Gastrointestinal Parasites

The most common gastrointestinal helminths that affected livestock in the study area were nematode infections belonging to the family Trichostrongyloidea. The nematodes that affected the animals mostly were *Trichostrongylous*, *Strongyloides*, *Ascaris*, *Bunostomum*, *Enterobius* and *Avitellina species*. Among the less common nematode infections included *Haemonchus*, *Toxoscaris*, *Ostergia*, *Acutellina*, *Cooperia*, *Oesophagystumum* species among others (Table 4.4). This agrees with previous studies that *Trichostrongyles* are the most common nematodes diagnosed on faecal flotation of ruminants (DFID-KARI, 1999; Odoi, *et al.*, 2007). Of the *Trichostrongyles* detected, *Strongyloides* were the most abundant parasite occurring in sheep, goats and cattle with 33%, 42% and 52% prevalence respectively. The prevalence of *Haemonchus contortus* in the study area was conspicuously low; this is in contrast with the notion that it is a common nematode infection of livestock under nomadic management of arid areas of Kenya (Waruiru *et al.*, 2000; Nganga, *et al.*, 2004).

The presence of dog nematode infection like *D. caninum* and *Toxoscaris* in ruminants indicated cross contamination of pastures by dog faeces or due to close association between communities living in Kerio valley, their animals and dogs that predispose livestock to canine parasites.

The overall prevalence of gastrointestinal helminths in cattle of 59.9% was recorded (Fig. 4.1), which corroborates with other findings from previous work (Odoi *et al.*, 2007; Tulu & Lelisa, 2016). Cattle infected with gastrointestinal helminths were likely to be adults than the young animals and more prevalent in Keiyo sub county, this is in

consistent with previous studies that found out that risk factors to helminth infection include host factors such as age, genetic resistance and environmental conditions i.e. climate, nutrition and management practices (Odoi *et al.*, 2007).

The farmers in Keiyo Sub County keep more of mixed animal breeds and they are kept in confinement as compared to those in Marakwet Sub County explaining the high rate of infections. Additionally, it also showed that cattle in Marakwet Sub County are more tolerant to helminth infections and/or they are capable of expelling the worms before establishment due to either genetic factors and acquired resistance as reported in neighbouring Uganda (Magona, *et al.*, 2011).

In small ruminants, the study showed that intestinal helminths were the major parasitic infections with the intestinal nematode infections being 58.3% and 74.5% in sheep and goats respectively. Intestinal helminth and protozoan infections were likely to infect sheep and goats in Marakwet than in Keiyo Sub County. While adults were more infected in sheep, the goats' age showed no bias to helminth infection whether they were adults or young, male or female, which agrees with findings documented in a previous study (Bogale *et al.*, 2014). Female sheep were shown to be more infected than the bucks, this is in sharp contrast to previous work which found males of small ruminants to be more infected with intestinal parasites than female (Magaji *et al.*, 2014).

Trematode species observed were *Fasciola*, *Schistosoma* and *Dicrocoelium* species. *Dicrocoelium* was only observed in cattle (9.5%). The presence of trematode infection has previously been associated with regions with high rainfall and poor drained soils, which favour survival of snail intermediate hosts (Jacobs, *et al.*, 2016). In the current study area, it can be postulated that the occurrence of trematode infections with snail intermediate hosts could be related to presence and proximity of Kerio River and its

tributaries that provide constant wetlands and flooding during rainy seasons provide important habitats for *Lymnea* snails.

Irrigation furrows that have existed for many years (MoA, 2005) in the valley also provide suitable habitat for fresh water snails. Fascioliasis was common in all types of livestock and has as such, have a potential of causing economic losses, depression of growth rates and liver condemnation at slaughter as earlier reported (Egbu, 2013). The occurrence of bovine schistosomosis in cattle (21.8%) is similar to what was found by Yabe *et al.* (2008).

Intestinal coccidia were also reported in the current study although at lower rates compared with intestinal amoebas and flagellates. Sheep and goats were more infected with *Balantidium* than cattle though all livestock types were similarly infected with intestinal amoebas. Goats were found to have higher prevalence of coccidian infections than sheep and cattle with 48%, 32% and 39% prevalence respectively. This corroborates with previous studies that found goats were more likely to be infected with coccidian species than sheep (Mohamaden *et al.*, 2018). However, no clinical symptoms and signs were observed in these livestock. Along the upper Kerio Valley, the location of livestock determined infection rates of protozoan infections with sheep and goats located in Marakwet Sub County being more infected than those in Keiyo Sub County but opposite for cattle occurred. This could basically be due to genetics and type of management systems.

The presence of cestode eggs (*Taenia* species and *Hymenolepis diminuta*) in faecal samples of goats and cattle was surprising since ruminants are normally intermediate hosts of most tapeworms. The effect of climate change on parasite biology, adaptability, and host range may cause shift in disease distribution as suggested by Moore, Shrestha,

Tomlinson, & Vuong (2012). Since the *Hymenolepis diminuta*'s eggs were found in goats and cattle, having its natural host to be rats (Gunn & Pitt, 2012), it only meant that forage and animal feed/forage were contaminated with infected rodent faeces. The presence of *Taenid* eggs could be due to poor sanitary conditions and high temperatures, which favour zoonotic infection (Hedrix & Robinson, 2006).

Coinfections by all possible combinations occurred in 28% - 53.5% of livestock. Double infections were more compared with those with triple or quadruple infections. Many factors may have contributed to this scenario including exposure and establishment of parasite infections and immune mechanisms towards different parasitic organisms.

5.1.2 Haemoparasites

Generally, vector-borne haemoparasites were less evident in livestock using microscopy (Table 4.6). This is probably due to limitations in terms of specificity and sensitivity of microscopy and its practicability due to large number of samples (Eisler *et al.*, 2004). It could also be due to use of prophylactic drugs and interventions through the use of acaricides by the livestock owners. In addition, some of haemoparasite infections result in a state of premunition immunity, which may cause the host to become long-term asymptomatic carrier or resistant to new infections all together as it has been reported by Bell-Sakyi *et al.*, (2004). Although the study showed low prevalence of haemoparasites, their effect however are known to manifest in production losses such as low milk and meat productivity, depressed growth and increase in susceptibility to other diseases (Isaac, *et al.*, 2016).

