OCCURRENCE OF Q FEVER AT THE WILDLIFE-LIVESTOCK INTERFACE OF AMBOSELI ECOSYSTEM, KENYA.

A thesis submitted in partial fulfilment of the requirements for the degree of Master of Science in Wildlife Health Management

Dr. Bilha Njeri Nguro, (BVM, UoN)

Department of Clinical Studies,

Faculty of Veterinary Medicine,

University of Nairobi

2017

DECLARATION

This thesis is my original work and has not been presented for award of a degree in any other university.

Signature _____ Date_____

Dr. Bilha Njeri Nguro, (BVM, UoN)

This thesis has been submitted for examination with our approval as the university supervisors:

Signature _____Date_____

Dr. Andrew G. Thaiyah (BVM, MSc, PhD)

Senior lecturer, Department of Clinical Studies, Faculty of Veterinary Medicine, University of Nairobi

Signature _____Date_____

Prof. George K. Gitau (BVM, MSc, PhD)

Professor, Department of Clinical Studies, Faculty of Veterinary Medicine, University of Nairobi

DEDICATION

To my father Christopher whose love is unconditional, my mother Ruth who is generous, kind hearted and hardworking, my brother Julian whose ability to make me laugh remains unmatched and my sister Christine who brings us all joy.

ACKNOWLEDGEMENTS

This thesis is a product of concerted efforts by various individuals and organisations. It may not be possible to mention here all who participated but I take this opportunity to thank them all. However, a few played very special roles which I wish to singly acknowledge.

I sincerely thank my supervisors, Dr. Andrew G. Thaiyah and Prof. George K. Gitau for their guidance, sound advice and encouragement from the development of the project proposal through fieldwork, laboratory work, data analysis and eventual preparation of this thesis. Their excellent supervision, constructive criticism and comments are highly appreciated.

My sincere thanks to Miss. Jane Kamau P.M., senior technologist in the Department of Clinical Studies, who assisted me in the field work and guided me through laboratory analysis.

I also appreciate Dr. Mwangi N.J., DVO Loitokitok Sub County for availing offices and helping to identify farmers.

My gratitude is further extended to USAID, through the One Health Central and Eastern Africa (OHCEA) for granting funds for the field work and laboratory analysis.

I would also like to appreciate the Kenya Wildlife Service (KWS) for their immense support throughout the wildlife (impala) sample collection process by providing vehicles and experienced rangers. Special thanks to Dr. Michael Njoroge (veterinarian KWS) for his immense help on logistics.

All Amboseli location pastoralists are highly appreciated for their willingness to provide the required data and information. I thank the chief Amboseli location, Mr. Daniel Kanchori for his help during the sheep and goats sampling exercise in the various selected Manyattas.

Most importantly, I am grateful to my family and friends for their support and continuous encouragement.

Finally I thank the almighty God for his providence, sustenance and his undeserved kindness.

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ABBREVIATIONS

AHP: Animal Health Practitioner

CBPP: Contagious Bovine Pleuropneumonia

CCPP: Contagious Caprine Pleuropneumonia

CDC: Centers for Disease Control and Prevention

CI: Confidence Interval

DVO: District Veterinary Officer

ECF: East Coast Fever

ELISA: Enzyme Linked Immunosorbent Assay

FMD: Foot and Mouth Disease

ID: Identity

Km: Kilometer

KWS: Kenya Wildlife Service

LSD: Lumpy Skin Disease

ml: Milliliter

NASPHV: National Association of State Public Health Veterinarians

nm: Nanometer

OD: Optical Density

OHCEA: One Health Central and Eastern Africa

OIE: World Organization for Animal Health

RVF: Rift Valley Fever

SD: Standard Deviation

Sq: square

µl: Micro liter

ABSTRACT

Q fever is an important zoonotic disease caused by *Coxiella burnetii* which is an obligate intracellular bacterium. Domestic ruminants, mainly goats and sheep, are the main source of Q fever outbreaks in humans. Very scant data is available on the role and status of Q fever in wildlife in Kenya. Direct risks of Q fever include infection of humans and animals while indirect risks include loss of income from livestock due to reduced production and reproduction. This sero-epidemiological survey was conducted to investigate the proportion of animals with Q fever antibodies and associated factors of the disease in impalas, sheep and goats at the wildlife-livestock interface of Amboseli ecosystem in Loitokitok sub-county of Kajiado County, Kenya.

