EPIDEMIOLOGY OF GASTROINTESINAL PARASITES OF HIROLA (*BEATRAGUS HUNTERI*, SCLATER, 1889) AND LIVESTOCK IN SOUTHERN KENYA

BY

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DECLARATION

Declaration by Candidate

This thesis is my original work and has not been presented for a degree in any other University. No part of this thesis may be reproduced without prior permission of the author and/or University of Eldoret.

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This thesis has been submitted with our approval as University supervisors.

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DEDICATION

I dedicate this work to my wonderful mother, Mary Kaari for her care and support, my fiancée Joy Tsonie for her special love and inspiration, and my brother and sisters for always being there for me unconditionally. You are all very special me.

ABSTRACT

The rapid decline in the population of Hirola over the last century has caused great concern among conservationists. Together with factors such as predation, drought, habitat loss, competition for resources with livestock and poaching, diseases including parasitoses have been implicated in this sudden decline. Parasite driven declines in wildlife have become increasingly common and pose significant risks to natural populations. A study on the epidemiology of gastrointestinal parasites of Hirola (*Beatragus hunteri*) was conducted between September 2009 and March 2010. The main goals were to determine the prevalence and intensity of gastrointestinal parasites in Hirola and livestock, to investigate the influence of area, season and host characteristics of age and sex on prevalence and intensity of gastrointestinal parasites in Hirola and livestock and to determine the level of aggregation of gastrointestinal parasites in Southern Kenya. The results showed that both Hirola and livestock were infected by a wide variety of strongyles, trematodes, cestodes and coccidia. These parasites differed significantly in terms of prevalence and intensity in the hosts with strongyles being the most prevalent (67.97%) and coccidia having the highest intensity of infection (537.78 oocysts per gram ±135.59SE). Season and age were found to be the main factors influencing infection patterns in both Hirola and livestock with significantly higher prevalence ($\chi^2 = 9.928$; df = 1, p = 0.002) and intensity ($F_{1, 409} = 23.36$; p = 0.001) were being recorded during the wet than dry season and higher prevalence being observed among the young than adult animals. Generally, females had higher prevalence but slightly lower intensities of infections than males though the difference was not statistically significant (prevalence: $\chi^2 = 0.023$; df = 1, p = 0.878; intensity: $F_{1, 409} = 0.010$; p = 0.921. Apparently, Hirola in Ishaqbini had slightly higher prevalence and intensity than those in Tsavo with contrary observations being made among cattle. The results also indicated high levels of aggregation of individual parasite taxa among the hosts ($k<1$). The features of these results showed the need to target the young Hirola (or calves) in the future control and management of gastrointestinal parasites at the Hirola-livestock interface in Southern Kenya as these appeared to be at a higher risk. This should consider the seasonal patterns influencing prevalence and intensity of the parasites. In future, a study to develop a checklist of gastrointestinal parasites of both Hirola and livestock in Southern Kenya is also recommended in order to better understand the transmission of gastrointestinal parasites between the two herbivores.

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ABBREVIATIONS AND ACRONYMS

EPG: Eggs per gram

FEC: Faecal egg count

FOC: Faecal oocyst count

GFEC: Geometric Faecal egg count

GFOC: Geometric faecal oocyst count

OPG: Oocyst per gram

OPERATIONAL DEFINITION OF TERMS

Co-infection - Simultaneous infection of a host organism by two or more pathogens.

Multiple infections - An infection in which an individual is infected by parasites of more than one species

Parasite aggregation-A distribution of parasites amongst hosts is said to be aggregated, or overdispersed, if parasites are found to co-occur in particular hosts more often than if the parasites were distributed at random amongst all hosts.

Superinfection - a new infection occurring in an individual having a pre-existing infection

Virulence - the degree of pathogenicity of a microorganism as indicated by the severity of disease produced and the ability to invade the tissues of the host; by extension, the competence of any infectious agent to produce pathologic effects

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CHAPTER ONE

INTRODUCTION

1.1. Background

Parasites are a component of a normal functioning ecosystem. Commonly they occur at low-intensities of infections which are often asymptomatic. However, anthropogenic changes may alter their transmission rates, host range, and virulence (Daszak *et al.*, 2000), posing a great threat to wild animals. Unfortunately, though parasites occasionally cause sudden and unexpected local decline in abundance of species (Cleaveland *et al.,* 2001), the risk posed by parasitoses in an effort to conserve endangered species is usually overlooked until a major problem occurs, (Muoria *et al.*, 2005).

The rapid decline in the population of the Hirola (*Beatragus hunteri*, Sclatter, 1889) over the last century has caused great concern among conservationists. Together with factors such as predation, drought, habitat loss and degradation, competition for resources with livestock and poaching, diseases have been implicated in this sudden decline (Andanje and Ottichilo, 1999; Butynski, 1999) and still pose a potential threat to the restoration of the species. Presently with a population of just about 600 animals in the wild (Andanje and Ottichilo, 1999; Butynski, 1999; Dahiye and Aman, 2002), Hirola are critically endangered (Mallon and Hoffmann, 2007).

Human settlements and domestic animals are a common sight within the Hirola range (Muthoni, 2009). Hirola, sheep, goats and cattle all belong to the taxonomic family

Bovidae and have similar feeding habits. This does not only raise the question of increased competition for resources with livestock (Butynski, 1999; Dahiye and Aman, 2002, Andanje, 2002) but also the potential for transmission of pathogens between Hirola and domestic animals. Since a number of the highly pathogenic gastrointestinal parasites such as the strongyle nematodes are generalists, able to infect multiple phylogenetically close host species (Zaffaroni *et al*., 2000; Matthee *et al.*, 2004; Turner and Getz, 2010),the intense Hirola-livestock interface possibly results in high potential for pathogen exchange especially those that can be acquired from contaminated environments. Furthermore, Bunderson (1985) also observed that although Hirola avoided livestock, their density was higher in areas that were heavily used but not overgrazed by livestock. They appeared to be attracted to areas where the traditional Somali herding practices were used; in which livestock was highly mobile and overgrazing was largely absent.

1.2. Statement of the Problem

Hirola is one of the world's most endangered genera of large mammals and perhaps the world's rarest and most endangered antelope. It is the sole survivor of a formerly diverse group, and is sometimes referred to as a living fossil. Once common throughout Eastern Africa, it has suffered a devastating decline in the last three decades, with numbers plummeting from around 14, 000 in the 1970s to an estimated 600 today (Andanje and Ottichilo, 1999; Butynski, 1999; Dahiye and Aman, 2002). For example, the republic of Somalia had 2,000 Hirola in 1979, but has few, if any, today (Butynski, 1999).

In 1963, a population of Hirola was introduced into Tsavo East National Park, about 200 km south-east of the south-eastern limit of the species' known natural range. It is believed that most of them perished soon after release and that the size of the "effective founder population" was only 11 to 19 animals (Butynski, 1999). Despite this, a second group was moved to the park in 1996 as one of the earnest efforts of their conservation. Of late, the translocated population in Tsavo East National Park numbers 105 individuals, an increase from the 56 to 76 animals in 1995/1996 (Andanje and Ottichilo, 1999). The surviving Hirola are threatened by drought, poaching, disease, competition with livestock and habitat loss, (Butynski, 1999; Andanje, 2002; Mallon and Hoffmann, 2007).

Intensive conservation efforts are needed if this rare antelope is to survive. Recommendations for the long-term conservation of Hirola in Kenya have been included in a conservation action plan (Magin, 1996) and a conservation evaluation report, (Butynski, 1999). As part of the conservation and management efforts, a species based action plan, targeting diseases, pathogens and parasite management is needed (Mallon and Hoffmann, 2007). Unfortunately, Hirola is among the least studied wildlife species in Africa (Butynski, 1999) and limited information on its gastrointestinal parasites.

So far, several studies have reported recurrent droughts and habitat degradation in Hirola's range as well as competition with domestic livestock (Butynski, 1999; Andanje

and Ottichilo, 1999; Dahiye and Aman, 2002). As Ezenwa (2004b) asserts, reduced resource availability and the subsequent undernutrition could lead to intensified parasitism. Indeed, malnutrition and its interaction with parasitism has long been identified as one of the main causes of die offs among wild bovids such as the African buffalo (Sinclair, 1974). Thus there was great need to determine the possible threat posed by parasites to this critically endangered antelope.

1.3. Objectives

The main objective of this study was to assess patterns of gastrointestinal infections in the critically endangered Hirola in comparison with those of livestock in Southern Kenya

1.3.1 Specific objectives

The specific objectives of the study were:-

1. To determine the prevalence and intensity of gastrointestinal parasites in Hirola and livestock in Ishaqbini Hirola Conservancy and Tsavo East National Park
2. To find out the influence of study site, season and host characteristics of age and sex on prevalence and intensity of gastrointestinal parasites in Hirola and livestock in Ishaqbini Hirola Conservancy and Tsavo East National Park
3. To determine the level of aggregation of gastrointestinal parasites in Hirola and livestock in Ishaqbini Hirola Conservancy and Tsavo East National Park

1.4. Hypotheses

H_0 1: There is no significant difference in the prevalence and intensity of gastrointestinal parasites in Hirola and livestock

H_0 2: There is no significant influence of study site, season and host characteristics of age and sex of the host on the intensity of gastrointestinal parasite infections in Hirola and livestock in Ishaqbini Hirola Conservancy and Tsavo East National Park

H_0 3: There is no significant difference in the frequency of distribution of gastrointestinal parasites in Hirola and livestock.

1.5. Justification of the study

Biologists recognize disease as a potential threat to restoration of endangered species, (Deem *et al.,* 2001). However, conventional wildlife managers have rated parasitic infections as less important unless they caused pathological disease (Gunn and Irvine, 2003). Accounts of parasitic infections in wild populations have primarily been restricted to few reports of clinical disease manifested through conspicuous mortality (Gulland, 1995).

In cases of cross infections, initial attention is almost exclusively focused on the role of wildlife as reservoirs of infection for humans and to lesser extent domestic animals. It is just recently that focus shifted to the study of the health of wild animals (Vázquez *et al*., 2010). In case of the critically endangered Hirola, no study has been conducted so

far on the gastrointestinal parasites afflicting their health and especially where Hirola and livestock interact.

Presently, the habitat of Hirola is increasingly being encroached by domestic animals, (Butynski, 1999; Dahiye and Aman, 2002; Andanje, 2002). The potential for closer and more frequent contact with Hirola is high, and so does the potential for pathogen transmission between Hirola and livestock. Such transmission may occasionally involve generalist or multi-host pathogens which are potentially highly pathogenic.

According to Cleaveland *et al*., (2002), generalist parasites are far more likely than specialist parasites to cause outbreaks and extinctions in small host populations. For transmission of most gastrointestinal parasites to occur, direct contact is not necessary; sharing the same habitat is enough, (Soulsby, 1982). Thus, considering that Hirola is phylogenetically related to domestic livestock, it is likely that they share a number of gastrointestinal parasites. Therefore, patterns of gastrointestinal infections in Hirola could not be studied solely as an independent unit if accurate predictions of infections were to be made.

The implications of having increased multi-host parasite prevalence in the Hirola-livestock interface may be potentially far-reaching. This is especially so in view of the fact that multi-host parasites have large reservoir populations, which act as maintenance hosts that can facilitate spill-over of the parasites into small, threatened host groups, leading to massive die-offs or extirpation (Daszak *et al*., 2000). Therefore,

there was need to assess and monitor livestock parasite infection levels as an important component in managing disease risks in the Hirola population. Furthermore, the concern for the outcomes of the Hirola-livestock interface is high especially in view of the resurgence of some wildlife and domestic animal diseases in the Hirola range that were previously under control. Such resurgence has involved the incursion of the rinderpest virus that is associated with cattle in the Somalia ecosystem (Wambwa, 2002).

Consequently, this being the first study on gastrointestinal parasites of the critically endangered Hirola, it will contribute immensely to the general understanding of gastrointestinal parasitosis of the antelope. And in the light of this, it is hoped that the baseline data provided in this study will be a fundamental resource in the current and future assessments and management of disease risks at the Hirola - livestock interface in Southern Kenya.

1.6. Scope of the Study

This study was conducted in Ishaqbini Hirola Community Conservancy and Tsavo East National Park between September 2009 and July 2010 to determine the epidemiology of gastrointestinal parasites at Hirola-livestock interface. The term “interface” here refers to sharing of the same space between Hirola, cattle, goats and sheep resulting to direct or indirect contact through soil, forage, and water with which another animal has recently been in contact and has left bodily discharges, such as faeces which could be harbouring parasite propagules. The study focused on prevalence, intensity and

aggregation of protozoan and helminthic parasites mainly dwelling in the gastrointestinal tract and whose infective stages were shed in faeces of their hosts. Variations in prevalence and intensity were examined on the basis of study area and season and also host characteristics including age and sex.

1.7. Conceptual Framework

The conceptual framework is based on transmission of food-borne pathogens (Figure 1.1). Although each gastrointestinal parasite has its own population dynamics and specific interactions with its hosts, there are common aspects underlying their transmission. In this study the main focus was on food-borne parasites that live in the gastrointestinal tract of animals; therefore, faecal shedding is the main route of exit of the parasites (Lanzas *et al.,* 2011).

In the environment humidity is required for successful development and survival of parasite stages and movement of infective stages (Nielsen *et al,* 2007). Thus, spatial and temporal variations in humidity and temperatures influence infection rates with the parasites. While feeding, susceptible host may get infected after ingesting the infective parasite stages with contaminated food or water. Species specific difference in the host such as immunity (which may be influenced by age and sex of the host) determines whether the parasite establishes and reproduces in the host. Therefore by recovering the eggs and/or oocysts from faeces of the animals, prevalence and levels of intensity and aggregation of the parasites can be determined.

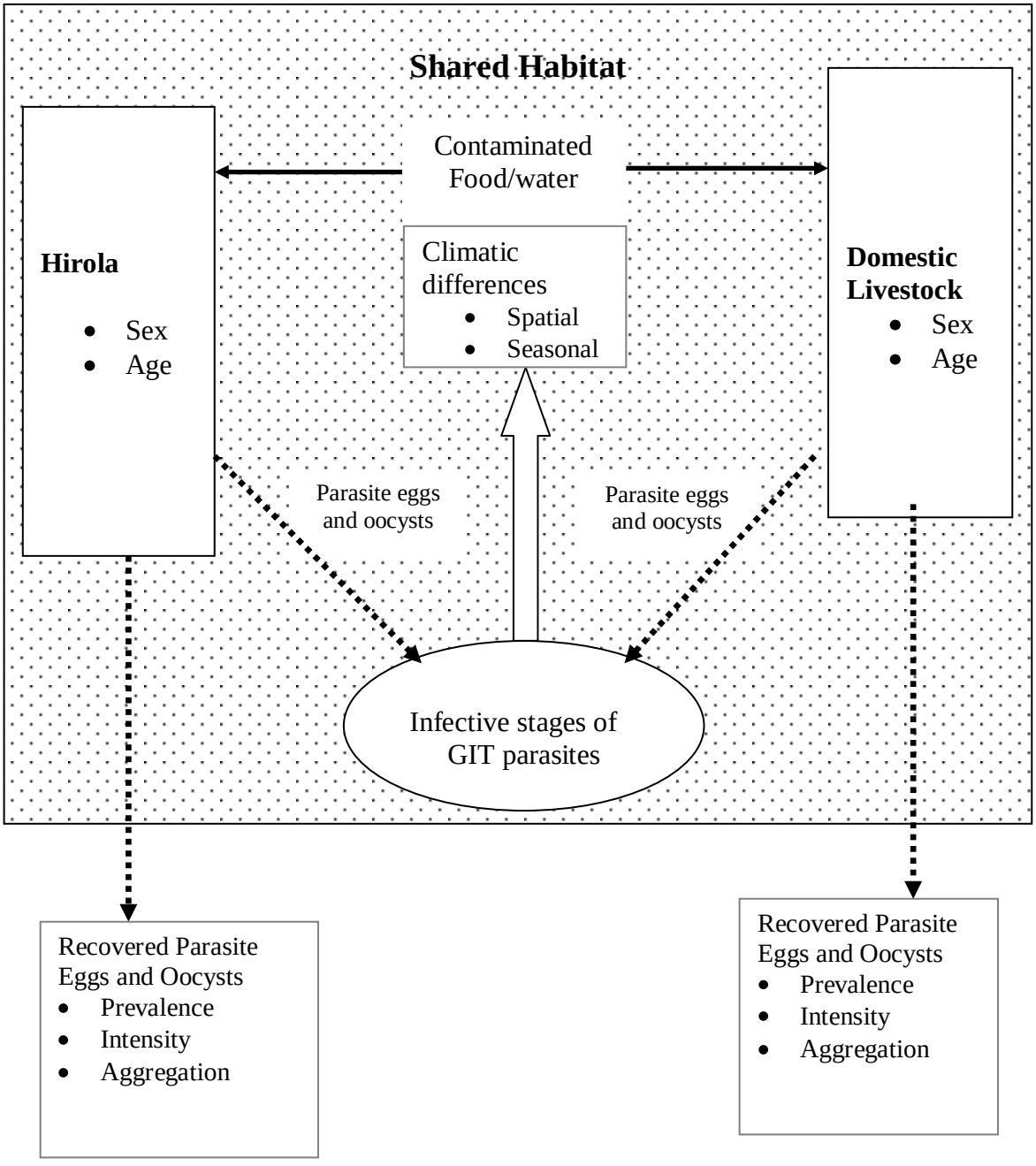

Figure 1.1. Conceptual Framework on epidemiology of gastrointestinal parasites in livestock and Hirola. (Source: Author, 2013)

Dashed arrows indicate presence of parasite eggs and oocysts in faeces of hosts whereas solid arrows in indicate presence of infective parasite stages in food and water.

CHAPTER TWO

LITERATURE REVIEW

2.1 Parasitic Infections of Ruminants and their Virulence

In recent years, interest in studying wildlife diseases has significantly increased. Initially, attention was almost exclusively focused on the role of wildlife as reservoirs of infection for humans and to a lesser extent domestic animals. More recently there has been a shift to the study of wild animal's health (Vázquez *et al*., 2010). In the same vein interest on parasites has increased tremendously in the recent past with the recognition that parasites are a major source of mortality to wild animal populations (Hudson *et al*., 2002; Moore and Wilson 2002, Vázquez *et al*., 2010). Parasites always gain their livelihood at the expense of the host animal (Samuel *et al*., 2001) and apparently, they are to be found in every ecosystem that is functioning in the normal way.

In ecology, the term "parasite" encompasses a wide range of organisms like viruses, bacteria, fungi, helminthes, protozoa and arthropods which diverge enormously in their mode of replication and transmission, generation times, elicited immune responses and diseases caused (Hudson *et al*., 2002). It is thus essential to specify the studied parasite type. In this study, the main focus is on helminthes and protozoa whose transmission stages that are shed in faeces. Broadly they include nematodes (roundworms); cestodes (tapeworms), trematodes (flukes) and coccidia, with the nematodes being by far the most important. Nearly all ruminants are exposed to these parasites during their lifetime and subsequently may succumb to the infections (Hoberg *et al.*, 2001).

In natural populations, a single-parasite genotype rarely infects only one host genotype and, similarly, a single-host genotype is rarely infected by just one parasite genotype (Rigaud *et al*., 2010). The same situation is true at the species level, with the majority of parasites having multiple host species and all hosts having multiple parasite species. These facts are not a recent discovery and could be considered as a '*lieu commun*' for most parasitologists (Combes, 2001; Pedersen and Fenton, 2007; Poulin, 2007). Presently, with more frequent pathogen outbreaks and shifts in their distributions linked to global climate change (Harvell, *et al*., 2009); the impacts of co-infections are increasing in importance especially at the wildlife-livestock interface. Similarly, we would expect this kind of a scenario for gastrointestinal parasite infection patterns at the Hirola-livestock interface.

While some empirical studies found increased virulence in multiple infections (Taylor e*t al*., 1998; Davies *et al*., 2002; Massey *et al*., 2004; Hôrak *et al*., 2006), some authors report no effect on virulence often when the competition is such that only one strain wins or even lower virulence in multiple infections (Hood, 2003; Hughes *et al*., 2004). According to Fellous and Salvaudon (2009), some parasites could be seen as protective to their hosts against various types of superinfections, which could lead to the evolution of mutualistic associations. It is also important to note that different species often have diverse resource needs and occupy different niches, making host-sharing possible, as is known for some gut macroparasites (Holmes, 2002). In such a case they 'cooperate and unite' to invade and colonize the host. This could have serious lethal outcomes.

However, the competitive outcomes of interspecific parasite interactions are complex and context dependent and this makes generalizations difficult (Lello *et al*., 2004).