Tick-borne parasites in livestock of Kerio valley were generally low with the prevalence of *Theileria parva* as common blood parasite at 4%. As stated earlier, this could be attributed to natural resistance phenomenon of these indigenous breeds of livestock that

operate at different levels (Gunn & Pitt, 2012). The animals kept by farmers are basically indigenous animals, which are out of their genetic make-up, are naturally resistant to pathogenic parasites and due to their geographical distribution, nutritional habitats and behavioural characteristics, the animals may not have come into contact with infected vectors or as a result of continuous inoculation of blood parasites by the vectors, have developed immunity that prevent clinical manifestation or high parasitaemia which is supported by previous studies (Wakelin, 1996; Gasbarre, *et al.*, 2001).

There were no cases of haemoparasites parasites among the young animals, possibly due to passive immune protection and unlikelihood of coming into contact with vectors, which agree with previous work done by Kamani *et al.*, (2010). It has also been established that when young animals become continuously challenged by parasite infected ticks especially when they are still protected by colostrums' immunity or passive immunity, it will result in high level of immunity in adulthood with consequent low incidence of clinical disease in few cases of infections (Jonsson *et al.*, 2012 and Gachohi *et al.*, 2013).

Molecular diagnosis however, demonstrated that livestock in the study area harbour infections with trypanosomes consisting of *T congolense* and *T brucei* which seem to be main parasites responsible for African animal trypanosomosis in Kerio Valley. The results showed that *T. brucei* accounted for significant proportion (19%) of the trypanosome infection in livestock in Kerio Valley. The identification of trypanosome in sheep also confirmed that even small ruminants are also readily fed by tsetse flies and are susceptible to trypanosome infection just like bigger ruminants, this agrees with the work done by Masiga *et al.* (2002), which showed that small ruminants to be susceptible to trypanosomes infection. Sheep were only found to be infected by *T. brucei*, probably

due to chance or possibly depended on complex epidemiological interactions that vary from one environment to another as it had been shown by Masiga *et al.*, (2002).

The use of PCR identification of trypanosomes has provided an understanding about the prevalence and types of trypanosome infections circulating in livestock and its vectors in the study area. The difference between prevalence of trypanosomes by microscopy and PCR procedure is undoubtedly due to high sensitivity and accuracy of molecular method. The frequency of trypanosome infections in small number of sampled animals was unexpected since Kerio valley is among the areas in which PATTEC-CK project of vector and disease control that had been running since 2005 (MoLD, 2011), and also due to the fact that most of the animals kept by farmers here are indigenous zebu which are genetically trypanotolerant (Rege *et al.*, 2001). Although *T. congolense* infections were identified, determination of the *T. congolense* types and strains in the study could not be proved due to costs, and unavailability of primers specific for their isolation.

5.1.3 Ectoparasites

Four different species of ticks were found on cattle in Kerio Valley, the most common ones being brown-ear ticks (*Rhipicephalus appendiculatus*), blue ticks (*Boophilus* species), black ticks (*Hyalomma* species) and *Ambyloma variegatum*; this is in consistence with previous studies in other parts of Kenya and neighbouring Uganda (Masiga *et al.*, 2002, Sang *et al.*, 2006, Magona *et al.*, 2011). Ticks were common in western Kerio valley especially Marakwet sub-County where livestock grazing are more of communal, which involves extensive vast movement of animals exposing animals to tick infested areas than those observed in Keiyo Sub County. The proximity to wildlife could have contributed to this phenomenon which agrees with previous studies that also

found that pastoralist communities that exist near wildlife parks allows the transfer of ticks and possibly pathogens to livestock (Sang et al., 2006).

The identification of tsetse flies, confirmed that *Trypanosoma* species found in livestock in this region are transmitted by tsetse flies. This also signifies the role of tsetse vector in the sustenance of animal trypanosomosis problem in the study area. According to Ministry of Livestock and Development (2011), Kerio Valley lies within the tsetse belts that form part of Lake Bogoria-Baringo-Kerio Valley-Koibatek-Turkwel fly belt where *Glossina pallidipes* and *G. morsitans* are likely to occur.

Scabies mites were also recorded which generally all are ectoparasites but the female scabies mites cause more harm as they burrow into the skin of the animal causing the damage of the skin (Gunn and Pitt, 2012). The two mite species i.e. *Chorioptes bovis* and *Psoroptes mangle* both identified in cattle may be due to poor management practices and/or lack of knowledge about mange mites by cattle owners as suggested previously (Beyechea, et al., 2014). The presence of mites was localized indicating unhygienic conditions and poor management at farm level than the entire study area.

5.2 Factors that affect Haematological Parameters of Livestock

The study confirmed relationships between the parasitic infections, age group and alterations of haematological indices of livestock in this study, which agrees with previous research (Van Wyk, et al., 2013) that among the parasites that affect blood profiles include *Anaplasma*, *Babesia* and trypanosomes infections. The study also demonstrated that haematological profiles of livestock in the study differed in different age groups and sex of the animal as compared to reference values as reported in the Mercks' Manual (2012) and Ministry of Livestock Manual).

5.2.1 Haematological parameters in relation to Standard reference values

Although the results (Table 4.11) showed the haematological profiles from apparently healthy cattle varied from the standard references blood values (Appendix 4, Merck's Manual, 2012) they did not differ significantly except for platelet counts (PLT) and white blood cell (WBC) counts. Significant high WBC counts observed, than standard reference values indicated that the cattle kept in Kerio valley are capable of generating antibodies in the process of phagocytosis and have high degree of resistance to diseases as it has been found in previous studies (Soetan *et al.*, 2013) thus making them adaptable to prevailing harsh environmental conditions and provide rapid and effective defence to infectious pathogens (Kubkomawa *et al.*, 2015).

Most of the blood value ranges of goats in Kerio Valley varied from standard reference values including WBC, MCV MCH and MCHC which were significantly higher ($p < 0.05$) and RBC, PLT and PCT being lower than reference values (Merck's Manual, 2012) which were also different from those found in other goats in previous (Tambuwal *et al.*, 2002; Opara *et al.*, 2010). This could be attributable to stress, hydration, nutritional composition and adaptations to hot, dry environments (Onasanya *et al.*, 2015) such as those found in Kerio Valley and also may due type of goat breeds that are kept in Kerio Valley. The higher values of WBC observed in this group of goats as in cattle reveal a good immune system, which provides a rapid and effective defence against infectious agents.

The overall haematological parameters observed in sheep were within the normal ranges except WBC counts which were higher ($p < 0.05$) than the standard reference values (Merck's Manual, 2012), just like cattle and goats it showed an effective protective immune system with rapid and potent strength of host immunity against infections..

5.2.2 Effect of parasites on haematological values

Although the lymphocytes, monocytes and granulocytes (Tables 4:11, 4:5, 4:19) in study livestock did not differ significantly with the standard reference values, they significantly differed between parasite-infected and non-infected livestock invaded by either blood or gastrointestinal parasites.