Manyattas closest to the park were selected purposively since they would offer the best opportunity for wildlife-livestock interaction. A semi-structured questionnaire was also administered in all the households in selected Manyattas to gather general household data, owner/household head data, livestock production data and data on knowledge on Q fever and other zoonotic diseases in the study area. From twenty Manyattas, 200 sheep and 300goats (10 sheep and 15 goats in each Manyatta) were selected randomly. In addition, 20 impalas were conveniently captured through darting and net capture from inside the National Park. From each of the animals selected, 5ml of blood was collected through jugular venepuncture into plain vacutainer tubes for preparation of the sera. The blood was then left to stand for 1 hour in a cool box so as to clot slowly to form clear sera which was then transferred into well labelled cryo vials and stored in a refrigerator at -5°C before transport to the laboratory where it was stored at -20°C awaiting analysis.

In the laboratory, the sera were tested for antibodies against Q fever using ELISA CHEKIT Q fever test kit (IDEXX, Westbrook, Maine, USA). Both optical density and percent positivity values were generated for sera antibodies.The association between serum antibody titres and the set of independent risk factors were tested through Fishers Exact Test and Mixed Logistic Regression.

Data gathered in the study area using questionnaires showed that the knowledge on Q fever in the pastoral communities was very poor as all the interviewed animal owners had never heard of Q fever. However, other diseases such as foot and mouth disease, rabies, poxvirus and tuberculosis among others were well understood (71%) and the pastoralists took preventive measures such as boiling milk before drinking (100%), avoiding sharing sleeping quarters with animals (100%)and up to date animal vaccinations (86%). The sero-proportion of animals that tested positive for Q fever antibodies in impalas was 25% [95% confidence interval: 6%, 44%]. In sheep the sero-proportion was 6% [95% confidence interval: 2.7%, 9.3%] while goats sero-proportion was 21.7% [95% confidence interval: 17%, 26.4%]. Based on the results of Mixed Logistic Regression analysis, there was statistically significant association between species ($p=0.007$) and sero-positivity. There was no indication of confounding or interaction in any of the factors.

This study showed the presence of Q fever antibodies in impalas, sheep and goats at the wildlife-livestock interface of Amboseli ecosystem which is rich in wildlife biodiversity that interacts with livestock, their owners and visitors, this interaction may result in zoonotic disease transmission.

CHAPTER 1

1.1 INTRODUCTION

Q fever, first described in 1937 (Davis *et al*., 1981; Maurin and Raoult, 1999), is a worldwide zoonosis that has long been considered an under-reported and under-diagnosed illness because symptoms frequently are nonspecific, making diagnosis challenging (CDC, 2013). *Coxiella burnetii*, the causative agent for Q fever, has been described as one of the most infectious organisms known and is a potential agent for bioterrorism (Jones *et al*., 2006).

C. burnetii is usually shed in urine, feces, milk and birthing fluids (OIE, 2012; CDC, 2013). Following parturition or abortion, high concentrations of the organisms are shed via the placenta, amniotic fluids and the abortus (OIE, 2012). Once *C. burnetii* is in the environment, it becomes small, dense and long lasting spore-like form that is highly resistant to heat and drying (Jones *et al*., 2006; OIE, 2012). The organism contaminates dust and can therefore be disseminated by wind for long distances. *C. burnetii* is considered highly infectious since a single inhaled organism can lead to clinical illness in a host (Jones *et al*., 2006; OIE, 2012).