Generally macroparasites exhibit an aggregated or over dispersed distribution within their host population (Wilson *et al*., 2002; Boag *et al*., 2001). Studies have also identified the importance of aggregation on the stability and dynamics of the host-parasite system (Dobson and Hudson, 1992). In macroparasites, host mortality and morbidity tends to be dose-dependent and so has most effect on individuals in the so-called 'tail' of the parasite distribution (Wilson *et al*., 2002). The proportion of hosts in this susceptible tail will be relatively larger when parasites are randomly distributed across hosts (and the variance of the distribution is low), than when the distribution is highly skewed (and the variance is high). As a consequence, parasites are likely to be relatively more important as both selection pressures (Poulin, 1993) and regulatory influences (May and Anderson, 1978) in the former case than in the latter.

2.2 Implications of Gastrointestinal Parasites on Animal Life

In domestic animals, clinical and sub-clinical signs, and parasitological and pathological features of gastrointestinal parasite infections have been reviewed by Parkins *et al*; (1989). The general consensus of ideas is that parasitoses and the resulting pathology can cause a direct negative effect on host survival especially among young animals. In addition, the financial costs of the subclinical effects on growth and reproduction and the expense of the antiparasitic drugs to combat these problems run into millions of dollars.

From studies of non-domesticated animals, Gulland (1992) provided evidence suggesting that gastrointestinal parasites also played an important role in regulating populations of free-ranging ungulates. According to Boyce (1990); Hudson *et al*., (1992); and Coop and Holmes (1996), parasites can impact host survival directly through pathologic effects and indirectly by reducing condition of fitness. Severe parasitoses can lead to blood loss, tissue damage, spontaneous abortion, congenital malformations and death, (Despommier *et al.,* 1995). Normally, sub-lethal parasite infections are more common than lethal ones, (Despommier *et al.,* 1995). Sub-lethal infections can reduce the fitness of an individual by impairing nutrition, decreasing ability of the individual to disperse, feed, escape from predators, and compete for resources or mates. They can also increase energy expenditure, (Hudson *et al.,* 1992; Coop and Holmes, 1996; and Packer *et al.*, 2003). These effects of parasites may be more difficult to detect and quantify, (Daszak *et al*., 2000).

In the light of all these revelations, is the concern among conservationists of the potential threats posed by parasites to the critically endangered Hirola. This is accentuated by the likelihood that Hirola is highly susceptible to diseases harboured by domestic livestock (Butysnki, 1999). According to Muthoni (2009), a total of 8, 434 domestic animals were found grazing in Arawale National Reserve along transect lines. There is no doubt that the scenario may be the same in the whole of the natural range of Hirola, or perhaps, even more intense.

Earlier, Bunderson (1985) observed that Hirola appeared to be attracted to areas that were heavily used by domestic animals under the traditional Somali herding practices. In such areas, livestock were highly mobile and overgrazing was largely absent. Since a number of the highly pathogenic gastrointestinal parasites are generalists (Soulsby, 1982), the close phylogenetic relationship between domestic ungulates and Hirola, possibly resulted in high potential for pathogen exchange especially those that could be acquired from contaminated environments. Several studies of both domestic and wild ruminants have also revealed that many gastrointestinal parasites infected multiple host species, both wild and domestic (Bindernagel, 1970; Wairuri *et al*., 1995; Zaffaroni *et al*., 2000; Harvell *et al*., 2009; Rigaud *et al*., 2010).

As the human population continues to increase thereby leading to more land degradation and habitat loss (Butynski, 1999; Andanje and Ottichilo, 1999; and Dahiye and Aman, 2002), it is inevitable that contact between Hirola and domestic livestock is bound to increase. Recurrent droughts as witnessed in the year 2007 to 2008 also makes the Hirola-livestock interface more intense due to increased incursion of livestock into otherwise protected areas. Furthermore, as host density increases, individual animals are more likely to come into contact with more parasite infective stages resulting in increased transmission rates (Ryder *et al*., 2007).

2.3 Factors Influencing Epidemiology of Gastrointestinal Parasitic Infections in Animals

Since parasites play an important role in host population dynamics, it is crucial to investigate factors that influence the epidemiology of gastrointestinal parasites in animals. Such knowledge is critical in the management and control of parasites.

The epidemiology of gastrointestinal parasite infections has been shown to depend on several factors. In broad terms, they include the host, the parasite and environmental factors. According to Urquhart *et al*., (1988), the ultimate occurrence of parasitic infections is as a result of host susceptibility, introduction of infective stages, alteration in host susceptibility, introduction of susceptible stock and the introduction of infection. Host factors include conditions such as nutritional status, physiological status, age, sex, breed and levels of acquired innate resistance (Tariq *et al*., 2008, Kanyari *et al*., 2009).

Poor nutrition increases susceptibility of the host to infection (Ezenwa, 2004a). When nutrition is good the animals are more able to tolerate infections (Faizal and Rajapakse, 2001). The physiological status of the host includes pregnancy and lactation. These can increase susceptibility of the animal to infections especially if nutrition is not increased to meet foetus and milk requirements. Under these circumstances, even low burdens can have detrimental effects on the food conversion efficiency of an animal, ultimately affecting foetal or neonatal growth. For example goats have been reported to be extremely susceptible during pregnancy and early lactation (Urquhart *et al*., 1988). In

case of Hirola, the peak calving period is late October and early November (Andanje, 2002) when short rains are commonly experienced in the area.

Animals develop immunity to parasitic infections with age. This has been attributed to increased resistance to infections and/or re-infections with age as a result of intake of small numbers of infective stages of the parasites which confer concomitant immunity (Assoku, 1981). Even though these animals develop immunity with age, the majority remain susceptible until they have been exposed to an infection, for instance if they are moved to an endemic area (Urquhart *et al.,* 1988). However, Boomker *et al.,* (1994) found an inverse relationship between age of goats and the mean nematode burden. In the study, very young kids were noted to have low nematode burdens. This was attributed to a diet consisting of milk and where only small amounts of vegetation containing infective larvae was consumed. In contrast, in a study by Magona and Musisi (2002), age did not have significant influence on the faecal egg count.

A number of studies have reported the influence of sex on susceptibility of animals to infections by gastrointestinal parasites (Morgan *et al.,* 2005; Tariq *et al.,* 2008; Kanyari *et al*., 2009). This disparity is attributable to genetic predisposition and differential susceptibility owing to hormonal control. In addition, according to Tariq *et al*., (2008), sex-related differences in behaviour may result in differences in exposure.

Some breeds of animals are more resistant to parasite infection than others. For example it has been shown that the Red Maasai sheep which was indigenous to East Africa was

more resistant to *Haemonchus contortus* than exotic breeds such as the Dorper (Ngingyi *et al*., 2002). Some of these variations were due to genetic factors in the host, but also they could be due to heterosis if the animals were not pure bred (Stear and Murray, 1994). In addition, factors such as the feeding behaviour based on level and time spent feeding (Halvorsen, 1986) and social organization of the animals are important in the preponderance of parasitic infections and are attributable to genetic differences between the individuals concerned. Ezenwa (2004b) also observed that along with group size and group living, territoriality and social class were important factors that shaped levels of parasite exposure and susceptibility in bovids.

The environment also plays a significant role in the epidemiology of gastrointestinal parasites. The development, survival and transmission of the free-living stages of parasites are influenced by micro-climate within the faecal pellets and herbage (Sissay *et al.,* 2008). These factors include sunlight, temperature, rainfall, humidity and soil moisture. The combined effects of these factors are responsible for fluctuations in the availability of infective stages on pasture and water and subsequently in the prevalence of the parasites in hosts (Regassa *et al.,* 2006; Sissay *et al.,* 2008, Turner and Getz 2010).This seasonal variation of parasite population dynamics has been described in a number of studies in both domestic and wild ungulates (Armour, 1980; Tembely *et al.,* 1997; Apio *et al.,* 2006; Regassa *et al*., 2006; Sissay *et al*., 2008).

Global warming and its influence on climate may play a role in the occurrence of parasitic diseases (Maposa, 2009; Polley *et al*., 2010). Back in 1960, Rowcliffe and

Ollerenshaw (1960) developed a model to predict the prevalence of fascioliasis in domestic sheep based on microclimatic observations in Great Britain. For African ungulates such data and models are not available to date (Apio *et al.*, 2006). However, many aspects of host and parasite ecology have been identified as potentially vulnerable to climate change. Among possible consequences are boundary shifts that can alter the structure and function of host-parasite assemblages (Hoberg and Brooks, 2007; Kutz *et al.,* 2008). The speed and extent of these shifts vary with place and time. For wildlife, the detection of these shifts may be hampered by a lack of baseline data for the occurrence and significance of pathogens and diseases (Maposa, 2009). However, according to Polley *et al.,* (2010), in exploring climate change as a cause of new patterns of disease, much can be learned from the many data-derived relationships between key climatic factors and host, parasite and disease ecology, and the integration of these with projections for climate change trajectories.

From this perspective, to get a comprehensive insight into the factors influencing epidemiology of gastrointestinal parasites in animals deserve a closer attention. In the present study the main focus was on seasonal and spatial (site) variations in the environment as well as differences in host age and sex.

2.4 Theoretical Framework

This study was based on the Mathematical Modelling on Transmission of Foodborne Pathogens by Lanzas *et al.,* (2011). The model is based on concepts of transmission of foodborne pathogens in a farm but also considers that while each foodborne pathogen

has its own population dynamics and specific interactions with its hosts, there are common aspects underlying their transmission. Most of the foodborne pathogens live in the gastrointestinal tract of animals; therefore, faecal shedding is the main route of pathogen excretion (Lanzas *et al.*, 2011). There is large variation in the pathogen shedding level among animals, as well as fluctuations in the level shed by a single animal.

Foodborne pathogens can often survive and even grow in the environment and in animal reservoirs other than farm animals (e.g., in pest and wildlife animals) (Oliver *et al.*, 2007). In the environment humidity is required for successful development and survival of parasite stages and movement of infective stages (Nielsen *et al.*, 2007). Thus, spatial and temporal variations in humidity and temperatures influence infection rates with the parasites. In this regard, the model is applicable when examining the transmission of gastrointestinal parasites in livestock and wildlife in a shared environment. Theoretically, there are multiple direct (e.g., faecal–oral and oral–oral) and indirect (e.g., through environment or mechanical vectors) routes of transmission (Lanzas *et al.*, 2011).

The most widely used mathematical models for infectious diseases are the susceptible-infectious-recovered (SIR) compartmental models (Lanzas *et al.*, 2011). In SIR models, the host population is divided into compartments according to its epidemiological status (e.g., susceptible, infectious, and recovered) (Figure 2.1). Susceptible animals are those not infected, but which may become infected later. Infectious animals are infected animals that shed the pathogen and, therefore, can infect other animals. Recovered

animals have immunity against the pathogen. Immunity lasts for a limited period after which animals become susceptible again.

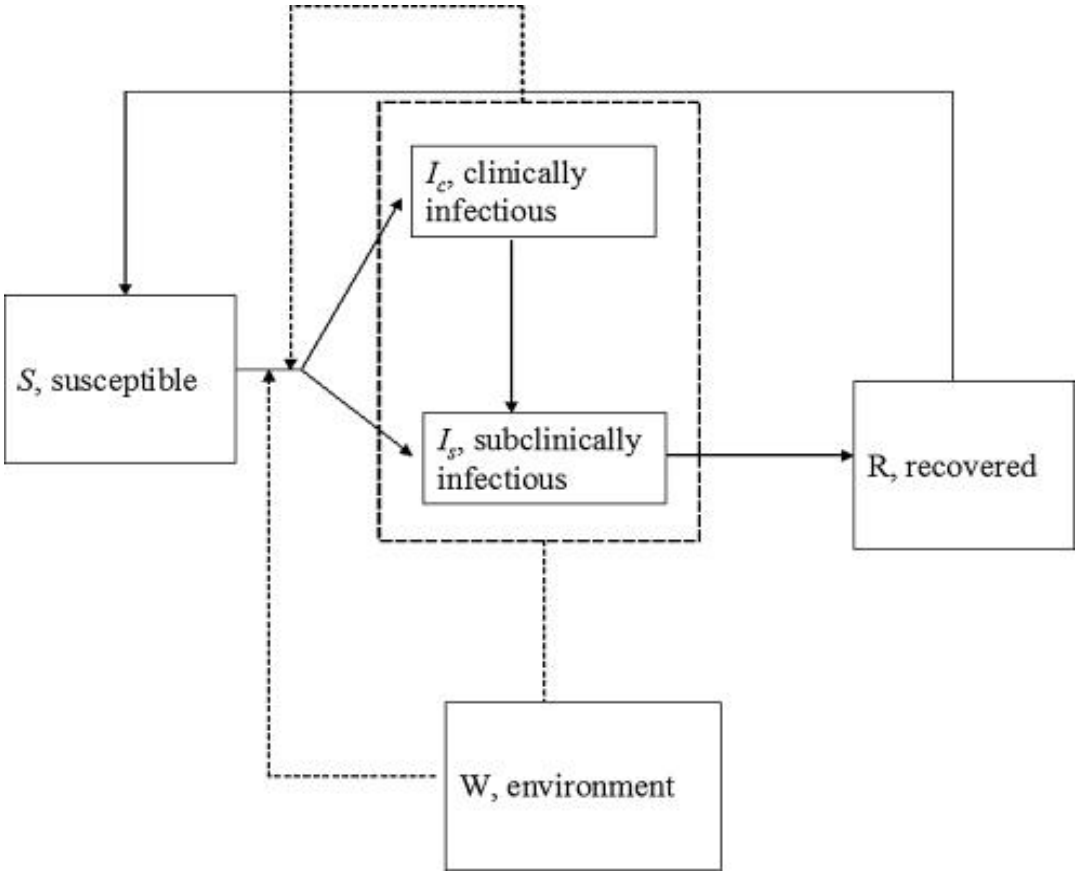

Figure 2. 1. Theoretical framework for transmission of foodborne pathogens within animal populations. (Source: Lanzas *et al.*, 2011)

Transition states for the animals include *S* (susceptible), I_c (clinically infectious), I_s (subclinically infectious), and *R* (recovered). Pathogens are shed to the environment, *W*. Transmission takes place directly, through contact with infectious animals, or indirectly, through the contact with the free-living pathogen in the environment. Solid arrows indicate movement of animals through the different transition states. Dashed arrows indicate sources of new infections.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study Area

3.1.1 Location and History

The present study was carried out in Ishaqbini Hirola Community Conservancy (IHCC) and Tsavo East National Park (Figure 3.1). Ishaqbini Conservancy is located in Masalani Division of Ijara District, Garissa County (01^{o} 55'32.4" S, 040^{o} 10' 17.6" E) in North Eastern Province of Kenya. The conservancy was registered in 2007 and covers approximately 72 km^2.

The conservancy surrounds the eastern sector of the Tana River Primate National Reserve where it is flanked by the lower part of River Tana, Kenya's longest and the only permanent river in the county. The river has rich riverine vegetation along its banks and areas of alluvial deposits as a result of seasonal flooding (Muchai *et al*., 2007). IHCC is one of the newest community conservation initiatives of the Northern Rangelands Trusts (NRT). It is managed by the local Somali pastoralist communities from Hara, Korissa and Kotile whose members come from the Abdullah clan of the Ogaden community.

The most important feature of this conservancy initiative is that it encompasses an area of land inhabited by the critically endangered Hirola. The last aerial survey conducted by Northern Rangelands Trust in conjunction with the Lewa Wildlife Conservancy in July 2008 recorded 152 Hirola in the conservancy area.

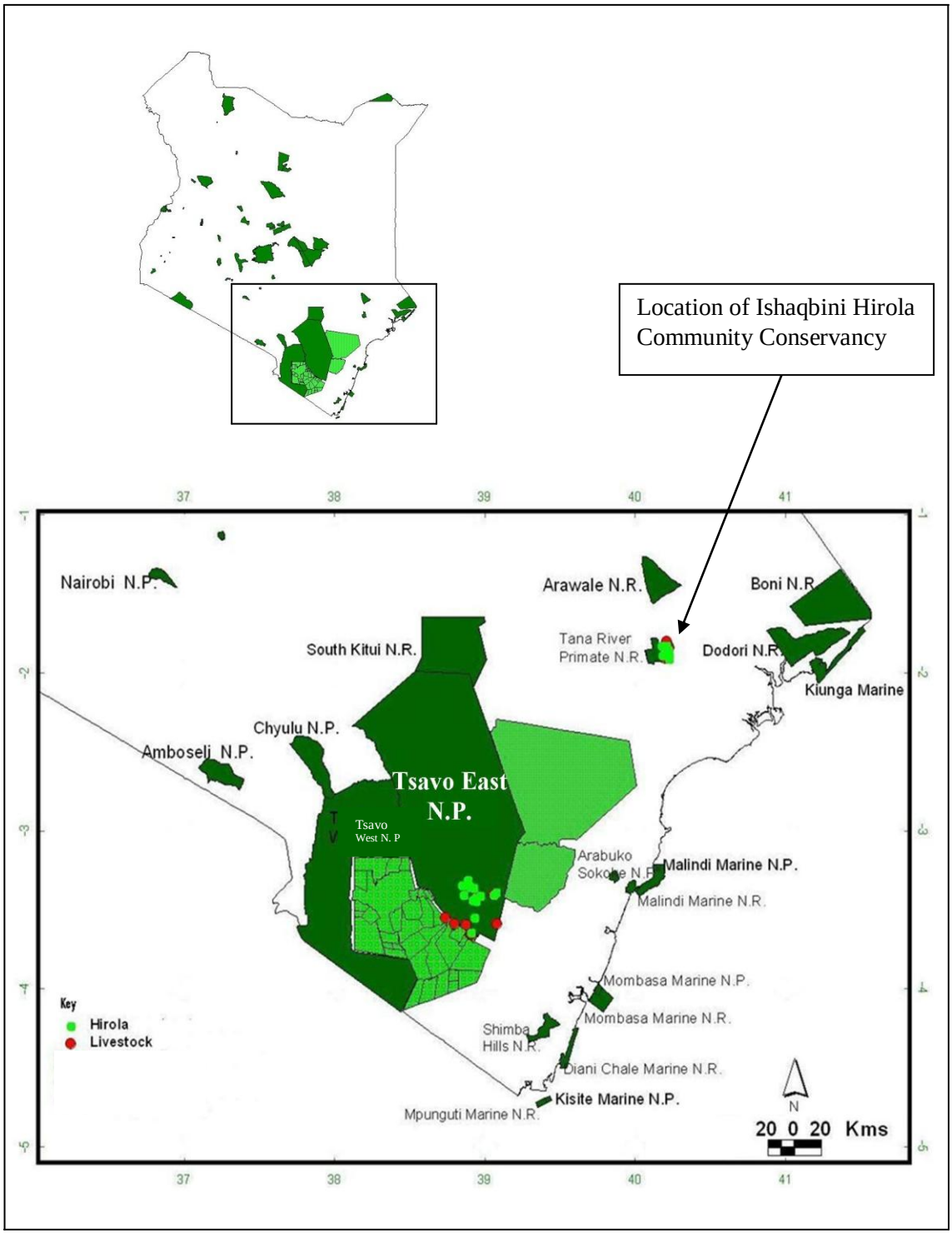

Figure 3.1. Map of Southern Kenya showing the location study sites
(Source: Author, 2013)

On the other hand, Tsavo East National Park (2° 45´ S and 38° 34´ E) is a vast, flat, monotonous and semi-arid area, located on the northern side of the main Mombasa-Nairobi highway, about 300 km southeast from Nairobi. It is Kenya's largest National Park covering an area of approximately 11,700 km^2. In 1963, a population of Hirola was introduced into the park, about 200 km south-east of the south-eastern limit of the species' known natural range. It is thought that most of these perished soon after release and that the size of the "effective founder population" was only 11 to 19 animals (Butynski, 1999). A second group was moved to the park in 1996. During both translocations the Hirola were released on Dika Plains, approximately *2* km south of Voi River. The translocated population in the park numbers approximately 105 individuals (Andanje and Ottichilo, 1999).

3.1.2 Climate

In both Ishaqbini Conservancy and Tsavo East National Park ecosystems, rainfall is distributed bi-modally, with long rains from April through June and short rains from November through December (Figure 3.2). Distinct dry seasons occur between the rains, particularly during January-March. The mean annual rainfall ranges from 350 mm in the northern extreme of the range to 700 mm on the southern edge of the range.

The preferred habitat of the Hirola lies in the 400-550 mm rainfall zone. Temperatures are high throughout the year. Annual daily minimum and maximum temperatures average about 2l°C and 30°C, respectively. The mean monthly temperature ranges are 22°-36°C, being lowest during May-July and highest during January-February (Butynski, 1999).