Reduced RBC and PLT in the blood of infected goats meant that there was increased destruction of red blood cells due to parasitic infection. As a consequence, there is probability of reduction in the levels of oxygen that reach the tissues and CO₂ to the lungs as reported previously (Isaac *et al.*, 2013, Etim *et al.*, 2014) and low process of clot-formation in case of an injury respectively (Etim *et al.*, 2014). However, MCH, MCHC and HCT/PCV levels of goats were within normal range ($p < 0.05$), which implies that there are adequate haemoglobin levels in the blood (Aster, 2004) among the goats of Kerio Valley.

Infection with intestinal or blood parasites had significant effect on haematological profiles of sheep in Kerio Valley. There was significant reduction in RBC, RDW and higher values of WBC, lymphocytes and PLT in infected sheep and this can be attributed to disruption of erythrocytes by parasites especially tick borne and nematode infections (Etim *et al.*, 2014) and as expected, elevation of defence cells such as WBC and lymphocytes is in attempt of the immune system of sheep for self-defence against infections.

However, mean blood values compared favourably between infected and non-infected cattle. This peculiar observation could be attributed to compensatory responses induced by cattle kept in Kerio valley that restore haematological levels especially the red blood cell factors as it was reported on *trypanosoma congolense*-infected goats and also due

to endemicity of parasites and animals are equally and continuously being exposed to them (Ng'wena *et al.*, 2011, Etim *et al.*, 2014).

5.2.3 Influence of sex, age and location on haematological parameters

In cattle, sex was observed to have an effect on monocytes, HCT, and MCV with cows having higher values than the bulls, which contrasted with a similar study on indigenous chickens (Onasanya *et al.*, 2015). In goats, sex had also significant effect on monocytes and granulocytes in which male goats had higher values than females. This is consistent with what was found in West African Dwarf goats (Daramola *et al.*, 2005). However, Sex of sheep did not have significant effect on haematological parameters.

The age of cattle and sheep had significant difference on various blood parameters. For instance, as the age of cattle increased, there was significant increase in WBC, Lymphocytes, mean corpuscular volume (MCV), and mean corpuscular haemoglobin concentration (MCHC). This observation was in accordance with the findings of previous studies, which reported that majority of haematological parameters increase with advancing age of animals (Addass *et al.*, 2012 and Onasanya *et al.*, 2015). Moreover, high values for WBC, lymphocytes and MCHC in older cattle and sheep in the study area signified enhanced adaptability to local environmental and disease prevalent conditions that have developed with time (Kubkomawa *et al.*, 2015). The age of goats did not have any effect on haematological values.

On the other hand, it was found that location played a role in haematological parameters of livestock in the study. The monocytes, granulocytes count, MCV and PLT were significantly higher in cattle of Marakwet Sub Counties than those in Keiyo sub counties and only MCHC was higher in cattle reared in Keiyo region than those reared in Marakwet. This was consistent with the work done by Etim *et al.*, (2014) which reported

that environment and management systems affect blood values of animals. The effect of location in goats was evidently observed for blood values: PLT, MPV, MCH, HGB, MCHC and granulocytes.

5.2.4 Effect of specific parasitic groups on haematological parameters

In this study, infection with helminths or protozoa generally lowered monocytes, granulocytes, Red blood cell counts (RBC), haemoglobin (HGB) and packed cell volume (Table 4:23, 4:24). Reduction in RBC, HGB and HCT were expected in parasitic infection due to lysis of red blood cells and its ratio to blood volume. Similar findings were recorded by Esmailnejad *et al.*, (2012), on the contribution of parasitic infections to anaemia.

Nematodes, flukes and protozoan infections were highly associated with lowered HCT values and RBC, this could be attributed to blood-sucking by adult worms, and destruction of these blood cells by protozoan parasites and bleeding from epithelial layers through replication and development of parasitic protozoa (Gunn & Pitt, 2012).

The presence of outlier blood values in healthy animals may simply be as a result of management practices, nutritional provisions, genetics or environment in which the animals are kept and not been majorly due to parasitic infections for instance the responses of the livestock to prevailing environmental and management systems.

CHAPTER SIX

CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

This study has provided information on intestinal helminth and protozoan parasite prevalence in livestock breeds reared under free-range management in upper Kerio Valley region. Livestock parasitism is a major a health problem and economically important infections of cattle, sheep and goats in Kerio valley. Although all the parasites in the study were prevalent, it appeared intestinal nematodes were most prevalent in all groups of livestock in upper Kerio Valley compared to other intestinal parasites diagnosed.

The study has provided information of blood parasite status of livestock comprising of tick borne diseases and tsetse fly transmitted trypanosomes. It has also given evidence of ticks and tsetse flies identified in the field possibly harbour infective stages of haemoparasites and subsequently transmit parasites to livestock.

The study confirmed the presence of four tick vectors that are capable of transmitting tick-borne diseases. These include *Rhipicephalus appendiculatus*, *Hyaloma sp*, *Boophilus* and *Amblyomma gemma* each of which are capable animal pathogens as well as zoonoses. The occurrence of mange mites in localized site i.e. Tot, was possibly due to poor husbandry at farm level than a common problem in the study site.

The use of molecular method utilizing PCR has greatly improved our understanding on epidemiology of animal trypanosomosis in Kerio valley as compared to the use of microscopy alone. Therefore, PCR method in diagnosis and identification of trypanosomes and other blood protozoan infection is a valid tool for the detection of

trypanosomosis. The outcomes of the study signified the existence of African Animal Trypanosomosis (AAT) in the upper Kerio Valley that is caused by *Trypanosoma brucei* and *T. congolense* with similar implications in the entire Kerio valley where tsetse fly thrive. The presence of *T. congolense* in study site poses potential danger to public health because of its capability of being zoonotic.

Infection with parasites had significant effect ($p < 0.05$) on various haematological parameters. The major blood parameters affected were the red blood cells (RBC), haemoglobin concentration (HCB), and hematocrit (HCT/PCV). White blood cells and its components were elevated as expected due to host immune response to infection. Thus, the haematological values from uninfected livestock obtained can be used to serve as reference for comparison to other farm animals under similar natural conditions.

Age, sex and location of livestock affected haematological values. While location had significant effect ($p < 0.05$) on blood values, age affected blood values of cattle and sheep while age was a factor in haematological parameters of goats and cattle. Haematological values found in the study can be used in selection and breeding of livestock suitable under semi-arid and hot conditions such as those found in the upper Kerio valley, as well as monitoring therapeutic regimes and nutritional programs.