Q fever outbreaks are frequently seen after birth or abortion, where the causative agent *C. burnetii* is shed leading to environmental contamination (Parker *et al*., 2006). There is also evidence that ticks play an important role in the transmission of Q fever from an infected to a susceptible animal (Barandika *et al*., 2007; Mediannikov *et al*., 2010; Knobel *et al*., 2013), ticks also shed *C. burnetii* in feces thus contaminating the environment (OIE, 2012; CDC, 2013). Drinking non-pasteurized infected milk can also lead to Q fever infection (OIE, 2012).

Q fever is listed as an important zoonotic disease in the OIE *Terrestrial Animal Health Code.* All member countries and territories are required to report any diagnosed cases to the OIE (OIE, 2012). Potasman *et al* (2010), Knobel *et al* (2013) and Wardrop *et al* (2016) provided evidence that human Q fever is present in Kenya and should be considered as a significant cause of respiratory illness, however, the disease remains under-diagnosed.

Q fever infects a broad range of mammalian hosts, including cattle, goats, sheep (Barandika *et al*., 2007; Astobiza *et al*., 2010; Kshash, 2012; Knobel *et al*., 2013; DePuy *et al*., 2014), camels (Mohammed *et al*., 2014), wild ruminants (Barandika *et al*., 2007; Hernandez *et al*., 2007; Dorko *et al*., 2009), sea mammals (Kersh *et al*., 2012) fish, amphibians and reptiles (Hernandez *et al*., 2007; Knobel *et al*., 2013) as well as humans (Fennolar *et al*., 2001; McQuiston *et al*., 2002; Marrie, 2009; Potasman *et al.*, 2010; Mediannikov *et al*., 2010; Porter *et al.*, 2011; Wardrop *et al.*, 2016). Studies show that the most frequent sources of human Q fever infection are domestic ruminants mainly sheep and goats (Mediannikov *et al*., 2010). Other domestic animals that have been linked to human infection include cattle, dogs and cats (Barandika *et al.*, 2007; Knobel *et al*., 2013).

In animals, *C. burnetii* mainly localizes in the reproductive system and may primarily cause abortion or infertility (Barandika *et al*., 2007; Astobiza *et al*., 2010; Kshash, 2012; OIE, 2012; DePuy *et al*., 2014; Mohammed *et al*., 2014). In humans, however, Q fever is associated with acute flu-like illness, hepatitis, pneumonia and chronic endocarditis (Fennolar *et al*., 2001; Marrie, 2009; Mediannikov *et al*., 2010; OIE, 2012; Wardrop *et al*., 2016).

This study was designed to investigate the occurrence of Q fever at the wildlife-livestock interface of Amboseli ecosystem, Kenya, which is rich in wildlife biodiversity that interacts with

livestock, their owners and visitors and may result in zoonotic disease transmission The study provided a valuable opportunity for generating Q fever proportion data in impalas, sheep and goats which could be linked to human Q fever. A one health approach to Q fever by wildlife, livestock and human health practitioners is required in the study area in order to better understand its epidemiology.

1.2 OVERALL OBJECTIVE

The overall objective of this study was to determine the occurrence of Q fever in impalas, sheep and goats at the wildlife-livestock interface of Amboseli ecosystem, Kenya.

1.3 SPECIFIC OBJECTIVES

1. To assess the pastoral community's knowledge, attitudes and practices at the wildlife-livestock interface of Amboseli ecosystem, Kenya.
2. To determine the sero-proportion and factors associated with Q fever in impalas, sheep and goats at the wildlife-livestock interface of Amboseli ecosystem, Kenya.