In Tsavo East National Park, rainfall ranges from 200-700 mm per year. The mean monthly temperature minima is 20°C and mean monthly maxima is 30°C. In Tsavo, Hirola live at an elevation of about 300-500 m which is slightly higher than on their natural range. Temperatures here are also marginally cooler.

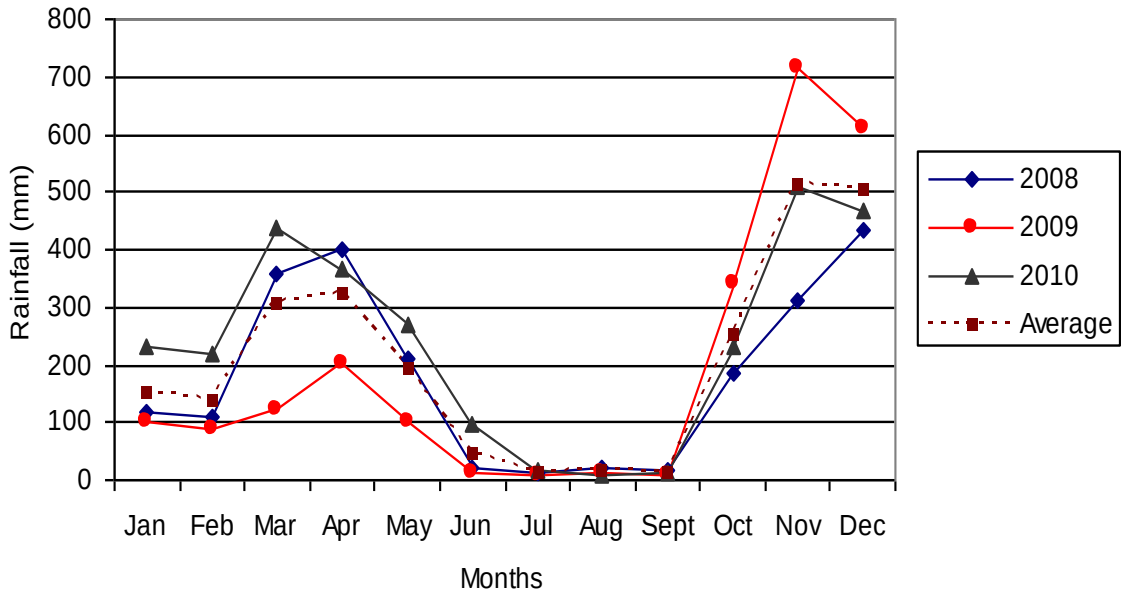

Figure 3.2. Monthly Rainfall trends in Tsavo East National Park between 2008 and 2010

3.1.3 Vegetation

Vegetation types within the natural range of the Hirola vary from lush savannah grassland in the south to open bush grassland in the centre, to dry thorn bush in the north. The natural range is bordered to the south by a humid coastal forest-savannah mosaic and to the west by a narrow band of riparian forest along River Tana. The region immediately to the west of River Tana is also arid and extremely over-grazed, with the

result that it is today largely an area of dense bush and little grass, and appears to be unsuitable habitat for Hirola (Butynski, 1999).

In Tsavo East National Park, the vegetation cover varies a lot. There are open plains alternating from grasslands and savannah bush land to semi-arid acacia scrub and woodlands. The vegetation is generally denser in the western part of the park where the annual rainfall is around 450 mm. In the eastern part, the annual rainfall is about 350 mm. The most vegetated areas of woodland and thickets are found along the rivers that cross the park. The southernmost part of the park, south of River Galana, is mostly open bush grassland. The northern part of the park is more or less dense *Acacia-Commiphora* woodland. Studies have shown that Hirola in Tsavo have habitat preferences similar to their natural range (Andanje and Ottichilo, 1999). Here, Hirola use fairly open, short, green grassland habitats where grass heights average about 17 cm. More shrubby areas are used during the dry season and more open areas during the wet season (Andanje and Ottichilo, 1999).

3.1.4 Animals

Apart from Hirola, Ishaqbini Conservancy holds several other animal species of conservation interest including the reticulated giraffe (*Giraffa camelopardialis*), cheetah (*Acinonyx jubatus*), African wild dog (*Lycaon pictus*), desert warthog (*Phacochoerus delameri*), Somali bush baby (*Galago gallarum*), buffalo (*Syncerus caffer*), lion (*Panthera leo*), leopard (*Panthera pardus*), Lesser kudu (*Tragelaphus imberbis*), bush-buck (*Tragelaphus sylvaticus*), Harvey's duiker (*Cephalophus harveyi*), Beisa oryx

(*Oryx beisa*), topi (*Damaliscus korrigum*), Tana River Red Colobus (*Procolobus rufomitrus*) and Tana Mangabey (*Cercocebus galeritus*).

Tsavo East National Park has a vast abundance of large mammals including great herds of elephant (*Loxodonta africana*, hippos (*Hippopotamus amphibious*), black rhino (*Diceros bicornis*), eland (*Taurotragus oryx*), lions (*Panthera leo*) and giraffe (*Giraffa camelopardialis*) plus a host of avifauna. Waterbucks (*Kobus ellipsiprymnus*), kudus (*Tragelaphus strepsiceros*) and dik-diks (*Madoqua kirkii*) are common along the banks of the Galana River.

3.2 Methods

3.2.1 Collection of Faecal Samples

Faecal samples were collected from late September 2009 to the end of March 2010. During this period, Hirola were searched and located in specific areas where particular groups were known to occupy.

After locating a group, size, composition and GPS location were recorded. The group was then monitored until one or more of the individuals in the group defecated. For each defaecation, the age and sex of the defaecator was recorded along with the position of the faecal sample.

The position of the faecal sample was recorded arbitrarily by estimating or describing how close or far the sample was from a given permanent mark such as a tree, shrub or

rock. The permanent mark was thereafter used to locate the position of the faecal pellets. This process was continued until an adequate number of stooling was recorded, and then collection of faecal samples would commence.

To ensure adequate sampling for each group, at least 6 samples were collected from groups with >10 individuals and at least 3 samples from groups with <10 individuals. For groups with less than five individuals it was possible at times to collect all samples from all the individuals. Samples from livestock were collected from sheep, goats and cattle found grazing within the study sites. They were also monitored and samples collected using the same procedure as for Hirola. Additional samples were collected from livestock in their resting sites in case they were located at the boundaries of the park in proximity to areas grazed by Hirola (Appendix V and VI).

Before collection, the faecal samples were examined macroscopically and noted for consistency, presence of blood, mucus, and adult or larval nematodes. A spatula was then used to scoop 10g of the sample from within the faecal mass into a collection tube containing 70% ethanol. The tube was closed and labelled with identification number, date, and time of collection, initials of the collector, site, and age/sex/identity of individual animal whenever it was possible. The tube was vigorously shaken to maximize contact between the sample and the preservative.

3.2.2 Processing of Faecal Samples

Faecal samples were evaluated for gastrointestinal parasites (GIP) using a combination of sedimentation and floatation techniques commonly used for non-invasive recovery of

parasites from faecal samples. Faecal floatation techniques are best for recovery and quantification of nematode and cestode eggs and protozoan cysts from faeces, but fail to recover trematode eggs or nematode larvae (Bowman, 2003, Turner and Getz, 2010). Sedimentation technique was used for qualitative analysis of faecal samples (MAFF, 1980) with the objective of identifying and evaluating the presence or absence (prevalence) of GI parasites in the samples.

3.2.2.1 Sedimentation Technique

Approximately 3 g of faeces was measured into a 250ml beaker. Then 46 ml of tap water was added and stirred thoroughly using a fork. Once a homogenous solution was obtained, the faecal suspension was filtered through a strainer into another 250ml beaker.

The filtered material was poured into a test tube and allowed to sediment for 5 minutes. The supernatant was removed carefully by decantation and the sediment resuspended in 5 ml of water. This was again allowed to sediment for a further 5 minutes and the supernatant discarded by decantation.

Using a teat pipette, a drop of the sediment was transferred onto 2mm x 2mm microslide. All observations made were recorded under serial slide 1 (sub sample one). A microscope with an in-built digital camera enabled detailed examination of the samples from a computer screen and also taking of photos of the parasite eggs, ova and larva (Appendix VII). The sediment in the 250 ml was agitated once more to prepare sub sample two, and the procedure was repeated to prepare sub sample three.

3.2.2.1 Floatation Technique

To quantify ova and oocyst output in faecal material, a modification of the McMaster faecal egg counting technique with saturated sodium chloride as the floatation solution (MAFF, 1980) was used. For each sample, 4g of faeces was carefully weighed out and put into a labelled vial. The sample was homogenized in 56 ml of saturated salt (NaCl) solution (specific gravity 1.2) after which it was sieved into a tube to remove large debris using a strainer.

After agitation, an aliquot was taken from the tube and pipetted into a single chamber of the McMaster slide. The tube was further agitated to fill a second chamber. The slide was then allowed to stand for 5 minutes. The two chambers were examined under x10 objective of a light microscope to identify and count all parasite eggs and cysts.

For each sample, all eggs and oocysts in the two chambers of the McMaster slide were counted and the total multiplied by 50, (a dilution factors) to determine the number of eggs and oocysts per gram of faeces. This was recorded as slide 1 after which two more McMaster slides were prepared and examined using the same procedure such that from the same sample, three subsamples were examined. This was done to increase sensitivity of the test.

3.3 Analysis of Data

In the data analysis “prevalence” was taken as the proportion of individuals examined that were shedding parasite propagules in faeces and “intensity” as the estimated number of parasite propagules shed per gram of faeces by infected individuals. To

determine whether prevalence of the parasitic infections was independent of the host, area and season, Chi-square test for association was used. The test was also used to determine whether the prevalence was independent of the individual's sex and age.

In the case of parasite intensity, the quantitative data on faecal egg counts (FEC) and faecal oocysts counts (FOC) was normalised using $\log_{10}(x+1)$ transformation and then analysis of variance (ANOVA) was used to test for differences in intensity of infection as inferred from mean eggs per gram (EPG) and oocysts per gram (OPG) of faecal material based on age and sex of the host, and seasonal and spatial variations. The results were thereafter back-transformed using antilogs (minus 1) and represented as the geometric faecal egg counts (GFEC) for strongyles and geometric faecal oocysts counts (GFOC) in case of coccidia.

Aggregation of parasites in hosts was determined by the frequency distribution of parasite burdens based on FEC and FOC. It was described using the negative binomial distribution whereby parameter *k* was used as a measure of aggregation of the parasites (Wilson *et al.,* 2002). The parameter $\boldsymbol{k}$ was determined by: $\boldsymbol{k} = (m^2 - s^2/n) / (s^2-m)$, where $\boldsymbol{k}$ is the corrected moment estimate; s^2= variance, m=mean and n = sample size. Based on this test, when k is large (>~20), the distribution converges on the *Poisson* (i.e. $s^2 \rightarrow m$); as k gets smaller, parasite aggregation increases until, as k approaches zero, the distribution converges on the logarithmic series (Wilson *et al.,* 2002). To test for the differences in aggregation of the parasites between hosts, Chi-square test for independence was used. Statistical significance for all analyses was determined at the 5% alpha level.

CHAPTER FOUR

RESULTS

A total of 410 samples were analysed for gastrointestinal parasites. These comprised of samples from Hirola (141), sheep (77), cattle (120) and goats (72).

4.1 Prevalence of Gastrointestinal Parasites

4.1.1 Overall prevalence

Analysis of data from Hirola, sheep, goats and cattle revealed that 79.75% (327) of the samples were positive for at least one classification of gastrointestinal parasite. The highest prevalence of infections was observed in goats with 83.33% (60) cases, whereas the lowest number of positive cases was observed in Hirola which had a prevalence of 73.05% (103) (Figure 4.1.) These differences in prevalence between the four hosts were not statistically significant ($\chi^2 = 1.025$; df = 3, P = 0.795).

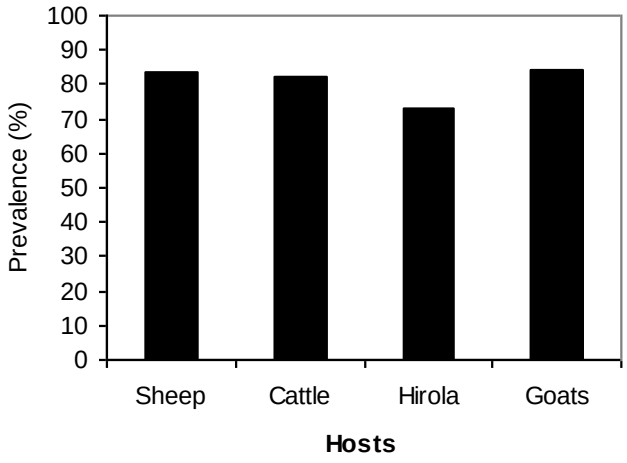

Figure 4.1. Proportions of animals testing positive for at least one classification of gastrointestinal parasites

As shown in figure 4.2 below, majority of the samples comprising 45.37% (186) of the samples analyzed for prevalence of gastrointestinal parasites had multiple infections whereas in 20.33% (83) no parasite eggs or cysts were detected. Infections with a single parasite group were observed in 34% (139) of the samples. These differences were statistically significant (χ^2 = 9.448; df = 2, P = 0.008).

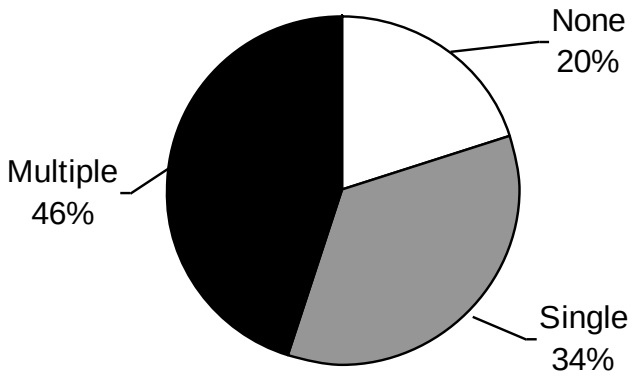

Figure 4.2. Overall proportions of multiple and single infections with gastrointestinal parasites in Hirola, sheep, goats and cattle

Patterns of multiple and single infections in host animals are shown in Figure 4.3. Goats had the highest cases of multiple infections (55.55 %; 40) whereas cattle had the highest cases of single infections (41.11%; 148). These differences in infection were statistically significant for sheep (χ^2 = 17.600; df = 2, P = 0.001); cattle (χ^2 = 11.282; df = 2, P = 0.004) and goats (χ^2 = 24.999; df = 2, P =0.001) but not for Hirola (χ^2 = 2.670; df = 2, P = 0.263).

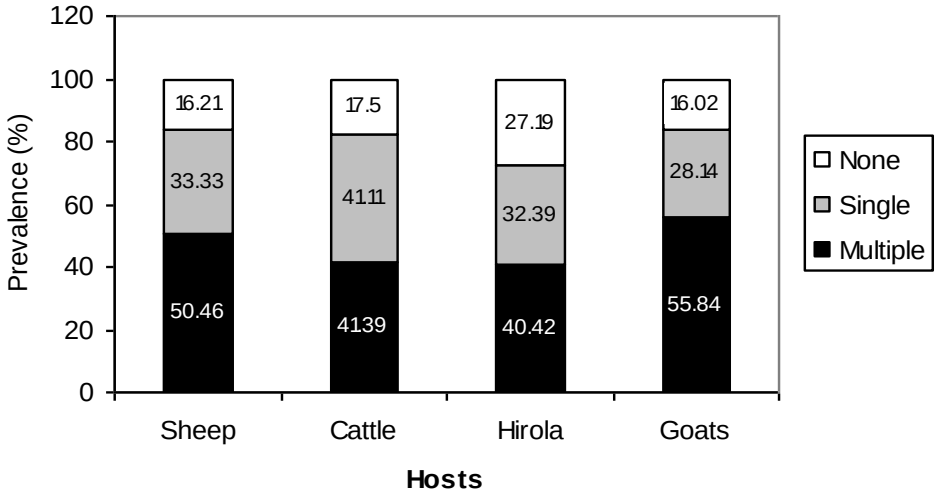

Figure 4.3. Patterns of multiple and single gastrointestinal infections in sheep, goats, Hirola and cattle

The patterns of prevalence of all parasites identified in this study are summarized in Table 4.1. In most cases it was not possible to identify the parasites beyond genus level especially the nematodes (Appendix VII). Their eggs and oocysts were conservatively assigned to taxonomic groups up to genus level to avoid identification errors especially in view of the fact that gastrointestinal parasites of Hirola have not been described earlier. Thus infection patterns were analysed based on four broad groups of parasites, namely: strongyles (nematodes), trematodes (flukes), cestodes (tapeworms) and coccidia (protozoans).

In addition some gastrointestinal parasites such as *Trichuris spp*, *Neoascaris spp* and *Monieza spp*, were recorded from one host each or just one individual and were not included in further comparison of infection patterns. In general, strongyles were present

in 67.80% (278) of the samples. This made them the most prevalent parasite group, whereas, with only 0.24% (1) cases, cestodes (*Monieza spp*.) were the least prevalent parasite group. The differences in prevalence across hosts, it was observed that samples that tested positive for *Haemochus* and *Cooperia*-like parasites were statistically significant (*Haemonchus sp*. χ^2 8.884=; df = 3, P = 0.031 and *Cooperia spp*; χ^2 = 34.929; df = 3, P=0.001) whereas others were not (Table 4.1).

Table 4.1 Summary of Prevalence of Gastrointestinal Parasites Identified

	Sheep(n=77)		Goats(n=72)		Hirola (n=141)		Cattle(n=120)		Overall(n=410)		P
	+ve	%	+ve	%	+ve	%	+ve	%	+ve	%	
Nematodes:	60	77.92	52	54.61	77	53.90	75	62.50	264	64.39	0.417
Strongyloides spp	21	27.27	20	27.78	36	25.53	27	22.50	104	25.37	0.878
Trichostrongylus spp	17	22.08	10	13.89	23	16.31	18	15.00	68	16.59	0.694
Dictyocaulus spp	2	2.60	1	1.39	1	0.71	2	1.67	6	1.46	0.795
Trichuris spp	0	0.00	1	1.39		0.00	0	0.00	1	0.24	**
Oesophagostomum spp	13	16.88	4	5.56	15	10.64	8	6.67	40	9.76	0.058
Bustonomum spp	11	14.29	4	5.56	10	7.09	13	10.83	38	9.27	0.328
Haemonchus spp	11	14.29	6	8.33	9	6.38	2	1.67	28	6.83	0.041*
Ostertagia spp	15	19.48	16	22.22	1	0.71	2	1.67	34	8.29	0.001*
Cooperia spp	2	2.60	3	4.17	3	2.13	10	8.33	18	4.39	0.109
Ascaris spp	1	1.30	1	1.39	3	2.13	3	2.50	8	1.95	0.667
Neoascaris spp	0	0.00	0	0.00		0.00	10	8.33	10	2.44	**
Coccidia: *Eimeria spp*	22	28.57	24	33.33	37	26.24	42	35.00	125	30.49	0.693
Trematodes											
Flukes	27	35.06	17	23.61	33	23.40	47	39.17	124	30.24	0.131
Cestodes:											
Monieza spp	0	0.00	1	1.39		0.00	0	0.00	1	0.24	**

+ indicates number of samples in which the parasites were found
* indicates statistically significant differences in prevalence across hosts
** indicates the parasite was found in one host only

4.1.2 Patterns of Prevalence of Specific Gastrointestinal Parasites Based on Host Characteristics

Sheep had the highest prevalence of strongyles followed by goats and cattle. Hirola had the lowest prevalence (Figure 4.4a). The prevalence levels of strongyles did not differ significantly across the hosts studied ($\chi^2 = 4.74$; df = 3, P =0.193).

Prevalence patterns of coccidia among the four animal groups studied is shown by Figure 4.4b. Goats had the highest positive 34.18% (24) cases of coccidian whereas Hirola had the fewest cases (26.71% (37). Cattle had a prevalence of 33.06% (40) and this prevalence was close to what was observed among goats. Sheep had a prevalence of 29.17% (22). The prevalence differences observed were not statistically significant ($\chi^2 = 0.871$; df = 3; P = 0.832).

As shown in figure 4.4c, with a prevalence of 38.89% (47) and 35.65% (27), most of the positive cases of trematodes were found among cattle and sheep respectively. Comparatively lower positive cases were found among Hirola and goats which had 24.11% (34) and 23.38% (17) respectively. Statistically, these differences in prevalence of trematodes across the four hosts were not significant ($\chi^2 = 6.177$; df = 3; P = 0.103).