6.2 Recommendations

- The high prevalence and occurrence of various parasite species, which sometimes occurred in co-infective state call for effective control measures so as to reduce mortality, morbidity and expenditure in treating parasitic diseases. This will also help in reducing the numbers of infective parasites in the environment hence, animals at risk of infection will not be exposed to the level of infection, which lead to disease.

- Concerted efforts are required to control vector borne infections and helminthiasis in livestock which can contribute significantly to a multi-disciplinary approaches in the control of animal diseases in Kerio valley and neighbouring areas where the animals occasionally move. Preventive measures applicable in Kerio Valley can also be extended to other areas where similar livestock diseases are endemic
- The presence of animal trypanosomes in Kerio Valley requires formulation of effective control measures, routine screening and surveillance that work in tandem with tsetse fly control methods for sustainable eradication of African Animal Trypanosomosis.
- Whereas domestic animals can easily be assessed to determine the prevalence of parasitic infections and evaluate their impact on animal health, full control cannot be achieved without consideration of controlling wild infectious parasites that originate from wildlife; wild foxes, wild dogs, rodents and other wildlife that are found in Rimoi game reserve and those in grazing areas..
- The damage to hides and skin and general health of infected livestock by mites cannot be underestimated and therefore, livestock owners and animal health officers in Kerio valley should put into consideration, mite control as part of ectoparasite control routines.

Suggestions for Further Research

- There is need to obtain true estimate of the vector borne parasite challenge by determining vectors density and specific parasites in vectors to determine their transmitting potential.
- Further research is needed to assess the economic impact of vector borne and helminth infections of livestock in relation to human health and livelihoods.
- Since *Fasciola sp* has potential to infect humans, then its presence can be source of zoonotic infections due to possible contamination of drinking water and edible water plants which contains infective metacecariae. Therefore, it will be important to investigate fascioliasis infection among the local population of Kerio valley especially those who live in close proximity to the rivers.
- With wide variety of tick species encountered during this research, their potential as vectors for various livestock other pathogens such as bacterial and viral diseases should further be investigated.
- Since this study focussed on livestock, a study is required to account for all ruminants including pigs and camelids.

REFERENCES

- Ahmed, A. H., MacLeod, T. E., Hide, G., Welburn, S. C., & Picozzi, K. (2011). The best practice for preparation of samples from FTA® cards for diagnosis of blood borne infections using African trypanosomes as a model system. *Parasites & Vectors* , 4, 68.
- Adass, P.A., David, D.L., Edward, A., Zira, K.E. and Midau, A. (2012). Effect of Age, Sex and Management System on Some Haematological Parameters of Intensively and Semi-Intensively Kept Chicken in Mubi, Adamawa State, Nigeria. *Iranian Journal of Applied Animal Science*, 2, 277-282.
- Al-Bulushi S, Shawaf T, Al-Hasani A (2017). Some hematological and biochemical parameters of different goat breeds in Sultanate of Oman “A preliminary study”. *Veterinary World*;10, 461–6.
- Aster, J. C. (2004). *Anaemia of diminished erythropoiesis*. In V. Kumar, A. K. Abbas, N. Fausto, S. L. Robbins, & R. S. Cotran (Eds.), *Robbins and Cotran Pathologic Basis of Disease* (7th ed., pp.638-649). Saunders Co. Philadelphia.
- Bell-Sakyi, L. K. (2004). Incidence and prevalence of tick-borne heamoparasites in domestic ruminants in Ghana. *Veterinary Parasitology* , 124 (1-2), 25-42.
- Beyecha K, Kumsa B, Beyene D. (2014). Ectoparasites of goats in three agroecologies in central Oromia, Ethiopia. *Comp Clin Path.*, 23, 21–8.
- Bogale, B., Chanie, M., & Melaku, A. (2014). Occurrence , Intensity and Parasite Composition of Gastrointestinal Helminth Parasites in Walia Ibex (*Capra walie*) at Semien Mountains National Park , Natural World Heritage Site , Northern Ethiopia, *Acta Parasitologica Globalis*. 5 (1), 19-25

- Borst, P. & Fairlamb, A. H. (1998). Surface receptors & Transporters of *Trypanosoma brucei*. *Annual Reviews*, 52, 745 - 778
- Bowman, D. D. (2009). *George's Parasitology for Veterinarians* (9th Edition ed.). Louis, Missouri, India: Elsevier Inc.
- Badran, I., Abuamsha, R., Aref, R., Alqisi, W., Alumor, J., (2012). Prevalence and diversity of gastrointestinal parasites in small ruminants under two different rearing systems in Jenin district of Palestine. *An-Najah Univ. J. Res.* 26, 1–18.
- Bronsvoort, B. W. (2010). No Gold Standard Estimation of Sensitivity and Specificity of Two Molecular Diagnostic Protocols for *Trypanosoma brucei* spp. in Western Kenya. *PLoS One.* 5 (1), 1-8.
- Byaruhanga, C., Collins, N. E., Knobel, D., Kabasa, W., & Oosthuizen, M. C. (2016). Veterinary Parasitology : Regional Studies and Reports Endemic status of tick-borne infections and tick species diversity among transhumant zebu cattle in Karamoja Region, Uganda : Support for control approaches. *Veterinary Parasitology: Regional Studies and Reports*, 1–2(2015), 21–30.
- Connor, R. J. (1994). The Impact of Nagana. *Onderstepoort Journal of Veterinary Research* , 61, 379-383.
- County Government of Elgeyo-Marakwet (2019);
<http://www.elgeyomarakwet.go.ke/index.php/about-us>. Date accessed: Jan 2019
- Courtin, D. B. (2008). Host Genetics in African Trypanosomosis. *Infection, Genetics and Evolution*, 8, 229-238.

- Daramola, J.O., Adeloye, A.A., Fatoba, T.A. and Soladoye, A.O. (2005). Haematological and Biochemical Parameters of West African Dwarf Goats. *Livestock Research for Rural Development*, 17, 95
- Desquesnes M., McLaughlin G., Zoungrana A., Davila A.M. (2001). Detection and identification of *Trypanosoma* of African livestock through a single PCR based on internal transcribed spacer 1 of rDNA. *International Journal of Parasitology*, 31(5–6): 610-614.
- Dlugosz, E. & Wisniewski M. (2006). Molecular diagnostic of parasites using rRNA gene sequence. *Wiad Parazytol*, 52 (4), 263-269
- Egbu, F. M. (2013). Haematological changes due to bovine fascioliasis. *African Biotechnology*, 1829-1833.
- Eisler MC, Dwinger RH, Majiwa PAO, Picozzi K (2004). *Diagnosis and Epidemiology of African Animal Trypanosomiasis*. In *The Trypanosomiases* Edited by: Maudlin I, Holmes PH. Miles MA: CABI Publishing; 253-267.
- Esmailnejad, B., Tavassoli, M., & Asri-rezaei, S. (2012). Investigation of hematological and biochemical parameters in small ruminants naturally infected with *Babesia ovis*. *Veterinary Research Forum*, 3(September 2009), 31–36.
- Etim, N. N., Williams, M. E., Akpabio, U., & Offiong, E. E. A. (2014). Haematological Parameters and Factors Affecting Their Values. *Agricultural Sciences*, 2(1), 37–47.
- FAO. (1992). Proceedings of the FAO expert consultation on the genetic aspects of trypanotolerance held in Rome, Italy.