1.4 STATEMENT OF PROBLEM

Q fever is a zoonotic disease with a worldwide distribution. It affects many animal species including domestic, wild and sea mammals as well as fish, amphibians, birds and reptiles. Ticks are considered as the natural primary reservoirs of *C. burnetii* (Barandika *et al*., 2007; Mediannikov *et al*., 2010; Knobel *et al*., 2013). In ruminants, Q fever is mostly associated with sporadic abortions or outbreaks of abortions and dead or weak offspring (Kshash, 2012; OIE, 2012; DePuy *et al*., 2014; Mohammed *et al*., 2014). Human Q fever is characterized by an acute or chronic illness (Mediannikov *et al*., 2010; OIE, 2012; CDC, 2013).

Potasman *et al* (2010) showed that 4(8%) of 50 safari travellers to Kenya contracted Q fever. Q fever could therefore be a strong deterrent for tourism in Kenya which is a major source of foreign exchange. There is limited data on Q fever in wildlife, livestock and humans in Kenya, which results in misdiagnosis hence under-reporting. In order to have proper diagnosis, treatment, control and prevention of Q fever; there is need for more epidemiological information on the disease. This study provided significant proportion data on Q fever in impalas, sheep and goats at the wildlife-livestock interface of Amboseli ecosystem which could be linked to human health outcomes.

1.5 JUSTIFICATION

Q fever has long been considered an under-reported and under-diagnosed disease because signs and symptoms are frequently non-specific making diagnosis challenging (CDC, 2013). At the wildlife-livestock interface of Amboseli ecosystem, there is high potential for humans and their domestic animals to come in contact with infected ticks from wild animals. Tourists and other personnel that handle animals or patrol this region can also come in contact with infected ticks or inhale infectious material leading to infection. It is important to recognise the fundamental role played by wild and domestic animals in the transmission of Q fever to humans; this requires extensive epidemiological surveillance. This study provided Q fever proportion data in impalas, sheep and goats at the wildlife-livestock interface of Amboseli ecosystem which can be used to alert wildlife, livestock and human health practitioners to the existence of the disease in this area.

CHAPTER 2: LITERATURE REVIEW

2.1 Epidemiology

Q fever is a zoonotic disease with a cosmopolitan distribution except in New Zealand. 1t was first identified in Queensland, Australia in 1935.The disease was named "Query (Q)" fever, since its etiopathogenesis was not known (Davis *et al*., 1981; Maurin and Raoult, 1999; Jones *et al*., 2006; Kshash, 2012). Although Q fever is present in virtually all 'animal kingdoms', including arthropods, the disease affects mostly humans, cattle, sheep and goats (OIE,2012; Knobel *et al*., 2013).

Since Q fever is endemic in many areas, it mostly results in explosive or sporadic cases. Due to under-diagnosis, its incidence is presumably greater than reported (CDC, 2013). Q fever advertence is typically increased during human-related outbreaks, which are mostly temporary and often constitute less than 300 acute Q fever cases (OIE, 2012). Netherlands reported the largest human community Q fever outbreaks in 2008 with 982 cases and then in 2009 with 2305 cases (OIE, 2012; CDC, 2013; Pinero *et al*., 2014).

Despite Q fever causative agent *C. burnetii* receiving attention as a potential bioterrorism agent (Jones *et al*., 2006) and having some high-profile outbreaks cases (OIE, 2012; CDC, 2013; Pinero *et al*., 2014), there is still exiguous data on the disease epidemiology in the sub-Saharan Africa (Knobel *et al*., 2013).

2.2 Host range and reservoirs

Q fever has been identified in a wide range of hosts, including; humans, birds, reptiles, arthropods (CDC, 2013; Knobel *et al*., 2013), wild and domestic mammals (Dorko *et al*., 2009).