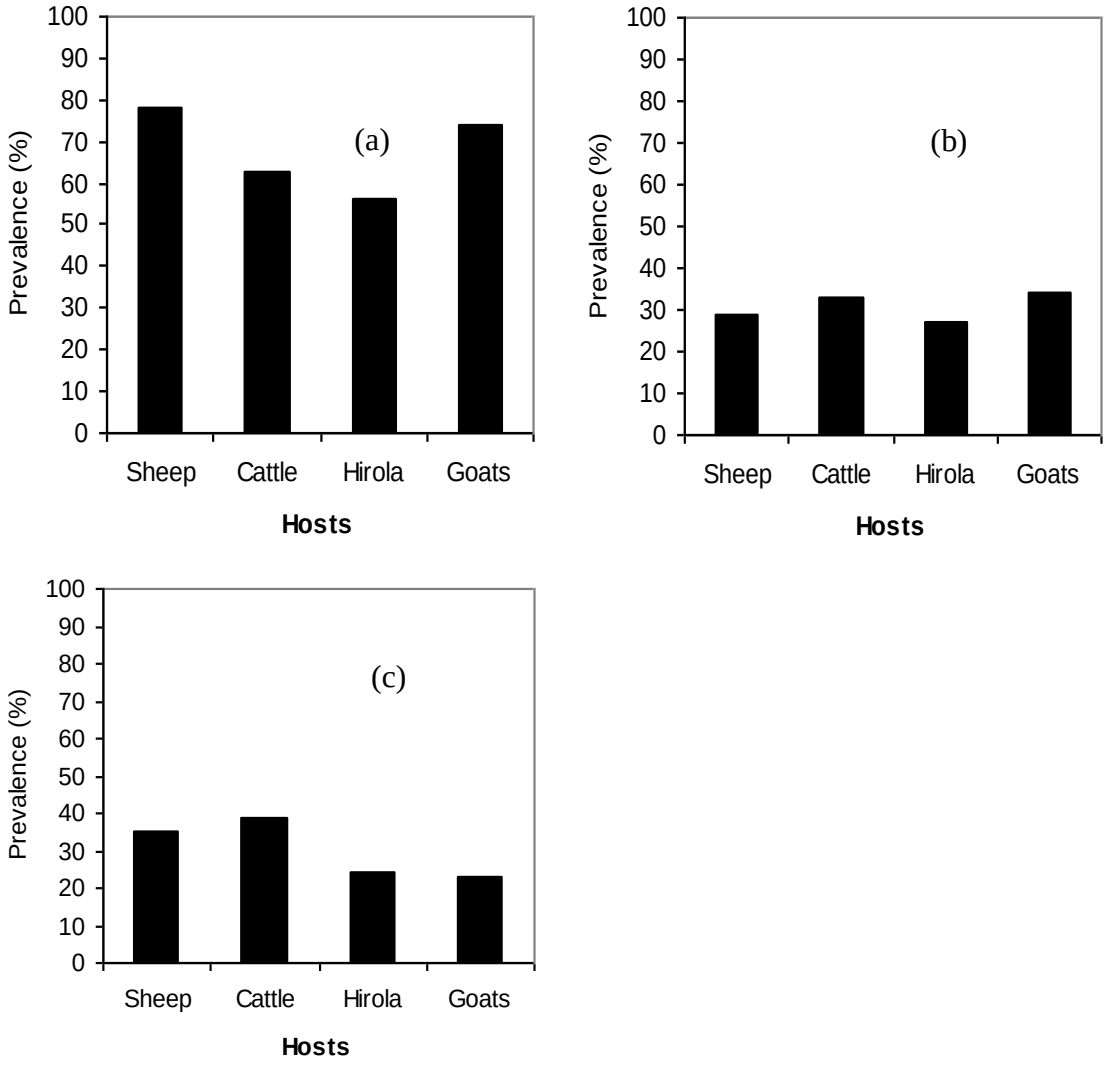

Figure 4.4. Proportions of animals infected with the three main gastrointestinal parasites

(a) Prevalence of strongyles, (b) coccidia, and (c) trematodes

4.1.3 Spatial and Temporal Variations in Prevalence of Gastrointestinal Parasites

Overall, the gastrointestinal parasites were more prevalent in Tsavo than in Ishaqbini (Figure 4.5). The differences observed were comparatively greater for coccidia and least for strongyles. For the latter, the prevalence level in the two areas was almost equal

(67.72%, and 67.68% for Tsavo and Ishaqbini respectively). Apparently, the area of sampling did not have significant effect on prevalence of any of the gastrointestinal parasites (strongyles; $\chi^2 = 0.004$; df = 1, P = 0.9997; trematodes; $\chi^2 = 0.089$; df =1; P = 0.765 and coccidia; $\chi^2 = 0.437$; df = 1; P = 0.508).

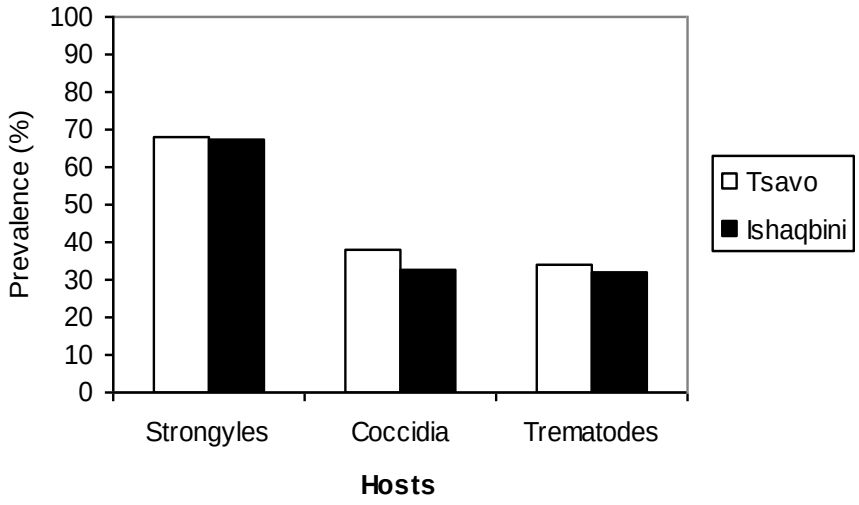

Figure 4.5. Overall comparisons for differences in prevalence of gastrointestinal parasites in study areas of Southern Kenya

Generally, strongyles were more prevalent among sheep and goats in Tsavo than in Ishaqbini (Figure 4.6a). However, with 72.55% (12) in Tsavo and 72.22% (43) in Ishaqbini, the discrepancy between the two areas was relatively small for goats. Among Hirola and cattle the prevalence of strongyles was higher in Ishaqbini than Tsavo. However, for all the species, the differences observed ere not statistically significant (Appendix Ia).

Figure 4.6b below depicts the prevalence patterns of coccidia among hosts in Tsavo and Ishaqbini. As shown, all hosts in Tsavo area had higher coccidia prevalence than those in Ishaqbini. The differences were more pronounced among cattle and least among Hirola. In case of cattle, the differences observed were statistically significant (χ^2 = 10.886; df = 1; P =0.001) whereas for sheep, goats and Hirola they were not, (Appendix Ib).

The highest number of samples that tested positive for trematodes belonged to sheep and cattle in Tsavo with 45.83% (4) and 42.06% (20) respectively (Figure 4.6c). In case of goats and Hirola in Ishaqbini, 23.89% (14) and 31.56% (16) respectively, were found to have higher positive cases of trematodes compared to their counterparts in Tsavo. The differences in prevalence between the two areas were however not statistically significant for any of the animal groups studied (Appendix Ic).

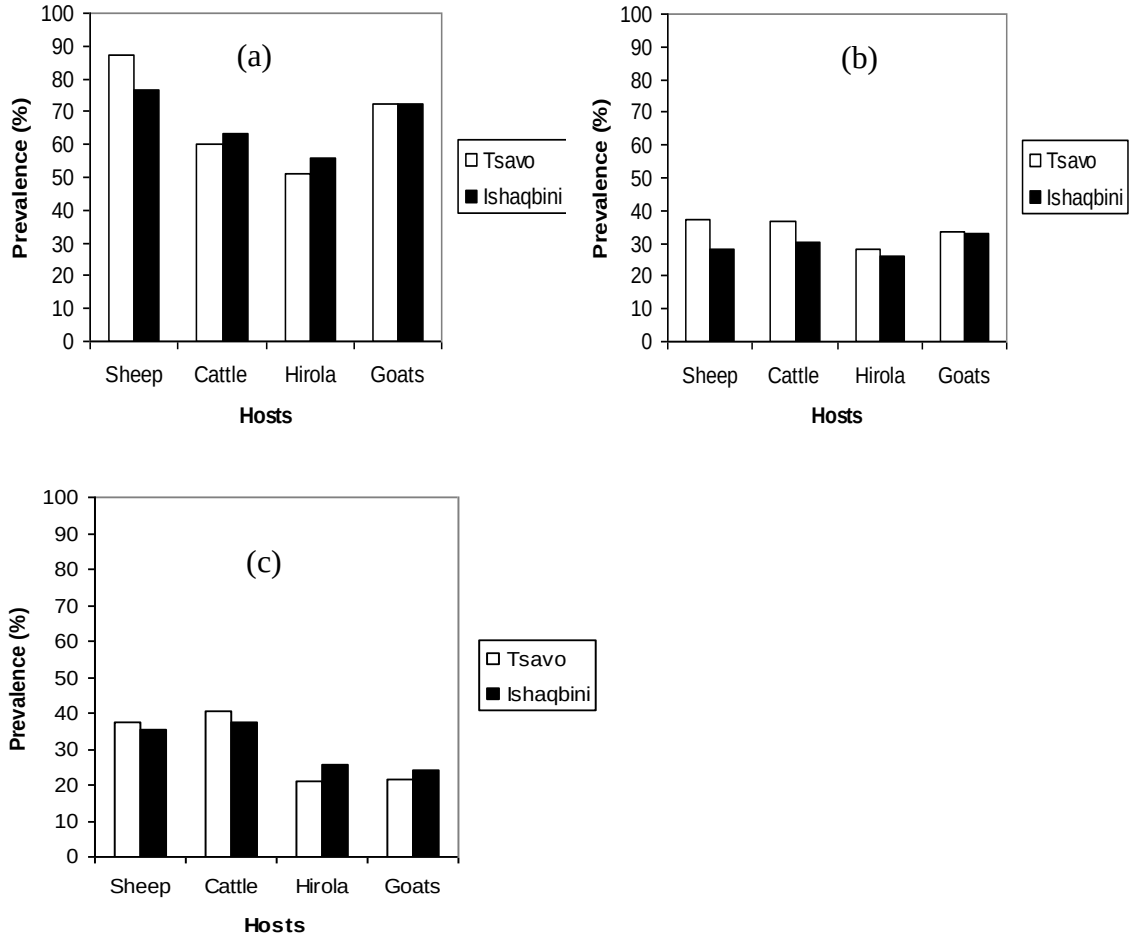

Figure 4.6. Prevalence of gastrointestinal parasites in Tsavo and Ishaqbini

(a) Strongyles, (b) Coccidia and (c) trematodes

Figure 4.7 shows the overall prevalence of gastrointestinal parasites based on seasons. As shown, all the three parasite groups were more prevalent during the wet season than during the dry season. The differences were greatest for coccidia and smallest for strongyles. In the case of coccidia, the differences observed were statistically significant

(χ^2 = 9.928; df = 1, P = 0.002) whereas for strongyles they were not (χ^2 = 1.203; df = 1, P = 0.273) and trematodes (χ^2 = 2.560; df = 1, P = 0.110) they were not.

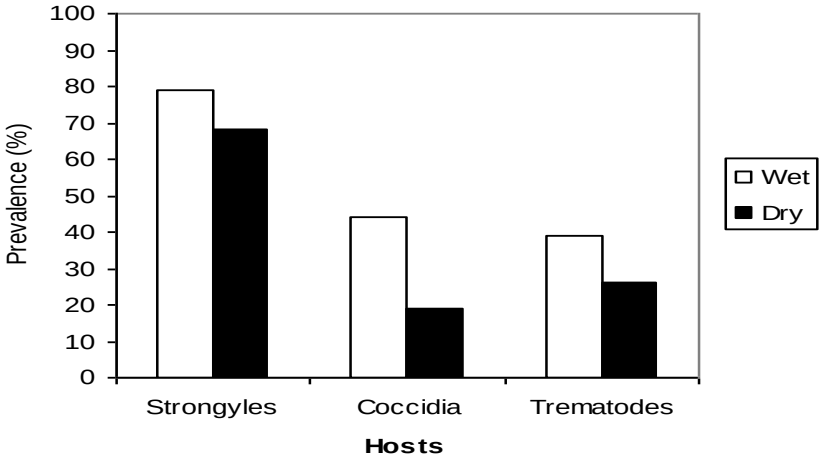

Figure 4.7. Overall comparisons for differences in prevalence of gastrointestinal parasites based on seasons

Among the four animal groups studied, season did not have significant effect on the prevalence of strongyles (Appendix Ie). However, it was observed that relatively more animals had been infected with strongyles during the wet season than during the dry season (Figure 4.8a). These differences were more pronounced in Hirola and goats and least in sheep.

The prevalence of coccidia was highest during wet season as compared the dry period (Figure 4.8b). The differences in prevalence between the wet and the dry periods were statistically significant for all animals (sheep, χ^2 = 15.327; df = 1, P = 0.001; cattle; χ^2 = 10.227; df = 1, P =0.001; Hirola; χ^2 = 3.840; df = 1, P = 0.050 and goats; χ^2 = 20.198; df = 1, P = 0.001).

More animals were infected with trematodes during the wet season than during the dry season (Figure 4.8c). The difference in prevalence of trematodes between the two periods was statistically significant for sheep (χ^2= 4.160; df = 1, P = 0.041) and Hirola (χ^2 = 4.811; df = 1, P= 0.028), whereas for cattle and goats they were not (Appendix Ig).

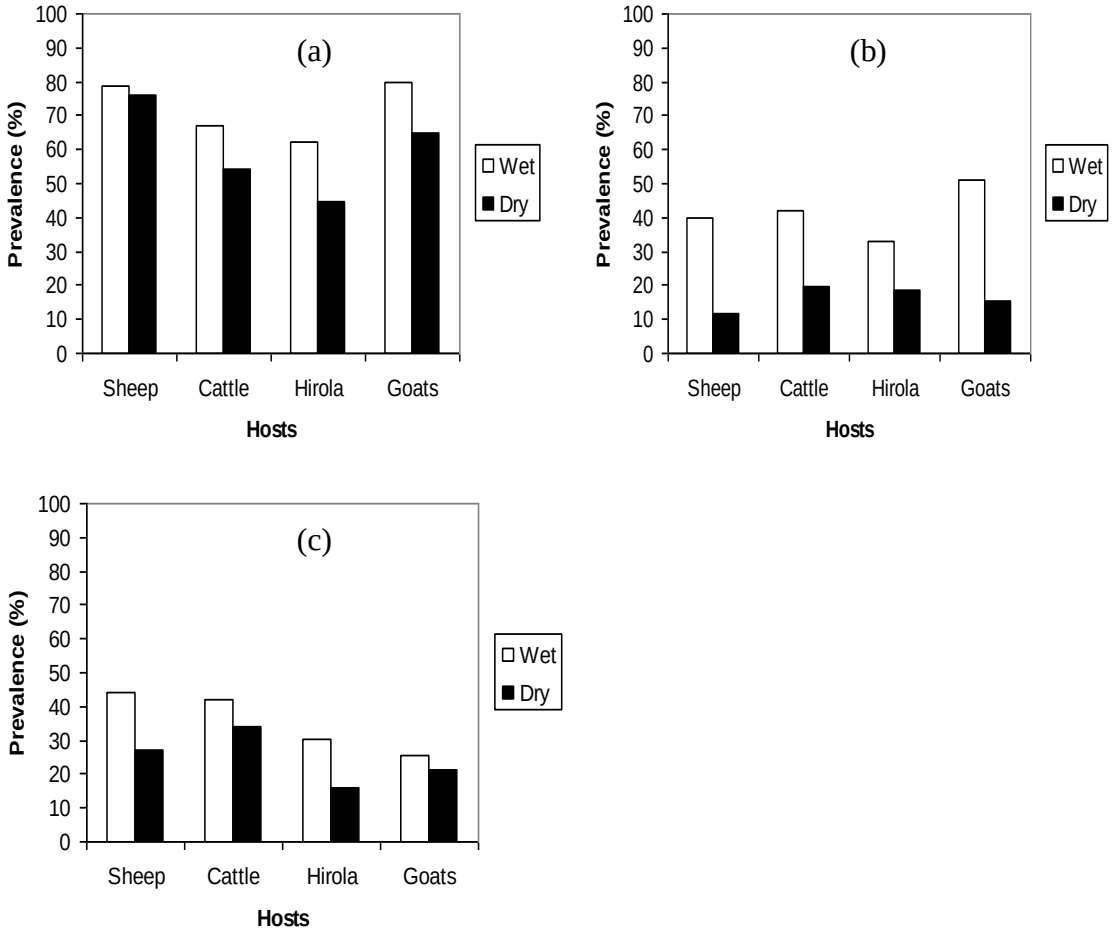

Figure 4.8. Prevalence of gastrointestinal parasites based on season

(a) strongyles, (b) coccidian and (c) trematodes

As shown by Figure 4.9 below, overall the gastrointestinal parasites generally were more prevalent in females than males. However, the differences were not statistically significant (strongyles; $\chi^2 = 0.023$; df = 1, P = 0.878; trematodes; $\chi^2 = 0.365$; df = 1, P = 0.546, coccidia $\chi^2 = 0.365$; df = 1, P=0.686).

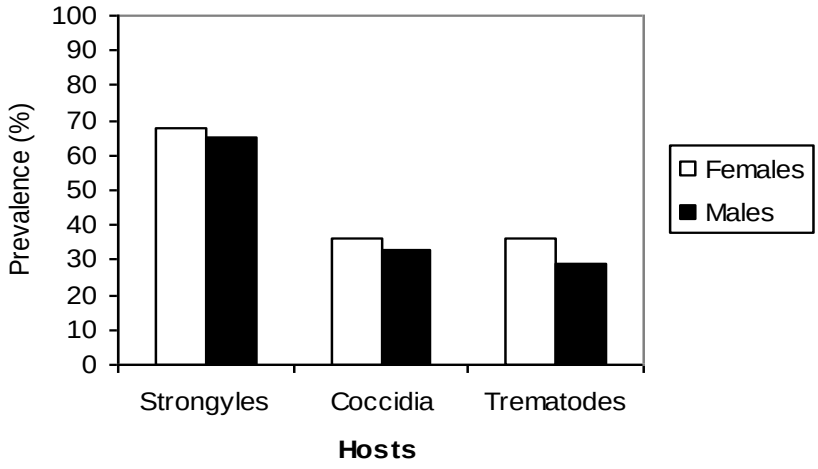

Figure 4.9. Overall comparisons for differences in prevalence of gastrointestinal parasites based on sex

4.1.4. Sex Variations in Prevalence of Gastrointestinal Parasites

After pooling all cases positive for strongyles species-wise, it was established that among sheep and goats, males had higher prevalence of strongyles than females (Figure 4.10a). However, in the case of sheep, 78.05% (32) in males and 77.42% (24) females, the difference was quite small. Contrarily to this, it was the females among Hirola (57.02%; 44) and cattle (70.59%, 36) that were found to have had higher prevalence of strongyles than males. In males, the prevalence was 51.06% (31) and 64.25% (44) in Hirola and cattle respectively. Tests for these differences showed that sex did not have a

significant influence on prevalence of strongyles in any of the animal groups (Appendix Ih).

As shown in Figure 4.10b analyses of samples positive for coccidia based on sex of the host revealed that apart from sheep, females had higher cases of infection than males. Among these, female sheep had the highest prevalence of 37.40% (12) while male Hirola had the lowest 23.83% (15). However, it was noted that these differences were not statistically significant for all the hosts studied (Appendix Ii).

Apart from sheep, females had comparatively higher cases of trematodes than males in all the hosts examined (figure 4.6c). The differences in prevalence based on sex of the host were not statistically significant in all the cases (Appendix Ij).