- Field, A. (2005). *Discovering statistics using SPSS*. 2ndEd. London: Sage Publications.
- Fitzpatrick, J. (2013). Global food security; The impact of veterinary parasites and parasitologists. *Veterinary Parasitology* , 195 (3-4), 233-248.
- Fox, M. (1993). Pathophysiology of *Ostertagia ostertagi* in cattle. *Veterinary Parasitology*, 46-143.
- Gachohi, J.M., Kitala, P.M., Ngumi, P.N., Skilton, R.A., Bett, B. (2013). Population attributable fractions of farm vector tick (*Rhipicephalus appendiculatus*) presence on *Theileria parva* infection seroprevalence under endemic instability. *Prev. Veterinary Medicine* 108, 103–113.
- Gasbarre, L. (1997). Effects of gastrointestinal nematode infection on ruminant immune system. *Veterinary Parasitology* , 72, 327-343.
- Gasbarre, L.C., Leighton, E.A., Sonstegard, T. (2001). Role of the bovine immune system and genome to gastrointestinal nematodes. *Veterinary Parasitology* 98 (1–3), 51–64.
- Geurden, T., Olson, M. E., Handley, R. M. O., Schetters, T., Bowman, D., & Vercruysse, J. (2014). Veterinary Parasitology World Association for the Advancement of Veterinary Parasitology (WAAVP): Guideline for the evaluation of drug efficacy against non-coccidial gastrointestinal protozoa in livestock and companion animals. *Veterinary Parasitology*, 204(3–4), 81–86.
- Gibson, W. (2009). Species-specific probes for the identification of the African tsetse-transmitted trypanosomes. *Parasitology*, 136, 1501–1507.

- Government of Kenya (2010). *National climate change response strategy*; Executive Brief. Pp 9 -11
- Gunn, A., & Pitt, J. (2012). *Parasitology; An Integrated Approach*. West Sussex: Wiley-Blackwell.
- Gutierrez, C., Corbera, J., & Morales, B. P. (2006). *Trypanosomosis in Goats: Current status*. N.Y.: New York Academic of Sciences.
- Huang, C., Wang, L., Pan, C., Yang, C., & Lai, C. (2014). Investigation of gastrointestinal parasites of dairy cattle around Taiwan. *Journal of Microbiology, Immunology and Infection*, 47(1), 70–74.
<https://doi.org/10.1016/j.jmii.2012.10.004>
- Hendrix, C. M., & Robinson, E. (2006). *Diagnostic Parasitology for Veterinary Technician*. Mosby Elsevier Inc.
- Intergovernmental Panel on Climate Change; (IPCC). (2007). *Impacts, Adaptation and Vulnerability. Contribution of Working Group II to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change*. (M. C. Parry, Ed.) Cambridge: Cambridge University Press.
- Isaac, L. A., (2013). Haematological properties of different breeds and sexes of rabbits. *18th Annual Conference of Animal Science Association of Nigeria* (pp. 24-27). Nigeria: Animal Science Association of Nigeria.
- Isaac, C., Ciosi, M., Hamilton, A., Scullion, K. M., Dede, P., Igbiosa, I. B., Turner, C. M. R. (2016). Molecular identification of different trypanosome species and subspecies in tsetse flies of northern Nigeria. *Parasites & Vectors*, 9(1), 301.
<https://doi.org/10.1186/s13071-016-1585-3>

- Jacobs, D. F., Gibbons, L., & Hermosilla, C. (2016). *Principles of Veterinary Parasitology*. Oxford, UK: WILEY Blackwell.
- Jonsson, N. N., Bock, R. E., Jorgensen, W. K., Morton, J. M., Stear, M. J. (2012). Is endemic stability of tick-borne disease in cattle a useful concept? *Trends in Parasitology* 28, 85–89.
- Kagenda, G. A., & Angwech, H. (2018). Cross-sectional prevalence of gastrointestinal helminth parasites in cattle in Lira District , Uganda. *Tropical Animal Health and Production*.
- Kamani J. A., Sannusi O.K., Eqwu G. I., Dogo T. J., Tanko S., Kemza A. E., Takarki D. S., Gbise, (2010): Prevalence and significance of haemoparasitic infections of cattle in North-Central, Nigeria. *Vet. World*, 3, 445-448.
- Kanyari, P. W. N., Kagira, J. M., and Mhoma, R. J. (2009): Prevalence and intensity of endoparasites in small ruminants kept by farmers in Kisumu Municipality, Kenya
- KARI. (1999). Integrated Helminth Control. *KARI technical note* , 2, p. 54.
- Khan, T., & Zafar, F. (2005). Haematological Study in Response to Varying Doses of Estrogen in Broiler Chicken. *International Journal of Poultry Science* , 4 (10), 748-751.
- Kothari, C. (2005). *Research Methodology* (2nd edition ed.). New Delhi: New Age International Publishers.

- Kristjanson, P.M., Swallow, B.M., Rowlands, G.J., Kruska, R.L., De Leeuw, P.N. (1999). Measuring the costs of African animal trypanosomiasis, the potential benefits of control and returns to research. *Agricultural Systems* 59, 79–98.
- Kubkomawa, I. H., Tizhe, M. A., Emenalom, O. O., & Okoli, I. C. (2015). Handling , reference value and usefulness of blood biochemical of indigenous pastoral cattle in tropical Africa : *Dynamic Journal of Animal Science and Technology*, 1(2), 18–27.
- Levinson, W. (1998). *Review of Medical Microbiology and Immunology* (9th Edition ed.). New York: Lange Medical Books/McGraw-Hill Publishers.
- Machila, N. W., McDermott, J., Welburn, C., Maudlin, I., & Eisler, M. (2003). Cattle owner's perceptions of African bovine trypanosomiasis and its control in Busia and Kwale Districts of Kenya. *Acta Tropica* , 86, 25-34.
- Magaji, A. A., Ibrahim, K., Salihu, M. D., Saulawa, M. A., Mohammed, A. A., & Musawa, A. I. (2014). Prevalence of Fascioliasis in Cattle Slaughtered in Sokoto Metropolitan Abattoir, Sokoto , Nigeria. *Advances in Epidemiology*, 1-5
- Magona, J. W., Walubengo, J., & Kabi, F. (2011). Response of Nkedi Zebu and Ankole cattle to tick infestation and natural tick-borne, helminth and trypanosome infections in Uganda. *Tropical Animal Health and Production*, 43(5), 1019–1033. <https://doi.org/10.1007/s11250-011-9801-9>
- Majiwa, P. H. (1986). Evidence for genetic diversity in *Trypanosoma congolense*. *Parasitology* , 93, 291-304.