The main reservoirs of Q fever are considered to be domestic ruminants (sheep, goats and cattle) (Barandika *et al*., 2007). Other domestic species (dogs, rabbits, cats, bird, camels etc) have also been implicated in human Q fever transmission (OIE, 2012; Knobel *et al*., 2013). The disease causative agent *C. burnetii* has also been isolated from approximately 40 species of ticks (Barandika *et al*., 2007; Mediannikov *et al*., 2010; CDC, 2013; Knobel *et al*., 2013)

2.3 Aetiology

The etiological agent of Q fever is *C. burnetii* of *Coxiellaceae* family in the order *Legionellales* of the gamma subdivision of *Proteobacteria* (Davis *et al*., 1981; Maurin and Raoult, 1999; OIE, 2012; Knobel *et al*., 2013; Mohammed *et al*., 2014). The organism is gram-negative, obligate intracellular bacteria found in small phagosomes which shelter a few organisms and fuse with each other and with other endocytic or phagocytic vesicles. After two or more days, most infected cells display one or more large replicative vacuoles (LRVs), in which the bacterium multiplies (Zamboni and Rabinovitch, 2004)

In female animals, *C. burnetii* usually localizes in the mammary glands and uterus (Dorko *et al*., 2009; Barandika *et al*., 2007; OIE, 2012), leading to shedding of copious amounts of the organism during parturition or spontaneous abortion hence environmental contamination. Once outside the host, *C. burnetii* becomes a spore-like form that is highly resistant to heat and drying and has been known to remain infective in the environment for several months (Kshash, 2012; OIE, 2012; Knobel *et al*., 2013).

2.4 Transmission and risk factors

In humans, the main routes of transmission are via inhalation of desiccated contaminated aerosol particles and through direct contact with infected animals, their products such as wool and reproductive tissues (Kersh *et al*., 2012; OIE, 2012; CDC, 2013). Another documented route of transmissions is ingestion of infected unpasteurized milk or milk products such as cheese (CDC, 2013; Knobel *et al*., 2013; Pinero *et al*., 2014). Person to person Q fever transmission is very rarely, however, the propagating circumstances include; blood transfusion and exposure during sexual intercourse or childbirth (OIE, 2012; CDC, 2013).

Animals may become infected by direct contact with infected animals and contaminated environments and/or from inhalation of aerosolized bacteria (NASPHV, 2013). Ticks are considered as the natural primary reservoir of *C. burnetii*, the agent of Q fever and transmission is as a result of bites and inhalation of contaminated aerosol due to dried excrement (OIE, 2012). There is data suggesting that ticks are responsible for the spread of the infection among wild animals and sometimes its transmission to domestic animals (Barandika *et al*., 2007; Hernandez *et al*., 2007; Dorko *et al*., 2009).

Q fever is an occupational disease in persons whose work involves contact with animals, such as slaughterhouse workers, veterinarians, and farmers, although infection is not limited to these groups (CDC, 2013). The disease also occurs among laboratory workers in medical and microbiological research facilities (Jones *et al*., 2006).

2.5 Clinical manifestations

2.5.1 Humans

Q fever infection can manifest in three ways, subclinical, acute or chronic. Symptoms associated with the acute form include; pneumonia characterized by coughing and chills, a self limiting febrile episode characterized by fever, severe headache and fatigue and/or granulomatous hepatitis (Fennolar *et al*., 2001; Marrie, 2009; Mediannikov *et al*., 2010; OIE, 2012; Wardrop *et al.,* 2016). Chronic Q fever is rare and only occurs in patients who have chronic fatigue syndrome, hepatitis, valvulopathies or vascular infections, the main clinical manifestation is endocarditis (Fennolar *et al*., 2001; McQuiston *et al*., 2002; Marrie, 2009; Mediannikov *et al*., 2010; Potasman *et al.*, 2010; Porter *et al.,* 2011; OIE, 2012; CDC, 2013; Wardrop *et al.*, 2016). Moreover, 20% to 40% of acute human Q ever cases usually end in a post-Q fever debility syndrome (Jones *et al*., 2006; CDC, 2013). In addition, Q fever infection in pregnant women can lead to placentitis hence early embryonic death, spontaneous abortion, premature birth or growth retardation (Fennolar *et al*., 2001; OIE, 2012; CDC, 2013). Since the clinical expressions are generally nonspecific, confirmatory diagnosis and treatment is frequently delayed (CDC, 2013).