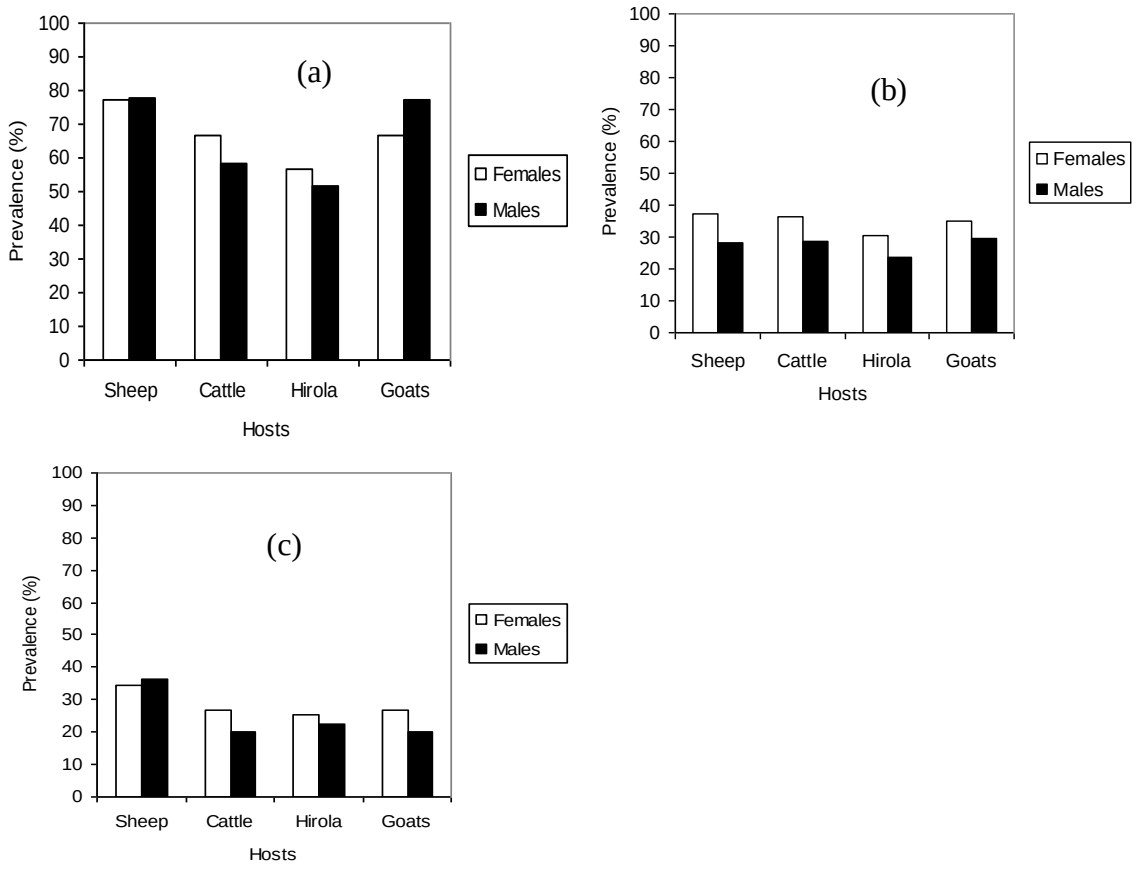

Figure 4.10. Prevalence of gastrointestinal parasites based on sex of host

(a) strongyles, (b) coccidian and (c) trematodes

4.1.5 Age Variations in Prevalence of Gastrointestinal Parasites

When all positive cases were pooled according to the age of the host, it was found that the parasites were more prevalent among the young animals than the adults. As depicted by figure 4.11 below, these differences were more pronounced in strongyles than in coccidia and trematodes. However, the differences showed were not statistically

significant (strongyles χ^2 =1.461; df = 1 P= 0.227; coccidia; χ^2 = 1.141; df = 1, P = 0.286 and trematodes; χ^2 = 0.544; df = 1, P = 0.461).

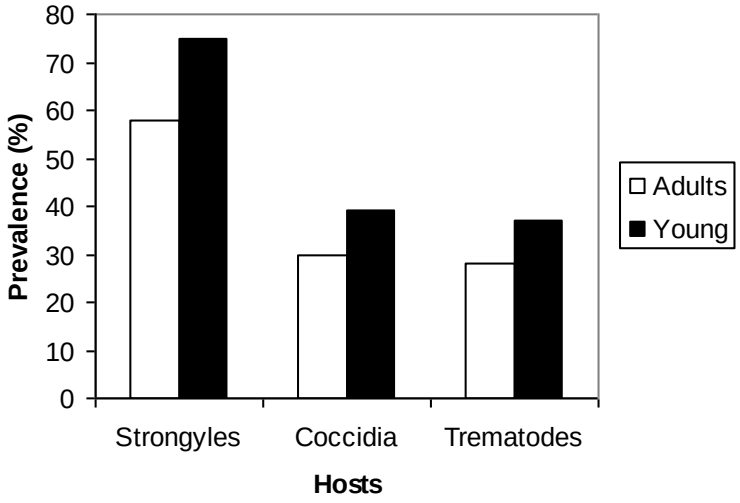

Figure 4.11. Overall comparisons for differences in prevalence of gastrointestinal parasites based on age of the hosts

Species specific differences in the prevalence of strongyles are shown in Figure 4.12a. As shown the differences were more pronounced among sheep and goats and least among cattle and Hirola. However, these differences were not significant in any of the hosts (Appendix Ik).

After pooling all samples positive for coccidia it was found out that more young animals were positive for coccidia than adults (Figure 4.12b). Among goats, the young were significantly more infected than the adults (χ^2=44.118; df =1, P = 0.020), whereas in sheep, cattle and Hirola, the differences in prevalence between adults and the young were not statistically significant (Appendix Il).

It was also found out that age did not have a significant effect on the prevalence of trematodes in the four hosts studied (Appendix Im).Apparently, among sheep, cattle and goats, there were higher number of positive cases in young animals than in adults (Figure 4.12c). In contrast, slightly more cases of the parasite were detected among the adult Hirola than the young.

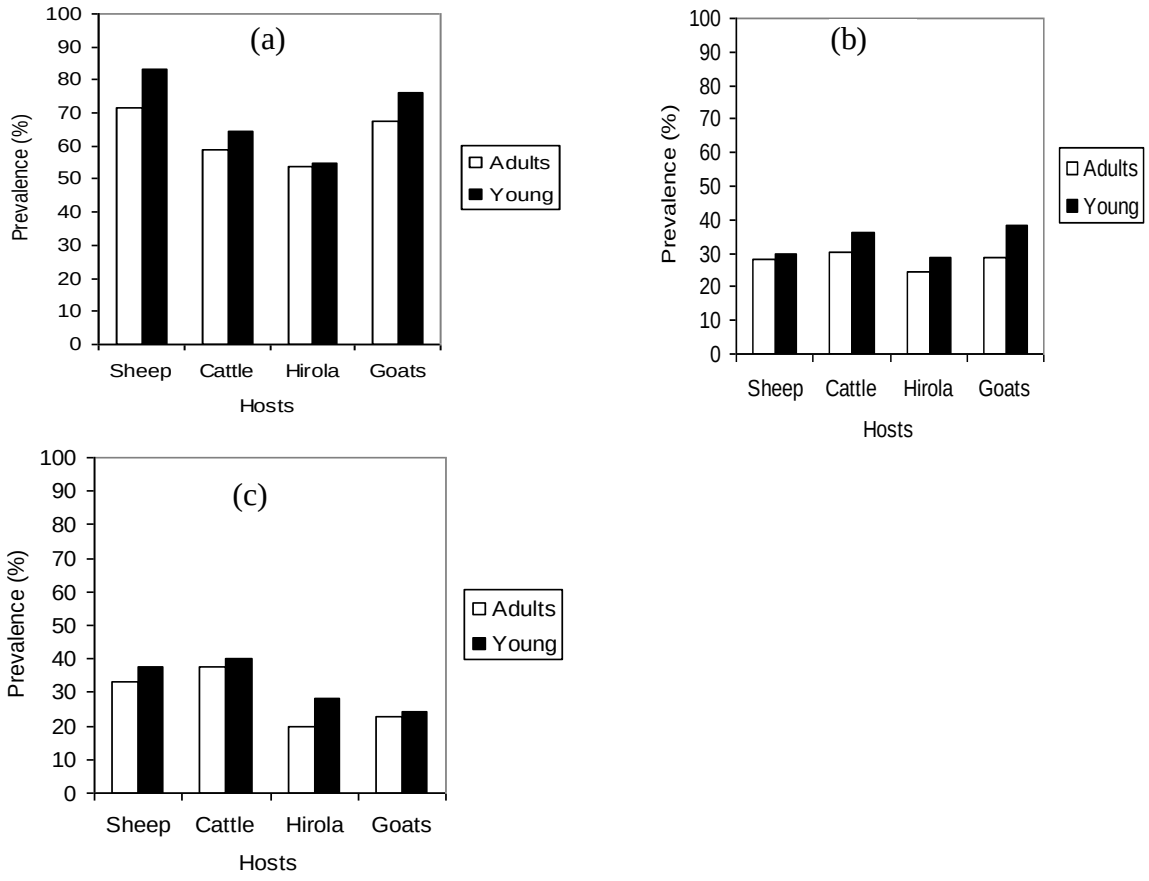

Figure 4.12. Prevalence of gastrointestinal parasites based on the age of hosts

(a) strongyles, (b) coccidian and (c) trematodes

4.2 Intensities of Gastrointestinal Parasites

4.2.1 Overall Counts for Eggs and Oocysts

Based on faecal egg counts (FEC), the mean eggs per gram (EPG) differed significantly across the hosts (ANOVA, $F_{3, 407}$= 9.376; P = 0.001). This is shown in Figure 3.13a in which eggsare represented as geometric faecal egg counts (GFEC). Goats had the highest mean eggs per gram (with 386.75 ± 43.55 SE) whereas Hirola (with 262.24 ± 33.42 SE) had the lowest. Also based on faecal oocysts counts (FOC), it was observed that the mean oocyts per gram (OPG) also differed significantly across the hosts (One - Way ANOVA; $F_{3, 407}$ = 5.91; P = 0.000). These results are depicted in Figure 4.13b as geometric faecal oocysts counts (GFOC). It was noted that just like in the case of strongyles, goats recorded the highest mean oocysts per gram (i.e. with 537.78 ± 135.59 SE) and Hirola the lowest (259.08 ± 63.47 SE).

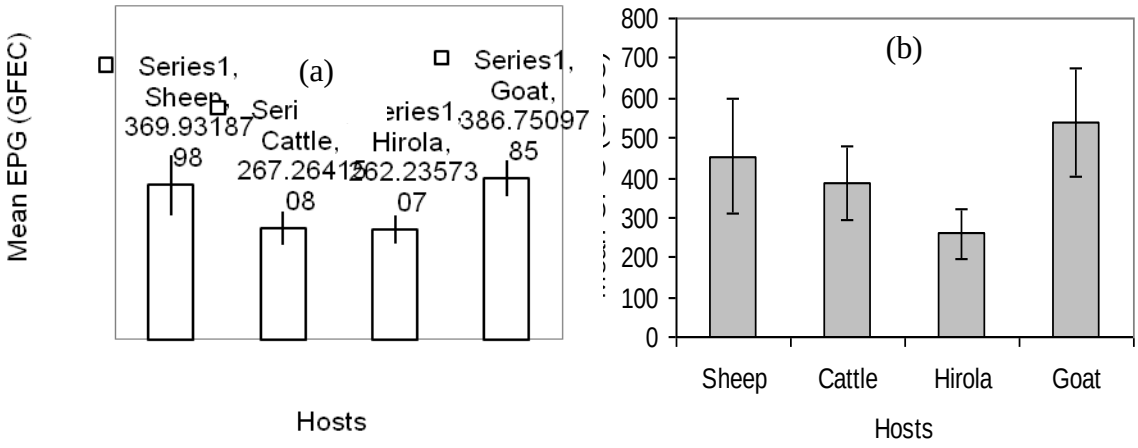

Figure 4.13. Overall mean Eggs per Gram and Oocysts per Gram ±SE

4.2.2 Variations in Intensity of Gastrointestinal Parasites based on Study Site

Overall, animals in Tsavo East National Park and its environs recorded a higher egg and oocyst counts per gram of faeces compared to those in Ishaqbini Hirola Conservancy (Figure 4.14a and 4.14b). It was observed that the mean eggs per gram obtained from faecal egg counts (FEC) in the two study areas were significantly different (One - Way ANOVA; $F_{1, 409} = 8.632$; $P = 0.003$). However, in the case of faecal oocysts counts, the differences in the mean oocysts per gram were not statistically significant ($F_{1, 409} = 0.542$; $P = 0.462$).

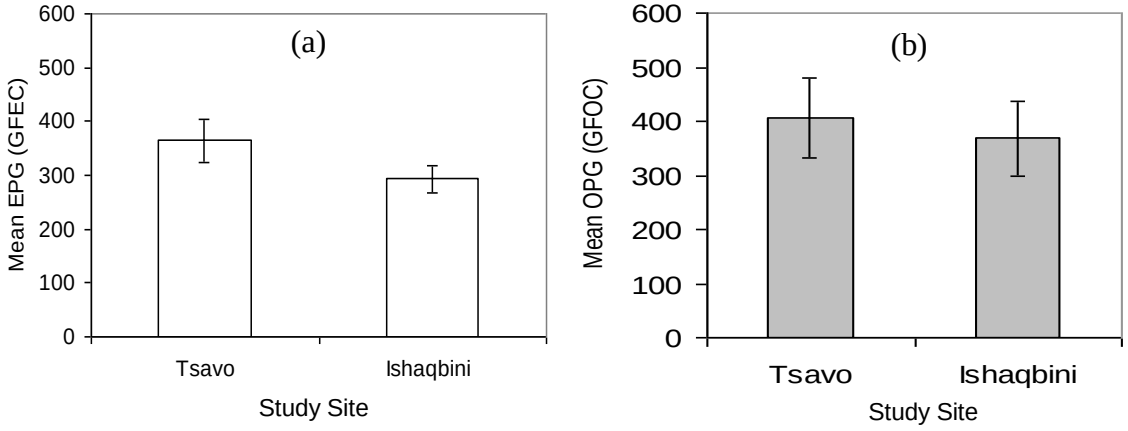

Figure 4.14. Comparisons for overall eggs and oocysts outputs by gastrointestinal parasites based on study site

Results for the analysis of faecal egg counts data based on site showed that apart from Hirola, the hosts had higher mean eggs per gram in Tsavo than their counterparts in Ishaqbini (Figure 4.15). In cattle, these differences were statistically significant ($F_{1, 119} = 14.584$; $P = 0.001$) whereas for the rest of the hosts they were not (Appendix IIa). The

highest mean EPG was observed among goats in Tsavo with 456.71±76.23 SE whereas the lowest EPG value was observed among Hirola in Tsavo (250.60±53.06 SE).

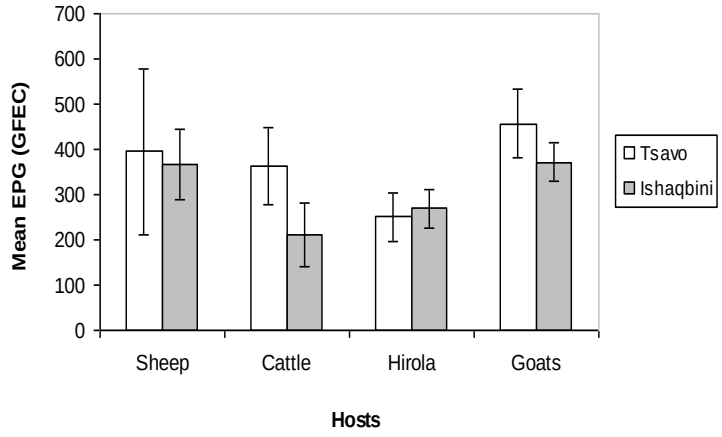

Figure 4.15. Mean Eggs per Gram (EPG) ± SE based on study site

Figure 4.16 below shows patterns of oocyst output per gram of faeces analyzed according to site of study. Except Hirola, all hosts in Tsavo had a higher oocysts output than their counterparts in Ishaqbini. These differences were more pronounced among cattle and least among sheep. The differences were statistically significant for cattle ($F_{1, 119}$= 5.420; P = 0.020) but not for sheep, Hirola and goats (Appendix IIb).

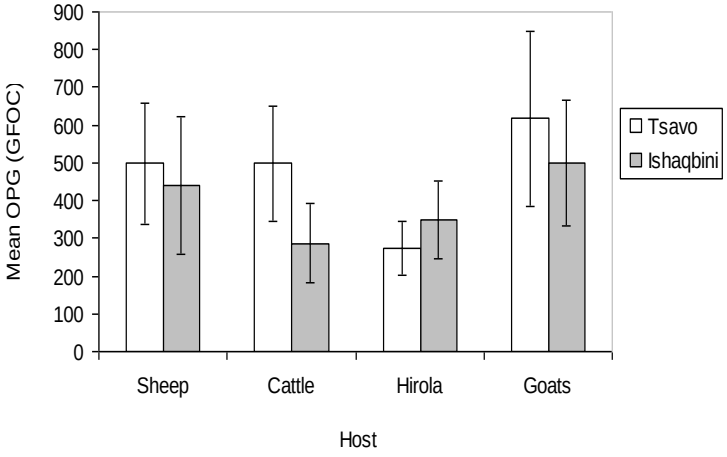

Figure 4.16. Mean Oocysts per Gram (OPG) ± SE based on study site

4.2.3 Seasonal Variations in Intensity of Gastrointestinal Parasites

When all the data on faecal eggs and oocysts counts was pooled according to season, it was observed that the intensity of gastrointestinal parasites was higher during the wet than in the dry season (Figure 4.17.b and Figure 4.17a).The differences in mean EPG and mean OPG counts between wet and dry season were statistically significant in both cases (EPG; $F_{1, 409}= 23.36$; $P = 0.001$; OPG; $F_{1, 409}= 3.29$; $P = 0.047$)

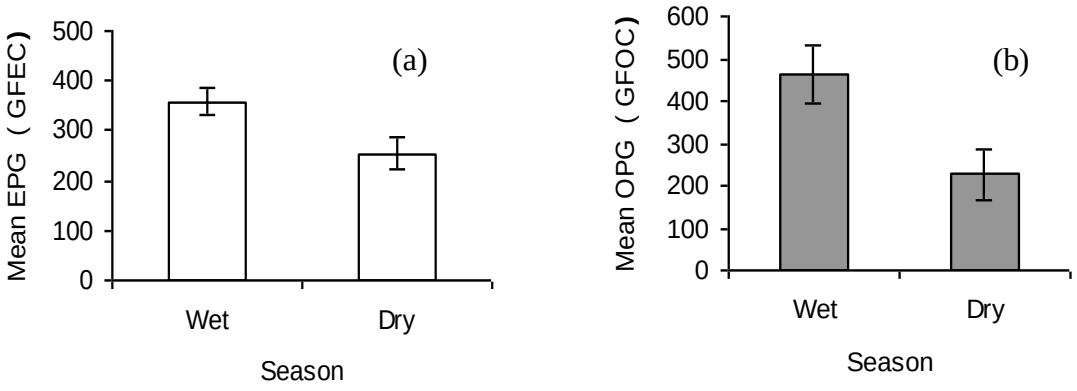

Figure 4.17. Comparisons for overall eggs and oocysts outputs by gastrointestinal parasites based on study site

The effect of season on faecal egg counts (FEC) is depicted by Figure 4.18a. For all hosts, higher FEC was recorded during the wet season than in the dry period. Differences were statistically significant for sheep ($F_{1, 71}= 9.36$; $P = 0.004$); cattle ($F_{1, 119}= 12.68$; $p = 0.000$) and Hirola ($F_{1, 76}= 9.23$; $P = 0.003$) but not for goats ($F_{1,140} = 3.08$; $P = 0.082$) (Appendix IIc).

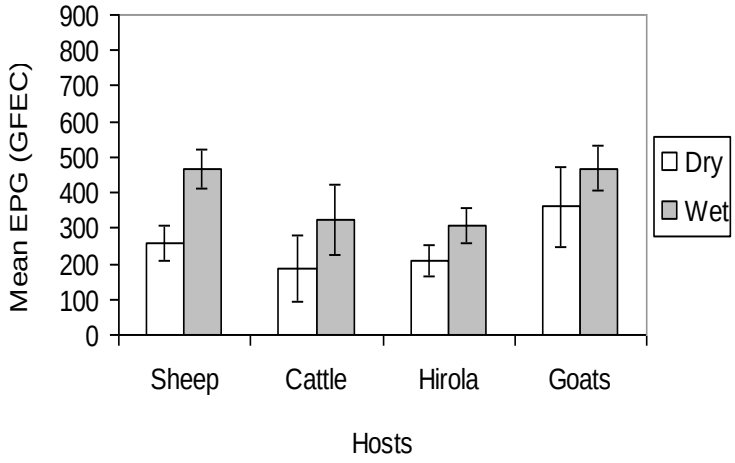

Figure 4.18. Mean Eggs per Gram (EPG) ± SE based on sex of host

The intensity of infection of coccidia was greater during the wet season than during the dry period (Figure 4.19) for all the hosts. The effect of season on coccidia intensity was higher in Hirola and cattle than it was in sheep and goats. For cattle and Hirola the differences were statistically significant ($F_{1, 119}$= 5.972; P = 0.016, and $F_{1, 140}$= 25.82; P =0.001 respectively) whereas for sheep and goats they were not (Appendix IId).