- Mansfield, J. (1990). Immunology of African Trypanosomosis. In J. Mansfield, & W. D.J. (Ed.), *Modern Parasite Biology: Cellular, Immunological and Molecular Aspects*. New York: W.H. Freeman
- Masake, R. A. (2002). The Application of PCR-ELISA to the detection of *Trypanosoma brucei* and *T. vivax* infections in Livestock. *Veterinary Parasitology* , 105, 179-189.
- Masiga, D. K., Okech, G., Irungu, P., Ouma, J., Wekesa, S., & Ouma, B. (2002). Growth and Mortality in Sheep and Goats under High Tsetse Challenge in Kenya, *Tropical Animal Health and Production* 34, 489–501.
- Merck Manual (2012). *Haematologic Reference Ranges*. Retrieved Dec 2015 from Merck Veterinary Manual: <http://www.merckmanuals.com/>
- McCusker. (2001). *Epidemiology in Community Health, A Self-teaching Manual for Rural Health Workers*, Revised Edition. English Press Ltd. Pg 5 – 139.
- Ministry of Livestock and Development (1985) *Annual Report*, Livestock Development Division, Ministry of Livestock Development, Nairobi, Kenya,
- Ministry of Livestock Development. (2010). *Strategic Plan 2008-2012*. pp. 1-3.
- Ministry of Livestock Development. (2011, February). *Strategy for Tsetse and Trypanosomiasis Eradication in Kenya*. 2011-2021. pp. 11-17.
- Minjauw, B., McLeod, A. (2003). *Tick-borne diseases and poverty*. The impact of ticks and tick-borne diseases on the livelihood of small- scale and marginal livestock owners in India and eastern and southern Africa. Research report, DFID Animal

- Health Programme, Centre for Tropical Veterinary Medicine. University of Edinburgh, UK, 116
- Mohamaden, W. I., Sallam, N. H., & Abouelhasan, E. M. (2018). International Journal of Veterinary Science and Medicine Prevalence of Eimeria species among sheep and goats in Suez. *International Journal of Veterinary Science and Medicine*, 6(1), 65–72. <https://doi.org/10.1016/j.ijvsm.2018.02.004>
- Moore, S., Shrestha, S., Vuong, H., (2012). Predicting the effect of climate change on African trypanosomiasis; Intergrating epidemiology with parasite and vector biology. *Ecology* 1-35
- Murray, R. K. (2003). *Harper's Illustrated Biochemistry a Lange Medical Book* (26th Edition ed.). USA: The McGraw-Hill Companies, Inc.USA, 622-701
- Mwacharo, J., & Drucker, A. (2005). Production objectives and management strategies of livestock keepers in South-East Kenya: implicatios for a breeding programme. *Tropical Animal Health Production* , 37, 635-652.
- National Museums of Kenya (NMoK -2010). Department of Museums, Sites and Monuments. The African Great Rift Valley - The Marakwet Escarpment Furrow Irrigation System; <https://whc.unesco.org/en/tentativelists/5503/:2> (accessed 7/09/2018)
- Nemetschke, L., Eberhardt, A.G., Viney, M.E., and Streit, S. (2010). A genetic map of the animal-parasitic nematode *Strongyloides ratti*. *Molecular and Biomechemistry Parastilogy*. 169, 124-127
- Nganga, C. J., Maingi, N., Kanyari, P. W. N., & Munyua, W. K. (2004). Development, Survival and Availability of Gastrointestinal Nematodes of Sheep on Pastures in

- a Semi-arid Area of Kajiado District of Kenya. *Veterinary Research Communications*, 28, 491–501.
- Ng'wena, Magak A G, Mwaniki, D. M., Chemwolo, L. K. and Ndiema. M. (2011). Effects of *T. Congolense* Infection on Hematological Indices, Liver , Spleen and Lymph nodes of Male Goats from Kerio Valley District ... *J. Agri. Pure Appl. Sci. Technol.*, 9(March), 1–15.
- Nzlawaha, J., Kassuku, A. A., Stothard, R.J., Gerald, C.C. and Eisler, M. C. (2014). Trematode infections in cattle in Arumeru District, Tanzania are associated with irrigation. *Parasites & Vectors*, 7: 107
- Odoi, A., Gathuma, J. M., Gachuri, C. K., & Omoro, A. (2007). Risk factors of gastrointestinal nematode parasite infections in small ruminants kept in smallholder mixed farms in Kenya. *BMC Veterinary Research*, 3(1), 6.
- Olafedehan, C. O. (2010). Effects of residual cyanide in processed cassava peel meals on haematological and biochemical indices of growing rabbits. *35th Annual Conference of Nigerian Society for Animal Production*, 212.
- Onasanya, G. O., Oke, F. O., Sanni, T. M., & Muhammad, A. I. (2015). Parameters Influencing Haematological, Serum and Bio-Chemical References in Livestock Animals under Different Management Systems. *Open Journal of Veterinary Medicine*, 5(August), 181–189
- Onono, J.O., Wieland, B., Rushton, J., 2013. Constraints to cattle production in a semiarid pastoral system in Kenya. *Tropical Animal Health and Production*. 45, 1415–1422.