2.5.2 Animals

Q fever infection in ewes, does and cows is mostly associated with reproductive disorders that present as late abortions, still births, premature birth, weak offspring or infertility (Barandika *et al*., 2007; Astobiza *et al*., 2012; OIE, 2012; Mohammed et al., 2014). Little is known about the pathogenesis of Q fever in wild animals (Barandika *et al*., 2007; Hernandez *et al*., 2007; Dorko *et al*., 2009; Kersh *et al*., 2012). Under laboratory conditions, *C. burnetii* inoculation of both

guinea pigs and mice results in a systemic infection, including pneumonia, hepatitis and splenomegaly (Roest *et al.,* 2013).

2.6 Diagnosis

In ruminants, serological survey coupled with microscopy on clinical samples have been traditionally used in Q fever diagnosis and also differentiating it from other diseases which are also normally associated with reproductive disorders such as brucellosis and trichomoniais (Field *et al*., 1983; Cowley *et al*., 1992; Fournier *et al*., 1998). Presently for clinical diagnosis, ELISA (enzyme-linked immunosorbent assay) and PCR (polymerase chain reaction) are considered as the methods of choice for direct detection and quantification (Kitterberger *et al*., 2009; CDC, 2013).

Q fever has no gold standard technique for diagnosis; therefore, efforts are emboldened for the development of a confirmatory method and reference reagents so as to provide quality control, harmonization and proficiency (OIE, 2012; CDC, 2013). In addition to ELISA (enzyme-linked immunosorbent assay), other serological test that can be used for Q fever diagnosis include, complement fixation test (CFT) and indirect immunofluorescence assay (IFA) (Kitterberger *et al*., 2009; OIE, 2012).

When PCR (Polymerase Chain Reaction) is employed as a diagnostic tool in the context of episodic abortions and/or stillbirths in a herd or flock, samples that should be collected include; vaginal swabs taken less than 8 days after parturition or abortion (to limit the number of PCR false-negative results), faeces, milk, urine, blood, aborted foetus and placenta (OIE, 2012; Mohammed et al., 2014).A Q fever positive herd or flock is one with the clinical signs (serial

abortions and/or stillbirths) and which the presence of *C. burnetii*, the agent of Q fever has been confirmed (OIE, 2012).

2.7 Treatment and control

Once a human Q fever case has been confirmed, the country's public health agencies must attempt to adjudicate the likely source of infection. In the course of investigating a single Q fever case, public health officials are encouraged to requisition assistance from their jurisdiction's animal health agencies. A large outbreak case or a cluster outbreak however requires a well-coordinated human health, animal health and public health response (OIE, 2012; NASPHV, 2013).

After the appropriate antibiotic therapy, the acute form of human Q fever in many cases resolves quite quickly. However, the chronic form of the disease normally requires prolonged antibiotic therapy of up to two years or more coupled with frequent serological monitoring of the patient (Fennolar *et al*., 2001; OIE, 2012; CDC, 2013). Without the required antibiotic therapy, complications as a result of chronic Q fever may be severe to fatal (OIE, 2012).

The treatment of choice for acute Q fever is doxycycline (Fennolar *et al*., 2001; Jones *et al*., 2006). In case of doxycycline contraindication due to allergic reactions, other antibiotic regimens that can be employed include; moxifloxacin, clarithromycin, trimethoprim/sulfamethoxazole, and rifampicin (Mediannikov *et al*., 2010; OIE, 2012; CDC, 2013). Treatment and management of chronic Q fever involves administration of the antibiotics used in the acute form for approximately 6 or more months coupled with serologic monitoring (Fennolar *et al*., 2001; OIE, 2012; CDC, 2013).