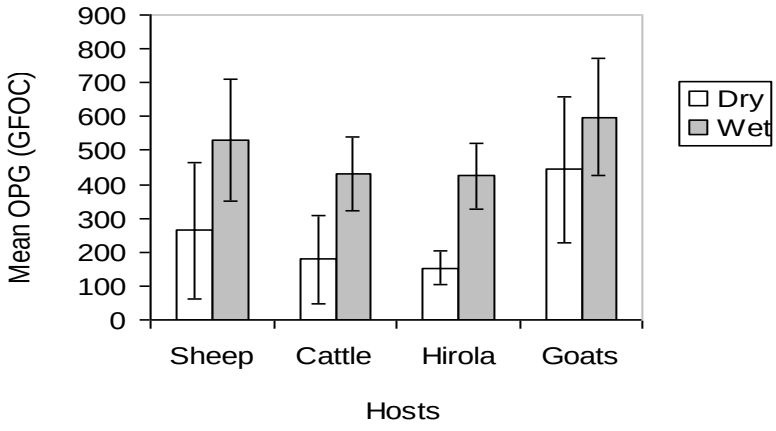

Figure 4.19. Seasonal Mean Oocyst Per Gram ± SE

4.2.4 Intensity of gastrointestinal parasites based on Sex of Host

After pooling all the data on faecal egg and oocysts counts on the basis of sex of the hosts, it was observed that only subtle differences existed between levels of intensities in the males and females (Figures 4.20a and 4.20b). In both strongyles and coccidia, the males recorded slightly higher intensities of the parasites than females. These differences were not statistically significant (strongyles, $F_{1,\ 409}$ = 0.010; P = 0.921; coccidia; $F_{1,\ 409}$= 0.04; P = 0.848).

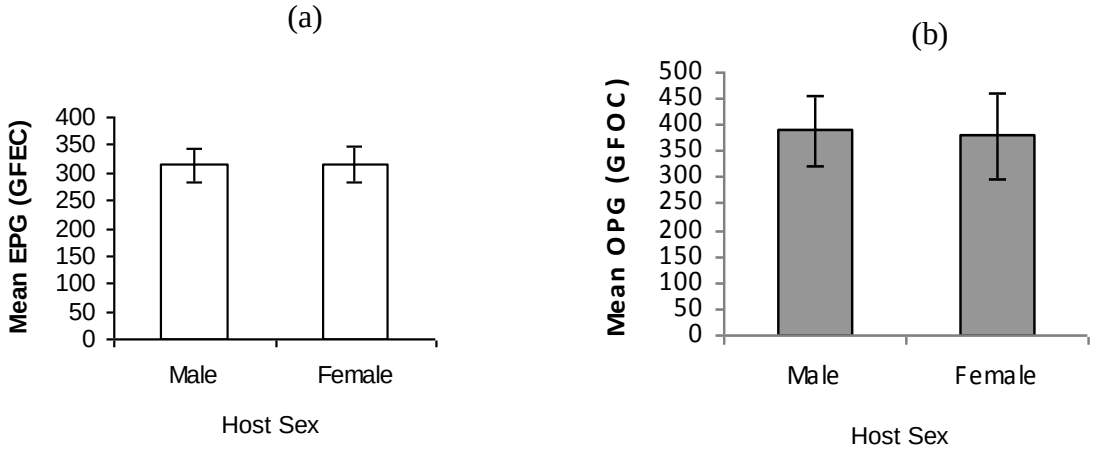

Figure 4.20. Comparisons for overall eggs and oocysts outputs by gastrointestinal parasites based on sex of hosts

Figure 4.21 below shows the patterns of strongyle intensities for each host on the bases of sex. Except in cattle, males recorded higher mean EPG than females. In cattle, the females had significantly higher EPG output than males ($F_{1,\ 119}$= 9.946; P = 0.002). In sheep, Hirola and goats, the differences in mean EPG between males and females was not statistically significant (Appendix IIe).

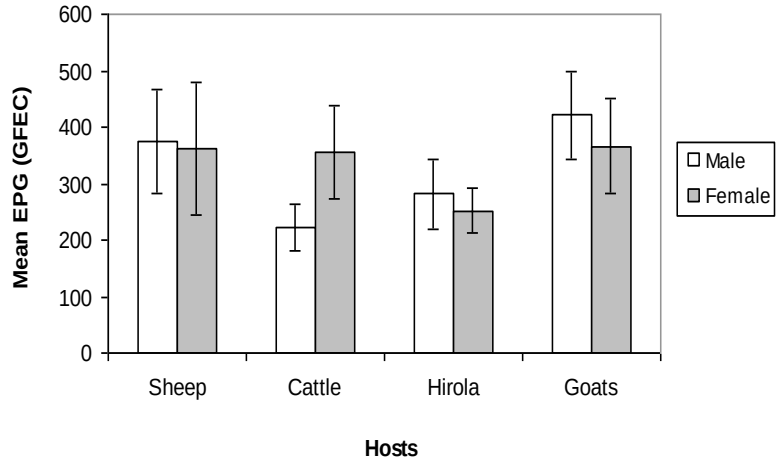

Figure 4.21. Mean Eggs per Gram (EPG) ± SE based on sex of hosts

Comparisons of FOC showed that female sheep and Hirola had higher mean OPG than males (Figure4.22). Female sheep had mean OPG value of 598.29±280.98SE while males had 405.87±164.46SE and among Hirola, females had 352.85±109.53SE while males had 285.85±74.14SE. Male cattle and goats were observed to have had higher OPG than females. In this case the differences were more pronounced in goats than in cattle. However, in none of the hosts were the differences statistically significant (Appendix IIf).

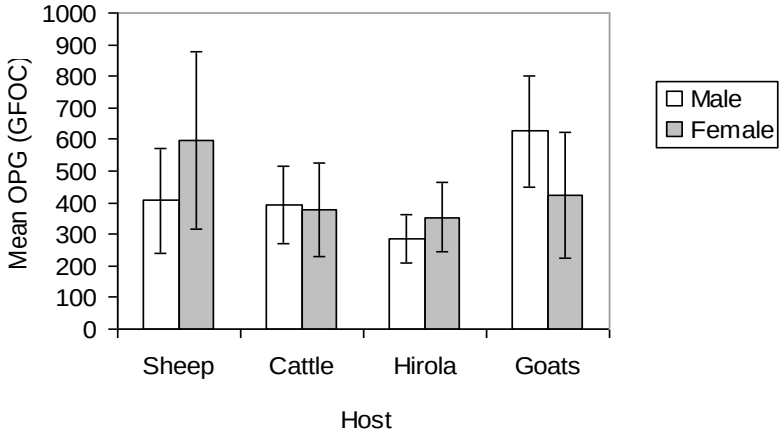

Figure 4.22. Mean Oocysts per Gram (OPG) ± SE based on season

4.2.4 Intensity of gastrointestinal parasites based on Sex of Host

In general, young hosts had higher intensities of gastrointestinal parasites than adults. These differences were more pronounced for coccidia (Figure 3.23a than strongyles Figure 3.23b). However age did not have significant influence in the intensities of the parasites (strongyles; $F_{1, 409}$= 0.010; P = 0.921; coccidia; $F_{1, 409}$ = 0.036; P = 0.848).

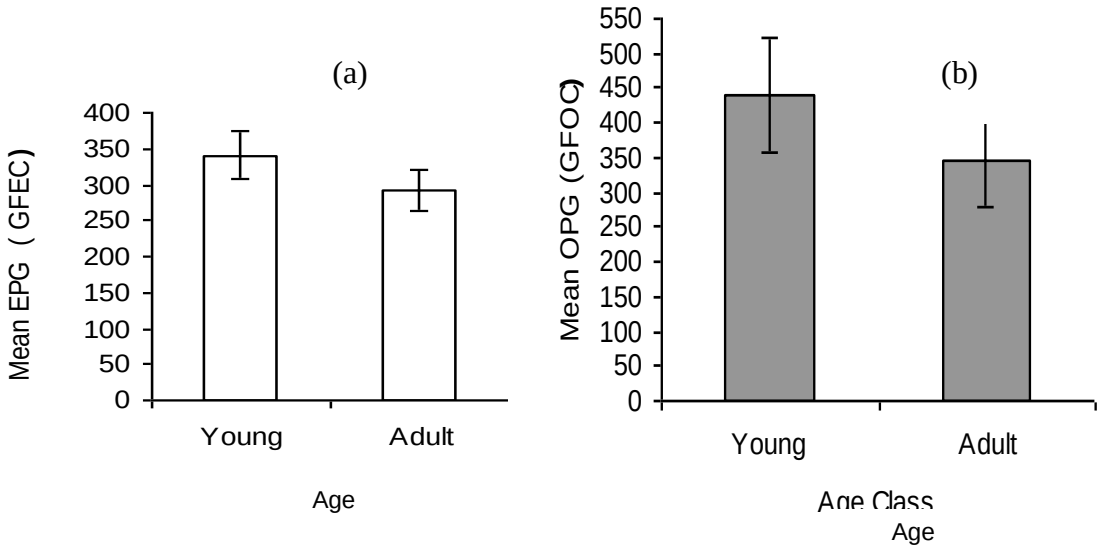

Figure 4.23. Comparisons for overall eggs and oocysts outputs by gastrointestinal parasites based on age of hosts

Except for cattle, the young had lower intensities of strongyles than adults (Figure 4.24). EPG differences in sheep were statistically significant ($F_{1, 71}$= 9.243; P = 0.003) but not in cattle, Hirola and goats (Appendix 2g).

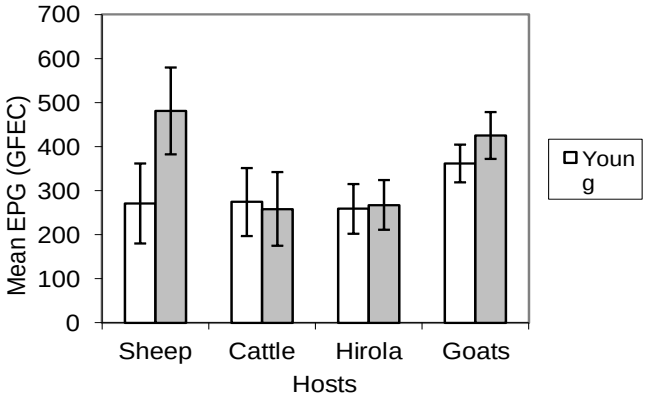

Figure 4.24. Mean Eggs per Gram (EPG) ± SE based on Age of Hosts

As indicated in the Figure 4.24, sheep and Hirola had similar patterns of FOC with adults recording higher mean OPG values than the young. For Hirola, the differences noted were statistically significant ($F_{1, 140}$= 5.183; P = 0.017) whereas for sheep, cattle and goats they were not (Appendix IIh).

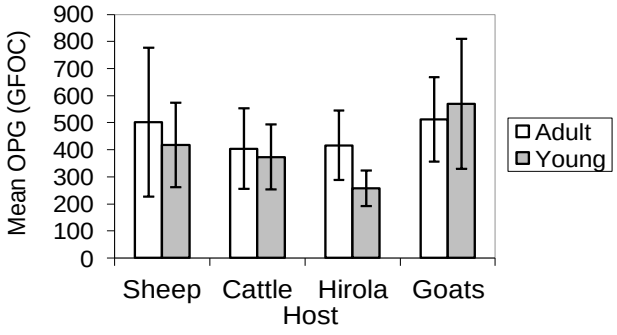

Figure 4.25. Mean Eggs per Gram (OPG) ± SE based on Age of Hosts

General Linear Models for the analysis of fixed effects of study site, season and age and sex of the host on EPG and OPG output are summarized in Appendices III and IV.

4.3 Aggregation of Parasites in the Host Populations

4.3.1 Frequency Distribution of Strongyles

Results for frequency distribution of nematodes in the hosts are represented by Figure 4.26. In all the four animal groups the distribution followed the negative binomial distribution. Very low values of *k* (<1) were obtained. This showed that strongyles were highly aggregated among the host populations. With a *k* value of 0.009, the highest level of aggregation was observed in cattle, whereas the least level of aggregation was observed in sheep and Hirola (*k = 0.064)*. Chi-square test for the differences in aggregation for the four hosts showed that they were not statistically significant (χ^2= 0.058; df = 3, P= 0.867).

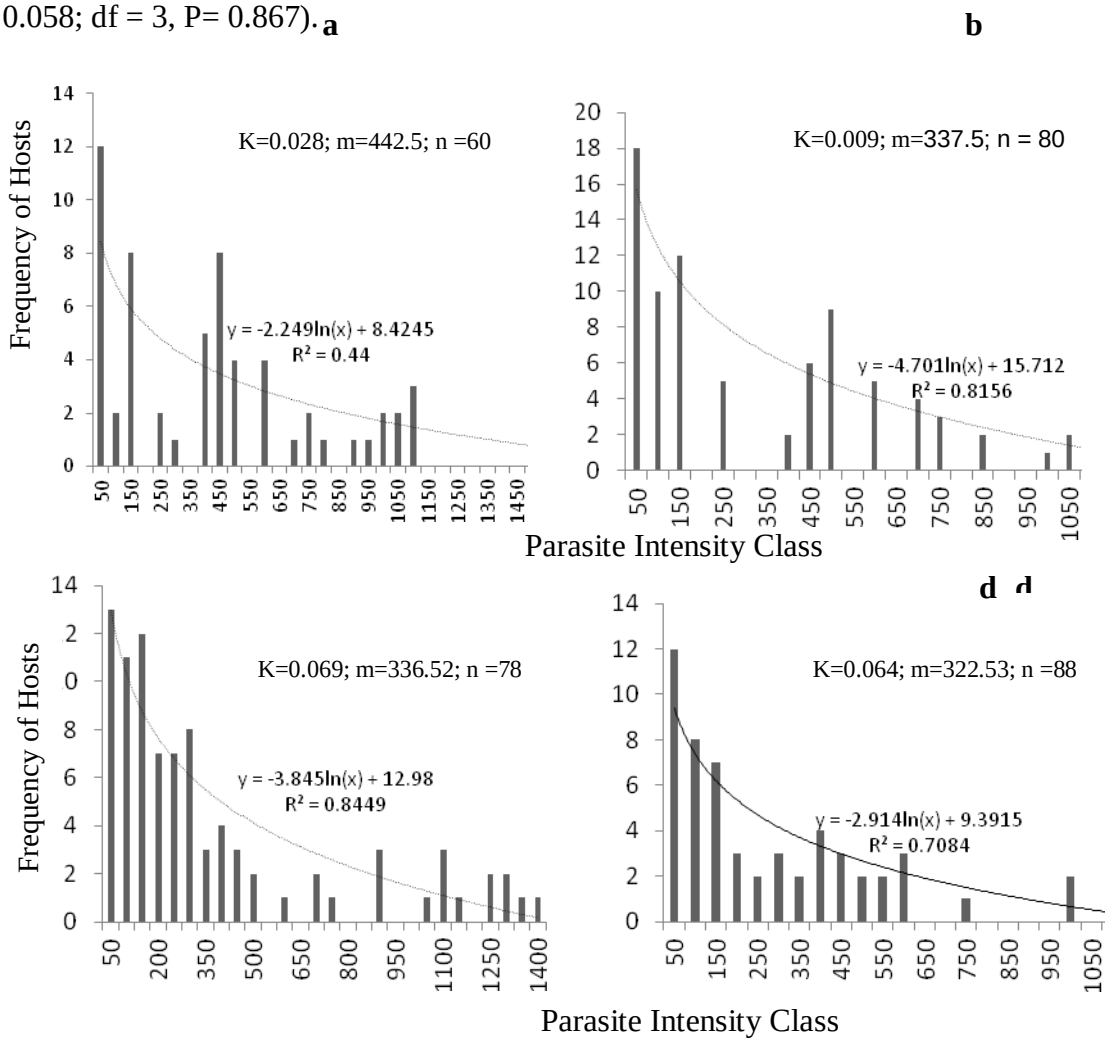

Figure 4.26. Frequency of distribution of strongyles in the hosts
(a) Sheep (b) Cattle (c) Hirola and (d) Goats

4.3.2 Frequency Distribution of Coccidia

Figure 4.27 shows patterns of aggregation of coccidia in the host population. The highest level of aggregation was recorded from cattle (k =0.003), while the lowest was recorded from goats (k = 0.18). However, the differences in aggregation between the hosts were not statistically significant (χ^2 = 0.523; df = 3, P = 0.756).

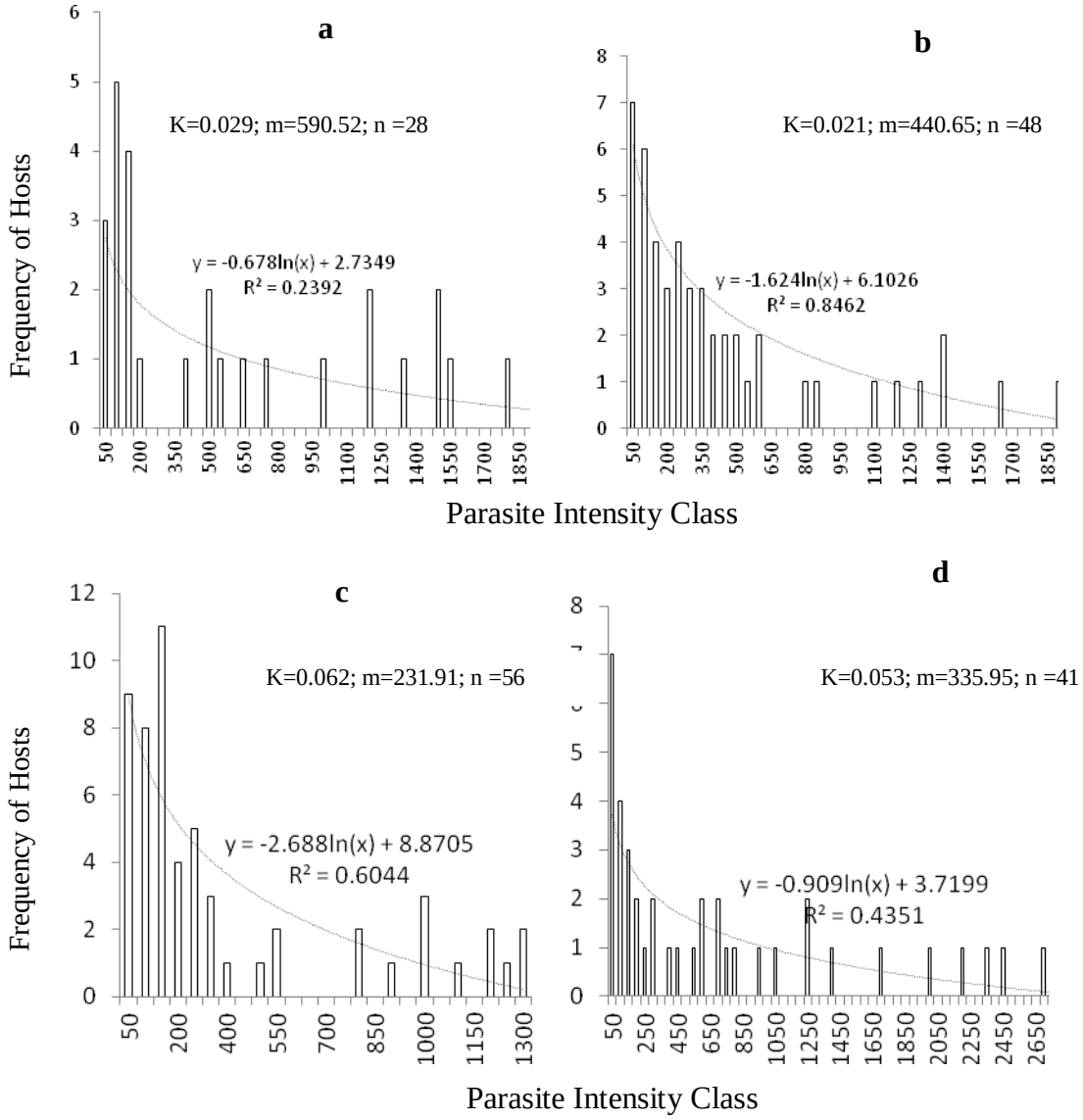

Figure 4.27. Frequency of distribution of coccida in the hosts
(a) Sheep (b) Cattle (c) Hirola and (d) Goats

CHAPTER FIVE

DISCUSSION

5.1 Prevalence of Gastrointestinal Parasite Infections

5.1.1 Overall prevalence

Results on gastrointestinal parasites revealed that both Hirola and livestock were infected by a wide variety of gastrointestinal parasites with strongyles being the most prevalent parasite group.