- Opara, M.N., Udevi, N. and Okoli, I.C (2010). Haematological Parameters And Blood Chemistry Of Apparently Healthy West African Dwarf (Wad) Goats In Owerri, South Eastern Nigeria. *New York Science Journal*, 3(8), 68 - 72
- Perry, B.D., & Young, A. S. (1995). The past and future roles of epidemiology and economics in the control of tick-borne diseases of livestock in Africa: the case of theileriosis. *Prev. Vet. Med.* 25, 107 -120
- Picozzi, K. T. (2002). The Diagnosis of Trypanosome Infections: Application of Novel Technology for Reducing Disease Risk. *African Journal of Biotechnology*, 1, 39-45.
- Randolph, S. (2005). Tick ecology: processes and patterns behind the epidemiological risk posed by ixodid ticks as vectors. *Parasitology*, 129, 37-67.
- Rege, J. E. O., Kahi, A., Okomo-Adhiambo, M., Mwacharo, J., & Hanotte, O. (2001). *Zebu cattle of Kenya: Uses, performance, farmer preferences and measures of genetic diversity*. Genetic Resource Research, 3 - 10
- Rehman, A., Nijhof, A. M., Sauter-louis, C., Schauer, B., Staubach, C., & Conraths, F. J. (2017). Distribution of ticks infesting ruminants and risk factors associated with high tick prevalence in livestock farms in the semi- arid and arid agro-ecological zones of Pakistan. *Parasites & Vectors*, 10(190), 1–15.
- Riond, B. M.-L. (2008). Concurrent Infections with Vector Borne Pathogens Associated with Fatal Anaemia in Cattle. *Comp Clin Pathol* , 17, 171-177.
- Sang, R., Onyango, C., Gachoya, J., Mabinda, E., Konongoi, S., Ofula, V., Miller, B. (2006). Tickborne arbovirus surveillance in market livestock, Nairobi, Kenya. *Emerging Infectious Diseases*, 12(7), 1074–1080.

- Seifert, H. (1996). *Tropical Animal Health*. Dordrecht/Boston: CTA Kluwer Academic Publishers. Dordrecht/Boston, 151 - 168
- Sekoni, V. (1994). Reproductive disorders caused by animal trypanosomosis. *A Review Theriogenology* , 42 (4), 557-570.
- Shah, S.S.A., Khan, M.L., Rahman, H.U. (2017). Epidemiological and hematological investigations of tick-borne diseases in small ruminants in Peshawar and Khyber Agency, Pakistan. *Journal of Advances in Parasitology* , 4 (1), 15-22.
- Shawaf, T., Hussen, J., Al-zoubi, M., Hamaash, H., & Al-busadah, K. (2018). Impact of season , age and gender on some clinical , haematological and serum parameters in Shetland ponies in east province , Saudi Arabia. *International Journal of Veterinary Science and Medicine*, 6(1), 61–64.
<https://doi.org/10.1016/j.ijvsm.2018.03.007>
- Short, E.E., Caminade, C., Thomas, B.N., (2017). Climate change contribution to the emergence or of parasitic diseases re-emergence. *Infectious Diseases*, 10
- Singh, S. K. (2012). Climate Change Impacts on Livestock and Adaptation Strategies to Sustain Livestock Production. *J Vet Adv* 2 (7), 407-412.
- Soetan KO, Akinrinde AS, Ajibade TO (2013). Preliminary studies on the haematological parameters of cockerels fed raw and processed guinea corn (Sorghum bicolor). *Proceedings*
- Sonenshine, D., & Mather, T. (1994). *Ecological Dynamics of Tick-Borne Zoonoses*. New York: Oxford University Press Inc.

- Tambuwal, F. M., Agale, B. M., & Bangana, A. (2002, March). Haematological and serum biochemical values of apparently healthy red Sokoto goats (p. 50-53). Proceeding of 27th Annual Conference of Nigerian Society of Animal Production, 17 -21
- Taylor, K., & Authie', E. (2004). *Pathogenesis of Animal Trypanosomosis*. (I. H. Maudlin, Ed.) Wallingford, Oxfordshire: CABI Publishing, 331–353.
- Taylor, M. A. (2007). *Veterinary Parasitology* (3rd Edition ed.). Blackwell Publishing, 798-837
- Thompson R.C.A, and Smith A (2011). Zoonotic enteric protozoa. *Veterinary Parasitology*, 182 issue 1 70-78
- Thrusfield M. (2005). *Veterinary Epidemiology*, 3rd edition. Blackwell science, Oxford, 233.
- Thumbi, S. M., Mcodimba, F. A., Mosi, R. O., & Jung, J. O. (2008). Parasites & Vectors Comparative evaluation of three PCR base diagnostic assays for the detection of pathogenic trypanosomes in cattle blood, 7, 1–7. <https://doi.org/10.1186/1756-3305-1-46>
- Thumbi, S., Jung'a, O., Mosi, O., & and McOdimba, F. (2010). Spatial Distribution of African Animal Trypanosomosis in Suba and Teso Districts in Western Kenya. *Biomedical Central (BMC) Research Notes* (3), 6.
- Thumbi, S.M., Bronsvort, M.B., Kiara, H., Toye, P.G., Poole, J., Ndila, M., Conradie, I., Jennings, A., Handel, I.G., Coetzer, J.A.W., Steyl, J., Hanotte, O., Woolhouse, M.E.J. (2013). Mortality in East African shorthorn zebu cattle under one year: predictors of infectious-disease mortality. *BMC Veterinay. Research* 9, 175.

- Tulu, D., & Lelisa, K. (2016). A Study on Major Gastro-Intestinal Helminths Parasites of cattle in Tulo District , West Hararghe Zone , South- Eastern Ethiopia. *Austin Journal of Veterinary Science and Animal Husbandry*, 3(2), 3–6.
- Uilenberg, G. (1995). International collaborative research: significance of tick-borne hemoparasitic diseases to world animal health. *Veterinary Parasitology* 57:19–41
- Van-Wyk, I. C., Goddard, A., Bronsvort, B.M.C., Coetzer, C., Booth, O., *et al.* (2013). Hematological profile of East African short-horn zebu calves from birth to 51 weeks of age. *Comp Clinical Pathology*, 22, 1029–1036.
- Walker A.R., Bouattour, A., Camicas, J.L., Estrada-Pena, A., *et al.*, (2003). *Ticks of Domestic Animals in Africa: a Guide to Identification of Species*. Edinburg: Bioscience.
- Waruiru, R., Kyvsgaard, N., & Thamsborg, S. (2000). Prevalence and Intensity of Helminths infections in Dairy Cattle in Central Kenya. *Veterinary Research Communications* , 24, 39-53.
- Wakelin, D. (1996). *Immunity to parasites: How parasitic infections are controlled* (2nd Edition ed.). Cambridge: University of Cambridge Press.
- Wiedosari E, Hayakawa H, Copeman B (2006). Host differences in response to trickle infection with *Fasciola gigantica* in buffalo, Ongole and Bali calves. *Tropical Animal Health and Production*, 38, 43-53.
- Wikipedia: http://en.wikipedia.org/wiki/Kerio_Valley. (accessed on: 27/09/2018)
- World Conservation Society, (2008). *'The Deadly Dozen': Wildlife-Human Disease Threats in the Age of Climate change*.