Measures that can be employed for the control and prevention of animal Q fever (especially domestic ruminants) include; vaccinations in countries where the vaccine is available (Roest *et al.,* 2013), proper manure management, having a segregated birthing area, removal and proper disposal of any risk material such as abortus and placenta, control of known reservoirs of *C. burnetii* such as rats and ticks, making changes to farm characteristics which could include building foot baths or visitor ban (Dorko *et al.,* 2009; OIE, 2012; CDC, 2013). In case of human Q fever outbreaks, suitable control and prevention options include; enforcing animal movement bans, culling of pregnant animals and other identified *C. burnetii* shedders and having a temporary breeding ban (OIE, 2012).

Recent studies in dairy cattle Q fever indicate that when antibiotics (oxytetracycline) are administered at the drying-off period, there is significant prevention of *C. burnetii* shedding around calving (Astobiza *et al.,* 2012). However, if the herd already has an established Q fever infection, the bacterial load shed by infected animals cannot be reduced by antibiotics (Astobiza *et al.,* 2012; Pinero *et al*., 2014). In a herd or flock with serial spontaneous abortions and/or stillbirths and where *C. burnetii* has been isolated as the causative agent, two parenteral injections of long acting oxytetracycline (20 mg/kg given 20 days apart) in late gestation are indicated to prevent any adverse pregnancy outcomes. However, data on treatment of domestic ruminant Q fever are sparse and inconclusive (Astobiza *et al.,* 2012; NASPHV, 2013; Roest *et al.,* 2013).

2.8 Q fever in Kenya

A serological study carried out in the 1950s in Kenya on patients with an acute febrile and respiratory illness confirmed the presence of human Q fever in the country. A more recent study

showed the seroprevalence of Q fever amongst Kenyans to range between 10% and 20 % (Knobel *et al*., 2013). Another investigation by Potasman *et al* (2000) found that out of fifty travellers (tourists) who had visited Kenya, four (8%) of them contacted Q fever.

In livestock, a distinct Q fever seroprevalence gradient has been reported with the lowest in cattle, higher in sheep and goats, and the highest in camels (DePuy *et al*., 2014). Ndeereh (2016) identified *C. burnetii*, the agent of Q fever in ticks collected from wild ruminants in Laikipia county, the pathogen was detected in the Rhipicephalus genus (*Rh. Appendiculatus*; 3.8%, *Rh. Pulchellus*; 3.0% and *Rh. evertsi evertsi*; 2.6%).

2.9 Importance

The importance of Q fever is related much more with human health and must be considered by veterinary services as of both economic and public health importance (Kshash, 2012; OIE, 2012; CDC, 2013).

CHAPTER 3: MATERIALS AND METHODS

3.1 Study area

The study was conducted at the wildlife- livestock interface within Amboseli ecosystem in Kajiado County, Kenya. Amboseli National Park is the second most visited conservation area after Maasai Mara National Reserve (Kenya information guide) and is approximately 260km (160 miles) South of Nairobi, on the border with the neighbouring country of Tanzania (Elephant voices; KWS) (Figure 3.1). Amboseli National Park lies between longitude 37°E and 37° 30' E and Latitude 2° 30' and 2° 45' S in Southern Kenya (Elephant voices; Kenya information guide).

Amboseli ecosystem offers some of the best opportunities to see African animals because its vegetation is sparse due to the long dry months (Amboseli ecosystem trust; Kenya safari guides). Average temperatures vary only slightly throughout the year. The minimum average daily temperature is 27°C and the maximum is 33°C (Kenya information guide; Okello *et al*., 2008). Droughts occur regularly in this area, and evaporation is high. The total annual rainfall is 240mm and usually expected during April and May, and again during November and December (Kenya information guide).

The Park itself is encompassed within a Pleistocene lake basin (Figure 3.2), formed when lava flowed from an erupting Kilimanjaro blocking off the course of the Pangani river, creating a lake, which is now the Amboseli basin. Over the course of time the lake dried up although the basin is still prone to seasonal flooding (Elephant voices; Kenya information guide).