In the tropics, strongyles have been shown to be the most common and economically important gastrointestinal nematodes (Agyei, 1997; Odoi *et al.,* 2007). The genera *Haemonchus, Trichostrongylus, Cooperia* and *Oesophagostomum* all belong to this group and are the most pathogenic gastrointestinal parasites to small ruminants. In Kenya these parasites have been reported widely in livestock (Ng'ang'a *et al.,* 2004; Odoi *et al.,* 2007) and in wild bovids (Ezenwa, 2003a; Ezenwa 2004a).

In this study, *Trichuris spp* was recovered from goats alone, but this does not discount the possibility that this parasite could be shared between groups. In other reports (Soulsby 1982) *Trichuris spp* was shown to have a wide host range in both livestock and wildlife, and are considered to be harmless except in very heavy infections such as is the case where there isa large soil intake by grazing animals during times of drought, in which case, there may be a sub-acute typhlocolitis, diarrhoea and ill thrift (Love and Hutchinson, 2003). *Toxocara* (*Neoascaris*) *vitulorum* identified in this study is a

nematode of cattle but it has also been reported widely in buffaloes (Souza *et al*., 2004; Raza *et al*., 2010).

Fluke infestations have been associated with areas with high rainfall and poorly drained soils and it was surprising to find flukes in the study areas especially, in Ishaqbini Hirola Conservancy. Nevertheless, it could be postulated that in the current study, the occurrence of flooding, especially in November-December and January at the time of sampling could have provided suitable habitats for propagation of the parasite intermediate hosts, the snail (*Lymnea spp*). A study by Shalaby *et al.,* (2011) on livestock from Somalia also gives evidence of prevalence of fascioliasis in animals from this agro-ecological zone. These parasites are known to cause anaemia, hypoproteinemia (manifested as submandibular oedema) and emaciation of the host (Love and Hutchinson, 2003).

The only cestode observed in this study was *Monieza spp.* Andit was recovered from one goatin Tsavo East National Park area. *Monieza spp*is generally regarded as relatively harmless (Love and Hutchinson, 2003). Previously, it has been reported from goats, sheep and cattle elsewhere (Kanyari *et al*, 2009) and several wild ruminants in Kenya and elsewhere in the tropics (Xiao and Herd, 1992; Ezenwa, 2003a; Ezenwa, 2004b; Apio *et al.*, 2006; Sissay *et al.*, 2008) and it appears to have a cosmopolitan distribution in these animals.

Intestinal protozoa such as the coccidia observed in this study can have a broad host spectrum (euryxenous), but *Eimeria* tend to specialize on a single species (Turner and Getz, 2010). Presently, they were recorded from both Hirola and livestock. These gastrointestinal parasites are common in livestock (Kanyari, 1993; Harper and Penhorn, 1999, Kanyari *et al.,* 2009) and have also been reported from wild ungulates (Ezenwa, 2003a; Ezenwa, 2003b, Ezenwa, 2004b; Apio *et al.,* 2006). *Eimeria* species causes coccidiosis (parasitic enteritis of small and large intestines) usually in younger animals (Love and Hutchinson, 2003). The disease is exacerbated by various stressors and other pathogens suchas viruses, bacteria and helminthes.

Overall, 79.75% (327) of the samples were positive for at least one of the groups of gastrointestinal parasites. Out of these, 45.37% (186) were multiple infections while 34% (139) were single infections. In natural host-parasites interactions, multiple infections are commonplace (Read and Taylor, 2001) and numerous studies show that they are an important determinant of virulence evolution (Taylor *et al.,* 1998; Davies *et al.,* 2002; Massey *et al;* 2004; Hôrak *et al.,* 2006). Indeed, Lo´ pez-Villavicencio *et al.,* (2010) asserted that the optimal virulence of a parasite under multiple infections was often different from that under conditions of single infection. Studies have also shown that when parasites of different genotypes or strains compete for limiting resources within a host, virulence per genotype is predicted to increase with lethal effects (van Baalen and Sabelis, 1995; Frank, 1996; Brown *et al.,* 2002).

In this study goats were observed to have the highest prevalence of multiple infections with 55.84 % (40), while Hirola had the lowest, 40.43% (57). Going by this

observation, it would mean that the threat posed by multiple infections in Hirola was lower than it was the case in livestock. However, it was difficult to predict immediately the implications of these infection patterns in Hirola based on the available data. Furthermore, some authors have reported no effect or even lower virulence in multiple infections (Hood, 2003; Hughes *et al*., 2004) often when the competition is such that only one strain wins.

The results from this study also indicated that the differences observed in overall prevalence of gastrointestinal parasites in the different hosts were not statistically significant. This strongly suggested co-infection with the parasites among the study species. Overall, goats had the highest prevalence (88.89%; 64). Similar observations were made by Kanyari *et al.,* (2009) and Ghanem *et al.,* (2009) in studies of gastrointestinal prevalence in livestock. However, the result contradicted observations made by Wairuri *et al*., (1995) in which sheep had a higher prevalence than goats. The scenerio in which goats had such high cases of infections could be due to slow development of immunity against gastrointestinal parasites.

Cattle and sheep are believed to have faced prolonged challenge by parasites over generations, but in goats, the decline of sufficient browsing area and expansion of crop husbandry has forced them to graze alongside cattle and sheep that had already developed good resistance (Regassa *et al.,* 2006). Perhaps the most plausible explanation for the high prevalence in sheep (84.42%; 65), is their feeding habit. Sheep tend to graze very close to the ground and this ultimately predisposes them to high chances of picking the infective larval stages.

Among the four hosts, Hirola had the lowest prevalence of gastrointestinal parasites. Like sheep and cattle, Hirola are grazers suggesting that they are potentially exposed to the infective larval stages in contaminated forage. Since species dependent factors result in certain species having higher prevalence levels than others, it is likely that Hirola have a higher genetic resistance to the parasites than livestock, or employs better feeding strategies than livestock, thereby avoid feeding on contaminated plants. Also feeding on plants containing high levels of anthelmitic substances such as tannins, perhaps not taken by livestock could lead to such low prevalence levels. In addition, given that Hirola and livestock have for decades shared the same environment (Burndeson, 1985), and that co-infection with the parasites between Hirola and livestock exists, then it is likely that Hirola have acquired better immunity against the parasites studied as compared to livestock. However, such a conclusion would warrant further investigation.

5.1.2 Spatial and Temporal Variations in the Prevalence of Gastrointestinal Parasites in Different Hosts

Generally all the parasites were more prevalent in hosts in Tsavo than in their counterparts in Ishaqbini, but the differences were not statistically significant. Prevalence of gastrointestinal parasites has been shown to vary considerably due to differences in environmental conditions such as humidity, temperature, rainfall and management practices (Magona and Musisi, 2002; Regassa *et al.*, 2006). In Tsavo National park, the amount of rainfall, humidity, temperature and elevation are on

average slightly higher than in Ishaqbini and the rest of the natural range of Hirola (Butynski, 1999). These differences could possibly have accounted for the higher prevalence of the parasites in Tsavo. Furthermore any slight variation in the environmental conditions can have significant effect on survival and development of free living stages of the parasite. For example according to Gupta *et al*., (1985) and Michalski *et al.,* (1990) infestation of paramphistomiasis can vary from 0.70% to 88.89 from place to place.

However, it was interesting to note that on the basis of locality, the parasites exhibited different prevalence patterns across the hosts from what was observed for the overall prevalence. For instance, both Hirola and cattle had slightly more cases of strongyles in Ishaqbini than in Tsavo while Hirola and goats had slightly more cases of trematodes in Ishaqbini than in Tsavo. In case of cattle, the most plausible explanation for this unexpected pattern was the difference in management practices. There were occasions when cattle from neighbouring ranches were encountered and sampled alongside other livestock found within areas utilized by Hirola. Most of those cattle tested negative for nematodes leading to the observed low prevalence. It is likely that this was due to routine use of antihelmintics and the good plane of nutrition that they had been subjected to. In the case of goats and Hirola, the cause for the unexpected pattern could not be immediately ascertained.

It was found that during the wet season more animals were infected by gastrointestinal parasites than during the dry season. These differences were statistically significant for

strongyles and coccidia but not for trematodes. Moisture is known to be a major factor enhancing the development, survival and transmission of infective stages of gastrointestinal parasites (Dunn, 1978; Armour, 1980), and hence the direct relationship between parasite prevalence with humidity and temperature.

5.1.3 Prevalence Patterns Based on Sex and Age of Hosts

The study revealed that sex of the host did not have significant effect on the prevalence of the parasites in livestock and Hirola. Overall, females appeared to have more cases of parasites than males. However after analyzing the prevalence patterns of each parasite based on sex of each host, the findings revealed a variation in patterns of infections from one host to the other. In the case of strongyles, female Hirola and cattle had slightly more cases than males while in sheep and goats, males were more infected than females. For coccidia, all the female hosts except cattle had a slightly higher prevalence than males. These results are consistent with reports from several authors (Morgan *et al.,* 2005; Raza *et al.,* 2007; Tariq *et al.,* 2008) which show that although sex played a significant role in the preponderance of infection, environmental and climatic conditions had an even greater role to play. The influence of sex on susceptibility of animals to infections could be attributed to genetic predisposition and differential susceptibility owing to hormonal control and also sex-related variation in behaviour that resulted in exposure (Tariq *et al*., 2008).

Apparently, young Hirola and livestock had more cases of gastrointestinal parasitoses than adults. In the case of coccidia the differences were statistically significant apart from coccidia infections in goats. This observation is in consent with previous studies in

Kenya, Ethiopia and Somalia (Ng'ang'a, *et al.,* 2004; Githigia, *et al.,* 2005; Regassa, *et al.,* 2006; Ghanem, *et al.,* 2009). According to Shah-Fischer and Say (1989), this condition could be due to the fact that younger animals were more susceptible than adult counterparts. Adult animals may acquire immunity through frequent challenge and then expel the ingested parasites before they establish infection.

5.2 Intensity of Infection of Gastrointestinal Parasites in the Host Animals

5.2.1 Overall Intensity of infection

Evaluation of the overall intensity of strongyle parasite infections showed that the mean number of eggs per gram of faeces differed significantly across the hosts. The geometric means for faecal egg counts (FEC) ranged from 262.24 ± 33.42SE in Hirola to 386.75 ± 43.55 SE in goats. Generally, these values were low indicating light levels of infection.

According to Hansen and Perry (1994), the degree of infection is considered light if the EPG values are between 600 and 800 in livestock. However due to the potential of rapid build-up of worms on pasture, Torres-Acosta and Hoste (2008) suggested that deworming of animals was necessary when the average FEC was between 100 and 200 eggs per gram. The mean oocysts per gram (OPG) of faecal material varied significantly across the hosts. The geometric faecal oocysts counts GFOC values ranged from 259.08 ± 63.47 SE in Hirola to 537.78 ± 135.59 SE in goats. This meant that the overall intensity of infection with these parasites was light in livestock as well as in Hirola.

In general, the overall intensity of infection in livestock was low compared to what had been previously reported by Maichomo *et al.*,(2004) but agreed with a report by Ghanem *et al*., (2009) on livestock from Somalia. In the case of Hirola the significantly lower intensity of infection in relation to livestock was surprising. Though currently there is no data on the species for use to benchmark these findings as either low or high, the observed levels were low as compared to reports from a number of other African bovids (Woodford, 1976; Boomker *et al.,* 2000). The results contradicted observations by Apio *et al*., (2006) in a study on bushbuck (in Uganda) which had a much higher intensity of coccidia, but a much lower intensity of strongyles. Ezenwa, (2003a) also observed higher intensities from a number of wild bovids in Kenya.

5.2.2 Spatial and Temporal Variations in Intensity of Infection

Overall, animals in Ishaqbini conservancy tended to have lower intensities of gastrointestinal parasites than those in the Tsavo area. This suggested that environmental differences between the two areas could have influenced the observed differences in intensities of infection of gastrointestinal parasites in these animals. However, though the intensities of coccidia in Hirola were higher in Tsavo than their counterparts at Ishaqbini, the intensity of infection of strongyles was unexpectedly higher in Ishaqbini. The reasons for this disparity could not be immediately ascertained.

Significantly higher intensities of infection of both strongyles and coccidia were recorded during the wet than dry season. This pattern was the same for all the animals sampled and it signified the role of environmental factors in determining the intensity of infection with gastrointestinal parasites. Armour (1980) observed that adult female

worms released their eggs preferentially when the environmental conditions were optimal for the successful survival and development of their infective larvae. In the environment humidity is required for successful development and survival of parasite stages and movement of larval nematodes (Nielsen *et al.*, 2007).

The seasonal patterns in parasitism indicated that the long dry season may have limited the development and survival of parasite stages in the environment and their transmission to hosts. A study by Jacquiet *et al.*, (1995) in the semiarid area of Mauritania found that young goats born during the dry season were free from gastrointestinal nematode infections until the following rainy season. This indicated a lack of transmission during the dry season. Thus, increased parasitism in the wet season may have been due to a resumption of transmission, or parasite activity in the case of arrested strongyle larvae. In addition, the pulse of new born naïve hosts during the wet season increased the number of susceptible hosts especially in the case of Hirola which mostly calf during the wet season between October and November.

5.3 Frequency Distribution of Strongyles and Coccidia

The frequency of distribution of strongyles and coccidia in the study animals showed that they were highly aggregated in all the hosts but the differences in aggregation were not statistically significant. Exhaustive empirical surveys have shown that, almost without exception, macroparasites are aggregated across their host populations, with most individuals harbouring low numbers of parasites, but a few individuals playing host to many (Shaw and Dobson, 1995).

This study revealed that gastrointestinal parasites were more aggregated in cattle than in other animals. Heterogeneities such as these are generated by variation between individuals in their exposure to parasite infective stages and by differences in their susceptibility once an infectious agent has been encountered (Wilson *et al;* 2002). However, in the current study, a comparison of ***k*** values for the four hosts showed that the differences in the levels of aggregation were not statistically significant. This observation suggested a very close similarity in the factors influencing aggregation (and also prevalence and intensity) in Hirola and livestock.

Studies have shown that aggregation of gastrointestinal parasites was influenced by factors such as age, sex, body condition, behaviour, and genetics of the host, genetics of the parasite and external heterogeneities such as spatial distribution of external infective stages (Wilson *et al.,* 2002). The effects of some of these factors on FEC and FOC are given in Appendices III and IV. Apart from these factors, the relatively high level of aggregation of strongyles and coccidia observed in cattle could also be attributed to differential use of salvage anthelmintics by livestock keepers (*pers observ.*) which could have led to some livestock having none or few infections while others would have comparatively higher cases of infection.

Parasite aggregation such as was observed in the current study has important implications for the population and evolutionary dynamics of the parasite and its host (Poulin, 1993). In macroparasites, host mortality and morbidity tends to be dose-

dependent (Wilson *et al;* 2002) and individuals in the tail end of the frequency distribution are the most affected by the parasites. In such a case, parasites are likely to exert a selection pressure, causing mortality and morbidity to a given proportion of individuals in the population (Poulin, 1993). In this way, aggregation of macroparasites has a regulatory influence on the population.

Wilson *et al.,* (2002) observed that over the years, the central theme of macroparasite studies has been the development of a theoretical and empirical understanding of the stabilizing role of aggregation in the population dynamics of parasitic helminths and their hosts. To gain a better understanding of the role played by aggregation of gastrointestinal parasites on the critically endangered Hirola, more studies need to be carried out to shed light on the sub-clinical and clinical significance of gastrointestinal parasitoses to those hosts falling in the tail of the frequency distribution.

CHAPTER SIX

CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

Both Hirola and livestock were infected by a wide variety of gastro-parasites including nematodes, trematodes, cestodes and protozoans. These parasites differed significantly in terms of their prevalence in the hosts meaning that parasite specific factors such as inherent differences in parasite fecundity, size, age and sex ratio could have influenced the prevalence patterns observed in this study. Nematodes were the most prevalent group while cestodes were the least. This could have serious implications to parasitism in these hosts given that among the nematodes observed, strongyles comprised some of the most pathogenic members, such as *Haemonchus, Trichostrongylus, Cooperia* and *Oesophagostomum.*

All the hosts had significantly higher cases of multiple parasites than single infestations. Thus, polyparasitism appeared to be a major cause of parasitoses among these animals due to the fact that multiparasite infections tended to be more severe than single parasite infestations. Goats had the highest prevalence whereas Hirola had the least.

Host species, age and sex, location and season were the factors mostly associated with prevalence patterns observed among the hosts. Amongst these factors, age and season had the greatest influence on the prevalence patterns. This implied that young hosts including those of Hirola were more susceptible to severe parasitoses than adults and that most animals tended to become infected during the wet season.

The hosts differed significantly in terms of intensity of gastrointestinal parasites. Goats had the highest intensity of strongyles and coccidia whereas Hirola had the lowest. However the overall intensity of infection with gastrointestinal parasites was light. Intensity levels were influenced by age and sex of the hosts, location and season but age and season appeared to be the underpinning factors influencing the intensity patterns in both Hirola and livestock.

The parasites were highly aggregated in the host populations with the greatest levels of aggregation being found among cattle and least among sheep. However, the differences in levels of aggregation among Hirola and livestock were not significant. This suggested that factors such as location, season, age and sex of the host nearly similar influence on the patterns of gastrointestinal parasite infections in Hirola and livestock.

Generally Hirola appeared to have lower prevalence and intensity of the parasites than livestock despite similarities in feeding behaviour. Sub-clinical infections may be very important in regulating Hirola population by reducing fitness, impairing nutrition, decreasing ability to disperse, feed, escape from predators, competition for resources or mates, or by increasing the energy expenditure. Hirola in Ishaqbini had a higher prevalence and intensity of strongyles than their counterparts in Tsavo.

6.2 Recommendations

Patterns of gastrointestinal infections among Hirola and livestock revealed in this study set the initial attempts towards unravelling the role of parasites in regulating Hirola population. Long term studies are necessary in order to identify and understand the

factors shaping these patterns. This will also help in coming up with models that would be of great use in monitoring and predicting gastrointestinal parasite infections especially in the free-ranging Hirola population. More specifically, this being a preliminary study on patterns of gastrointestinal parasites of the critically endangered Hirola, the following recommendations are made:

6.2.1 Recommendations to management

a) There is great need to target the young Hirola (or calves) in the future control and management of gastrointestinal parasites at the Hirola-livestock interface in Southern Kenya as these appeared to be at a higher risk.

b) Control and management of gastrointestinal parasites at the Hirola-Livestock in Southern Kenya should consider the seasonal patterns influencing prevalence and intensity of the parasites.

6.2.1 Recommendations for further research

c) It is important to further investigate and develop a checklist of gastrointestinal parasites of both Hirola and livestock. This will help understand the sharing of gastrointestinal parasites between Hirola and livestock and also the direction of transmission of the parasites (if there is any).

d) There is need to investigate the effects of the high levels of low parasitoses in Hirola. Subclinical parasitoses can also play a role in regulating Hirola population.