WHO. (2009). WHO Library Cataloguing-in-Publication Data Protecting health from climate change: connecting science, policy and people . © *World Health Organization 2009*.

Yabe J, Phiri IK, Phiri AM, Chembensofu M, Dorny P, Vercruysse J. (2008). Concurrent infections of *Fasciola*, *Schistosoma* and *Amphistomum* spp. in cattle from Kafue and Zambezi river basins of Zambia. *Journal of Helminthology*, 82, 373–6.

Zvinorova, P. I., Halimani, T. E., Muchadeyi, F. C., Matika, O., Riggio, V., & Dzama, K. (2016). Prevalence and risk factors of gastrointestinal parasitic infections in goats in low-input low-output farming systems in Zimbabwe. *Small Ruminant Research*, 143, 75–83.

APPENDICES

Appendix I: Sequences of primers

Primer ID	Sequence	Species	Product Size	Cycles No.
ITS1 CF	CCG GAA GTT CAC CGA TAT TG	<i>Trypanozoon;</i> <i>T. congolense</i> <i>savannah,</i> <i>T congolense kilifi;</i> <i>T. congolense</i> <i>forest, T. vivax</i>	480; 700; 620; 700; 250	
ITS1 BR	TTG CTG CGT TCT TCA ACG AA			
ITS11	GAT TAC GTC CCT GCC ATT TG	<i>T. congolense</i> <i>forest;</i>	1513; 1422	Two rounds, 35 cycles, each, 95 C for 7 min; the second round PCR was 94 C for 1 min, seeded with 1 ul of the first 55 C for 1 min, round 72 C for 2 min.

		<i>T. congolense</i>	
		<i>kilifi</i>	
ITS12	TTG TTC GCT ATC GGT CTT CC	<i>T. congolense</i>	1413
		<i>savanna</i>	
ITS13	GGA AGC AAA AGT CGT AAC AAG	<i>T. brucei s.l.</i>	1207 - 1224
ITS14	TGT TTT CTT TTC CTC CGC TG	<i>T. theileri; T.</i>	988; 954;
		<i>simiae tsavo; T.</i>	850; 611
		<i>simiae; T. vivax</i>	

Appendix II: Normal Haematological Ranges of Livestock

a) Haematological value ranges from uninfected cattle summaries

<i>Blood</i>	<i>Kerio valley</i>	<i>Reference values</i>	<i>Reference values</i>
<i>Parameters</i>	<i>blood values</i>	<i>(Merck's Manual</i>	<i>(Vet lab Techniques</i>
			<i>– MOL, Kenya</i>
WBC 10 ³ /mm ³	2 - 23	4 – 12	4 – 12
Hgb (g/dl)	7 – 15.6	10 - 15	8 – 14
RBC	4 – 9.2	5 - 10	5 – 10
Granulocytes%	0.2 - 10.4		
PCV/HCT (%)	20 – 46.3	30 - 45	24 – 48
MCV um ³	33 – 57	39 - 55	40 – 60
MCH pg	12.5 – 19	13 - 17	11 – 17
MCHC g/dl	30 – 38	30 – 36	26 – 34
PLT 10 ³ /mm ³	38 – 1005	300 - 800	
Lymphocytes %	27.6 - 90.5	45 - 75	45 – 75
Monocytes %	0.1 – 5	0 – 8	2 – 7
Neutrophils%	0.9 – 57	15 - 33	15 - 45
Eosinophils%	0 – 4.8	0 – 20	2 – 20
Basophils%	0.4 – 7.4	0 - 2	0 – 2
Leucocytes %	0 – 36.1		
NRBC%	2.8 – 127		

b) Normal Reference values (Merck Manual (2012) in comparison with haematological parameters of uninfected sheep in Kerio Valley

Blood values	Sheep	Reference values (Merck's Manual)	Reference values (Vet lab Techniques – MOL, Kenya)
WBC $10^3/\text{mm}^3$	2.0 - 14	4 – 8	4 – 12
Hgb (g/dl)	2.0 - 12	9 – 15	8 – 16
RBC	1.0 - 8	9 – 15	8 - 16
PCV/HCT (%)	28 - 53	27 - 45	24 – 50
MCV μm^3	33 – 44	28 – 40	23 – 48
MCH pg	9 – 12	8 – 12	9 – 12
MCHC g/dl	26 – 31	31 – 34	29 – 35
PLT $10^3/\text{mm}^3$	43 - 3556	800 - 1100	
Lymphocytes %	63 - 93	40 - 55	40 – 70
Monocytes %	4 – 12.4		1 – 6
Neutrophils%	5.3 – 42.9	10 – 50	10 – 50
Eosinophils%	0.1 – 2.8	0 – 10	1 – 10
Basophils%	0.5 – 4.5	0 – 3	0 - 3
Leucocytes %	5.8 – 23.2		

Appendix III: Research pictures

Drawing of blood from Jugular vein (Marakwet West)



Drawing of blood from Jugular vein (sheep) (Marakwet East)

Source: Author 2017



Sample Processing in the field (Marakwet West)

(Source: Author, 2016)



Deworming/drenching after sample collection (Keiyo North)



Sample collection (Soy- Keiyo South)

(Source: Author, 2016)



Maseno University molecular lab

(Source: Author, 2017)

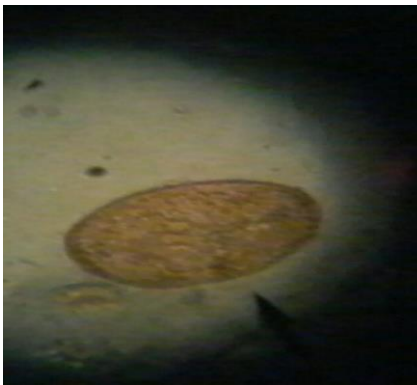


Strongyloide egg from x200

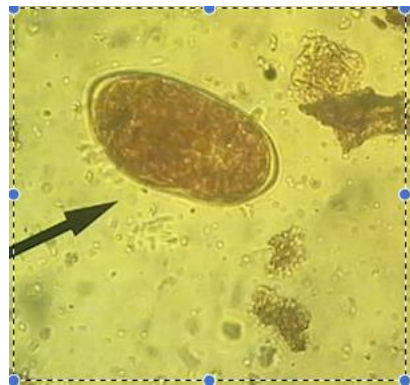


Taenid egg found in goat x200

(Source: Author, 2017)



Fasciola egg from goat x200



Heamonchus contortus egg x200

(Source: Author, 2017)



Sporulated oocyst from sheep



Tsetse fly x40

(Source: Author, 2017)

Appendix IV: Research Permit

Appendix V: Ethical clearance

Appendix VI: Similarity Report