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APPENDICES

Appendix I. Statistical Tests for Factors Influencing Prevalence of Gastrointestinal Parasites

An asterisk **(*)** indicates significant differences

Appendix I (a): Differences in prevalence of strongyles according to area

Host	Tests for Prevalence of between study sites		
	$\chi 2$	Df	P
Sheep	0.730	1	0.393
Cattle	1.191	1	0.275
Hirola	0.341	1	0.559
Goats	0.001	1	0.978

Appendix I (b): Differences in prevalence of coccidia according to area

Host	Tests for Prevalence between study sites		
	$\chi 2$	Df	P
Sheep	0.353	1	0.552
Cattle	**10.886**	**1**	**0.001***
Hirola	0.057	1	0.811
Goats	0.551	1	0.457

Appendix I (c): Differences in prevalence of trematodes according to area

Host	Tests for Prevalence between study sites		
	$\chi 2$	Df	P
Sheep	1.634	1	0.201
Cattle	0.301	1	0.584
Hirola	0.386	1	0.534
Goats	0.118	1	0.731

Appendix I (d): Overall Differences in prevalence based on season

Host	Tests for Prevalence between seasons		
	$\chi 2$	Df	P
Strongyles	1.203	1	0.273
Coccidia	**9.928**	**1**	**0.002***
Trematodes	2.560	1	0.110

Appendix I (e): Differences in prevalence of strongyles based on season

Host	Tests for Prevalence between seasons		
	$\chi 2$	Df	P
Sheep	0.072	1	0.788
Cattle	0.910	1	0.340
Hirola	2.840	1	0.092
Goats	1.360	1	0.244

Appendix I (f): Differences in prevalence of coccidia based on season

Host	Tests for Prevalence between seasons		
	$\chi 2$	Df	P
Sheep	**15.327**	**1**	**0.001***
Cattle	**10.227**	**1**	**0.001***
Hirola	**3.840**	**1**	**0.050***
Goats	**20.198**	**1**	**0.001***

Appendix I (g): Differences in prevalence of trematodes based on season

Host	Tests for Prevalence between Sex of hosts		
	$\chi 2$	Df	P
Sheep	**4.160**	**1**	**0.041***
Cattle	0.862	1	0.353
Hirola	**4.812**	**1**	**0.028***
Goats	0.513	1	0.474

Appendix I (h): Differences in prevalence of strongyles based on Sex of Host

Host	Tests for Prevalence between study sites		
	$\chi 2$	Df	P
Sheep	0.003	1	0.960
Cattle	0.569	1	0.450
Hirola	0.329	1	0.566
Goats	0.814	1	0.367

Appendix I (i): Differences in prevalence of coccidia based on Sex of Host

Host	Tests for Prevalence between Sex of hosts		
	$\chi 2$	Df	P
Sheep	0.018	1	0.892
Cattle	1.271	1	0.260
Hirola	0.045	1	0.832
Goats	0.429	1	0.512

Appendix I (j): Differences in prevalence of trematodes based on Sex of Host

Host	Tests for Prevalence between Sex of hosts		
	$\chi 2$	Df	P
Sheep	0.067	1	0.796
Cattle	1.047	1	0.306
Hirola	0.213	1	0.645
Goats	0.224	1	0.881

Appendix I (k): Differences in prevalence of strongyles based on age of Host

Host	Tests for Prevalence between Sex of hosts		
	$\chi 2$	Df	P
Sheep	2.510	1	0.113
Cattle	0.825	1	0.364
Hirola	0.920	1	0.337
Goats	3.376	1	0.067

Appendix I (l): Differences in prevalence of coccidia based on Age of Host

Host	Tests for Prevalence between study sites		
	$\chi 2$	Df	P
Sheep	1.060	1	0.303
Cattle	0.041	1	0.834
Hirola	0.420	1	0.517
Goats	5.411	1	0.020

Appendix I (m) Differences in prevalence of trematodes based on Age of Host

Host	Tests for Prevalence between study sites		
	$\chi 2$	Df	P
Sheep	0.647	1	0.412
Cattle	0.126	1	0.723
Hirola	0.097	1	0.756
Goats	2.432	1	0.119

Appendix II. Statistical Tests for Factors Influencing Mean EPG and OPG of gastrointestinal Parasites

An asterisk **(*)** indicates significant differences

Appendix II (a) Differences in Mean eggs per gram based on study sites

Host		F	df	Within groups	p
Sheep	EPG * Study Area	0.073	1	76	0.788
Cattle	**EPG * Study Area**	**13.278**	**1**	**119**	**0.001***
Hirola	EPG * Study Area	0.297	1	140	0.586
Goats	EPG * Study Area	1.765	1	71	0.186

Appendix II (b) Differences in Mean oocysts per gram based on study sites

Host	Test	F	df	Within groups	Sig.
Sheep	OPG * Study Area	0.114	1	76	0.736
Cattle	**OPG * Study Area**	**5.420**	**1**	**119**	**0.022***
Hirola	OPG * Study Area	1.594	1	140	0.209
Goats	OPG * Study Area	0.629	1	71	0.430

Appendix II (c): Differences in Mean eggs per gram based on season

	Test	F	df	Within groups	Sig.
Sheep	**EPG * Season**	**9.363**	**1**	76	**0.003***
Cattle	**EPG * Season**	**12.676**	**1**	**119**	**0.001***
Hirola	**EPG * Season**	**9.217**	**1**	140	**0.003***
Goats	EPG * Season	3.078	1	71	0.082

Appendix II (d): Differences in Mean oocysts per gram based on season

	Test	F	df	Within groups	Sig.
Sheep	OPG * Season	3.393	1	76	0.071
Cattle	**OPG * Season**	**5.972**	**1**	**119**	**0.016***
Hirola	**OPG * Season**	**25.824**	**1**	140	**0.001***
Goats	OPG * Season	1.317	1	71	0.254

Appendix II (e): Differences in Mean eggs per gram based on sex of host

	Test	F	df	Within groups	Sig.
Sheep	EPG * Sex	0.025	1	76	0.874
Cattle	**EPG * Sex**	**9.946**	**1**	**119**	**0.002***
Hirola	EPG * Sex	0.691	1	140	0.407
Goats	EPG * Sex	0.904	1	71	0.343

Appendix II (f): Differences in Mean oocysts per gram based on sex of host

	Test	F	df	Within groups	Sig.
Sheep	OPG * Sex	1.241	1	76	0.270
Cattle	OPG * Sex	0.034	1	**119**	0.855
Hirola	OPG * Sex	1.082	1	140	0.300
Goats	OPG * Sex	2.332	1	71	0.130

Appendix II (g): Differences in Mean eggs per gram based on age of host

	Test	F	df	Within groups	Sig.
Sheep	**EPG * Age**	**9.243**	**1**	76	**0.003***
Cattle	EPG * Age	1.234	1	**119**	0.269
Hirola	EPG * Age	0.158	1	140	0.692
Goats	EPG * Age	0.060	1	71	0.808

Appendix II (h): Differences in Mean oocysts per gram based on age of host

	Test	F	df	Within groups	Sig.
Sheep	OPG * Age	0.331	1	76	0.568
Cattle	OPG * Age	0.109	1	**119**	0.742
Hirola	**OPG * Age**	**5.813**	**1**	140	**0.017***
Goats	OPG * Age	0.182	1	71	0.671

Appendix III. GLM Models of the Fixed Effects and Faecal Eggs Counts

(With eggs per gram as the dependent variable). The fixed effects included in the best fitting models are highlighted in **bold**.

Tests of Between-Subjects Effects	F	Df	Sig.
Study Area	0.487	1	0.485
Host	**6.150**	**3**	**0.001**
Sex	0.276	1	0.600
Age	0.344	1	0.558
Season	3.666	1	0.056
Study Area * Host	0.737	3	0.530
Study Area * Sex	3.729	1	0.054
Host * Sex	**2.990**	**3**	**0.030**
Study Area * Host * Sex	1.748	3	0.156
Study Area * Age	1.362	1	0.244
Host * Age	0.239	3	0.869
Study Area * Host * Age	1.389	3	0.245
Sex * Age	1.360	1	0.244
Study Area * Sex * Age	2.139	1	0.144
Host * Sex * Age	0.393	3	0.758
Study Area * Host * Sex * Age	**3.942**	**2**	**0.020**
Study Area * Season	3.441	1	0.064
Host * Season	**3.532**	**3**	**0.015**
Study Area * Host * Season	2.718	1	0.100
Sex * Season	2.952	1	0.086
Study Area * Sex * Season	0.198	1	0.657
Host * Sex * Season	2.555	3	0.054
Age * Season	**6.045**	**1**	**0.014**
Study Area * Age * Season	1.413	1	0.235
Host * Age * Season	**3.746**	**3**	**0.011**
Sex * Age * Season	0.002	1	0.967

Appendix IV. GLM Models of the Fixed Effects and Faecal Oocysts Counts (With oocysts per gram as the dependent variable). The fixed effects included in the best fitting models are highlighted in **bold**.

Tests of Between-Subjects Effects	F	df	Sig.
Study Area	0.207	1	0.650
Host	**5.388**	**3**	**0.001**
Sex	0.314	1	0.576
Age	0.317	1	0.574
Season	**20.387**	**1**	**0.001**
Study Area * Host	0.121	3	0.948
Study Area * Sex	0.778	1	0.378
Host * Sex	1.370	3	0.252
Study Area * Host * Sex	0.081	3	0.971
Study Area * Age	4.168	1	0.042
Host * Age	0.489	3	0.690
Study Area * Host * Age	2.474	3	0.062
Sex * Age	0.027	1	0.870
Study Area * Sex * Age	0.054	1	0.817
Host * Sex * Age	1.391	3	0.245
Study Area * Season	0.005	1	0.946
Host * Season	0.745	3	0.526
Sex * Season	**3.960**	**1**	**0.047**
Host * Sex * Season	0.099	1	0.753
Age * Season	0.104	1	0.747
Host * Age * Season	0.042	1	0.838
Sex * Age * Season	2.011	1	0.157

Appendix V. Collection of samples from livestock

Plate 1. Monitoring and collection of faecal from livestock in a boma located near an area utilised by Hirola (Tsavo East National Park) (Source: Author, 2013)

Plate 2. Collection and recording of faecal samples from livestock (Tsavo East National Park) (Source: Author, 2013)

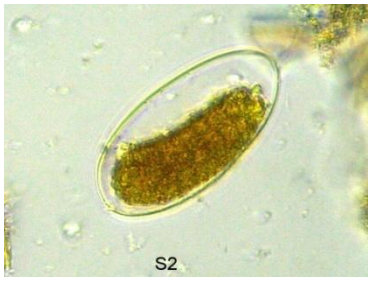
Appendix VI. Collection of samples from Hirola

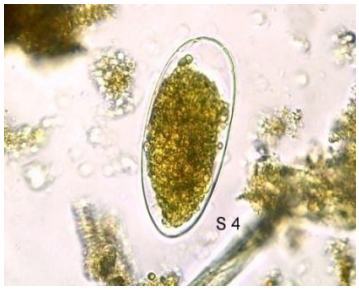
Plate 1. Two adult Hirolas and a sub-adult (in the middle) (Source: Author, 2013)

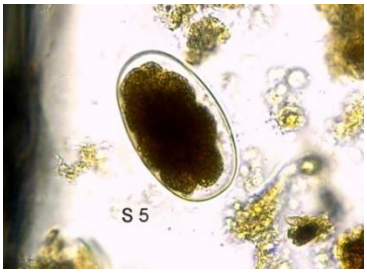
Plate 2. Collection and recording of faecal samples (Source: Author, 2013)

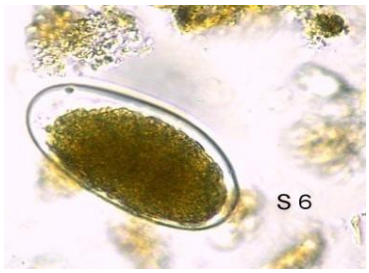
Appendix VII. Plates of Eggs, Oocysts and Larvae of Gastrointestinal Parasites Observed in the Study Animals (Source: Author, 2013)

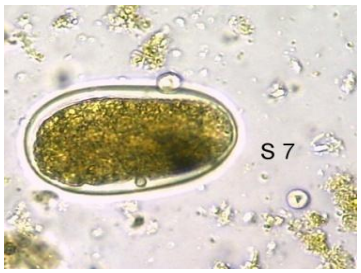
(a) Variants of Strongyle eggs

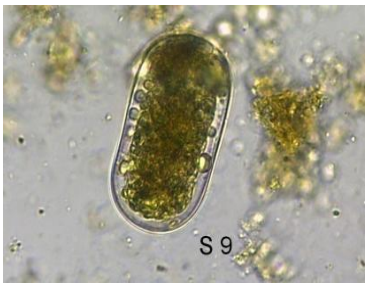


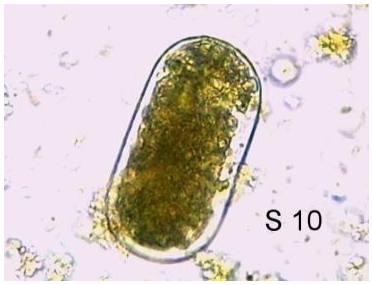


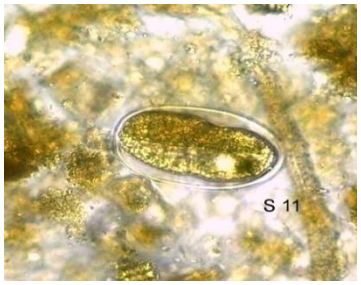


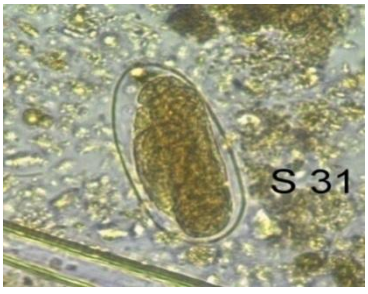


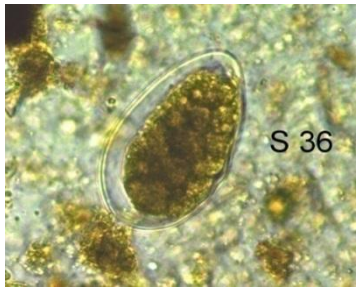


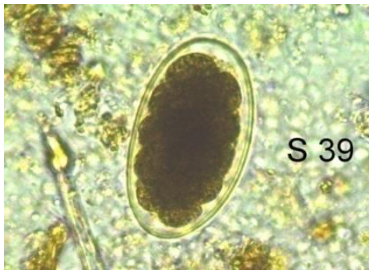






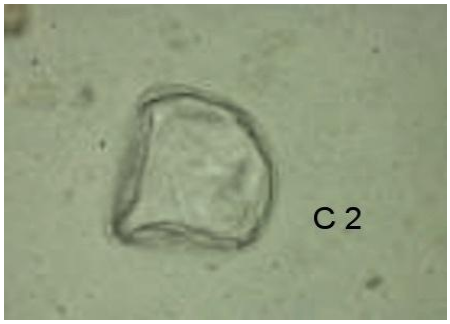




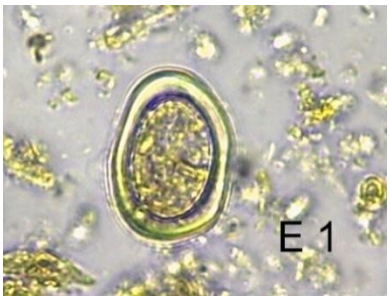


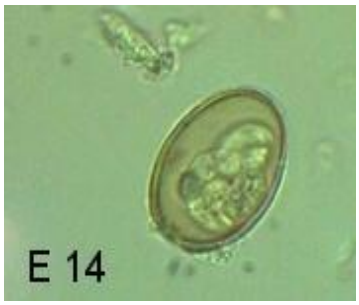
(b) Nematode *(Neoascaris spp)*

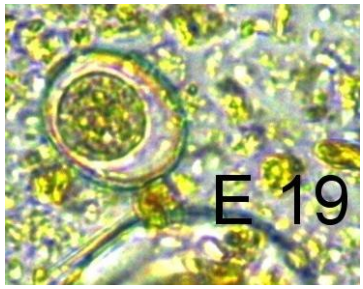
(c) Cestode (*Monieza Spp*) egg



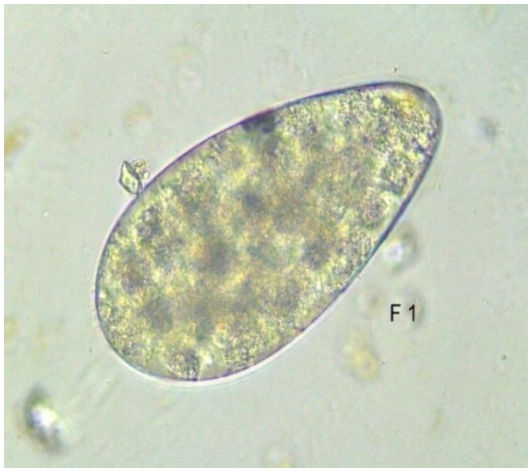
(c) Variants of Coccidia (*Eimeria spp*) cysts

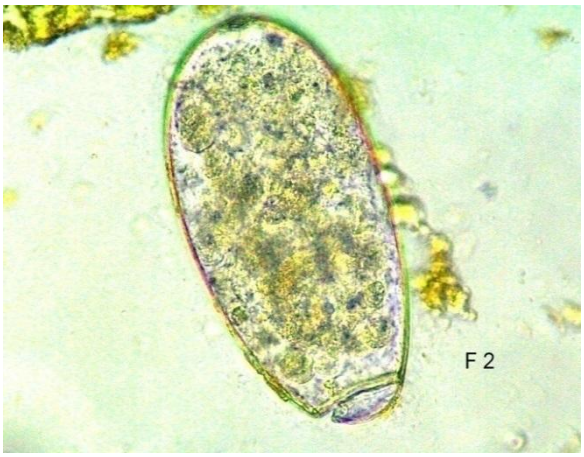


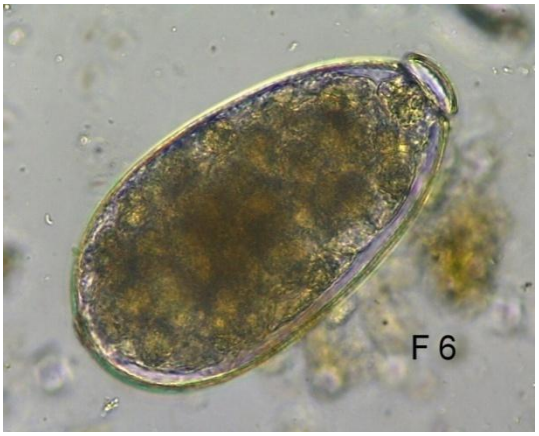


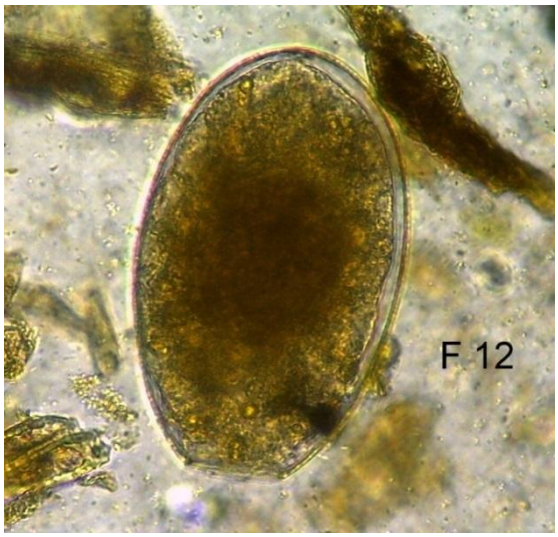


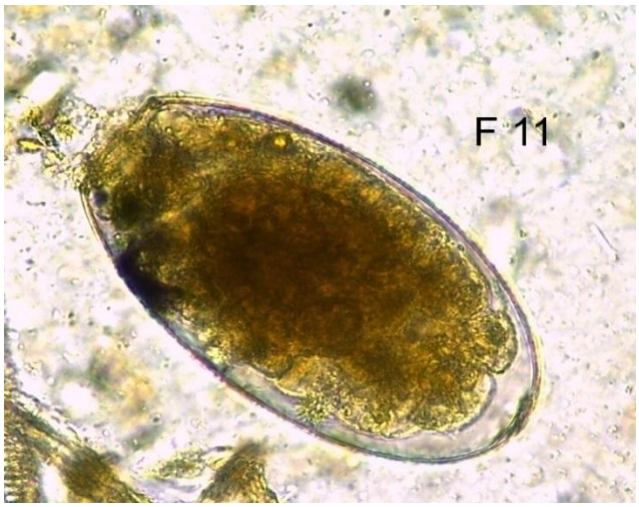
(e) Variants of Trematode (Fluke) eggs

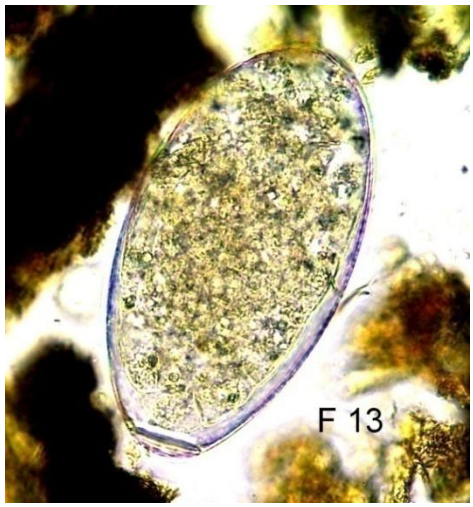












(f) Larvae of nematodes