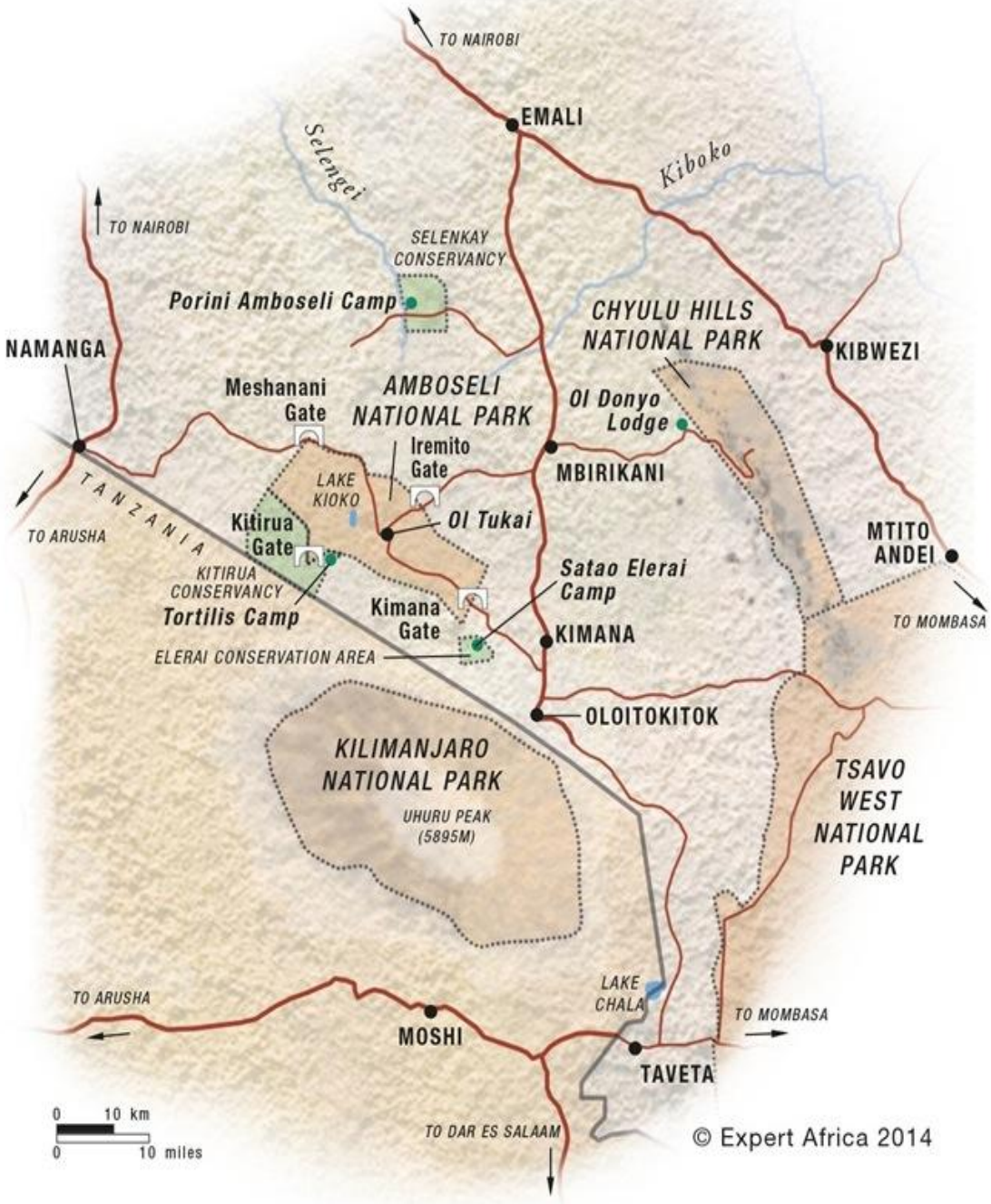

Figure 3.1: Map showing the location of Amboseli National Park in Kenya

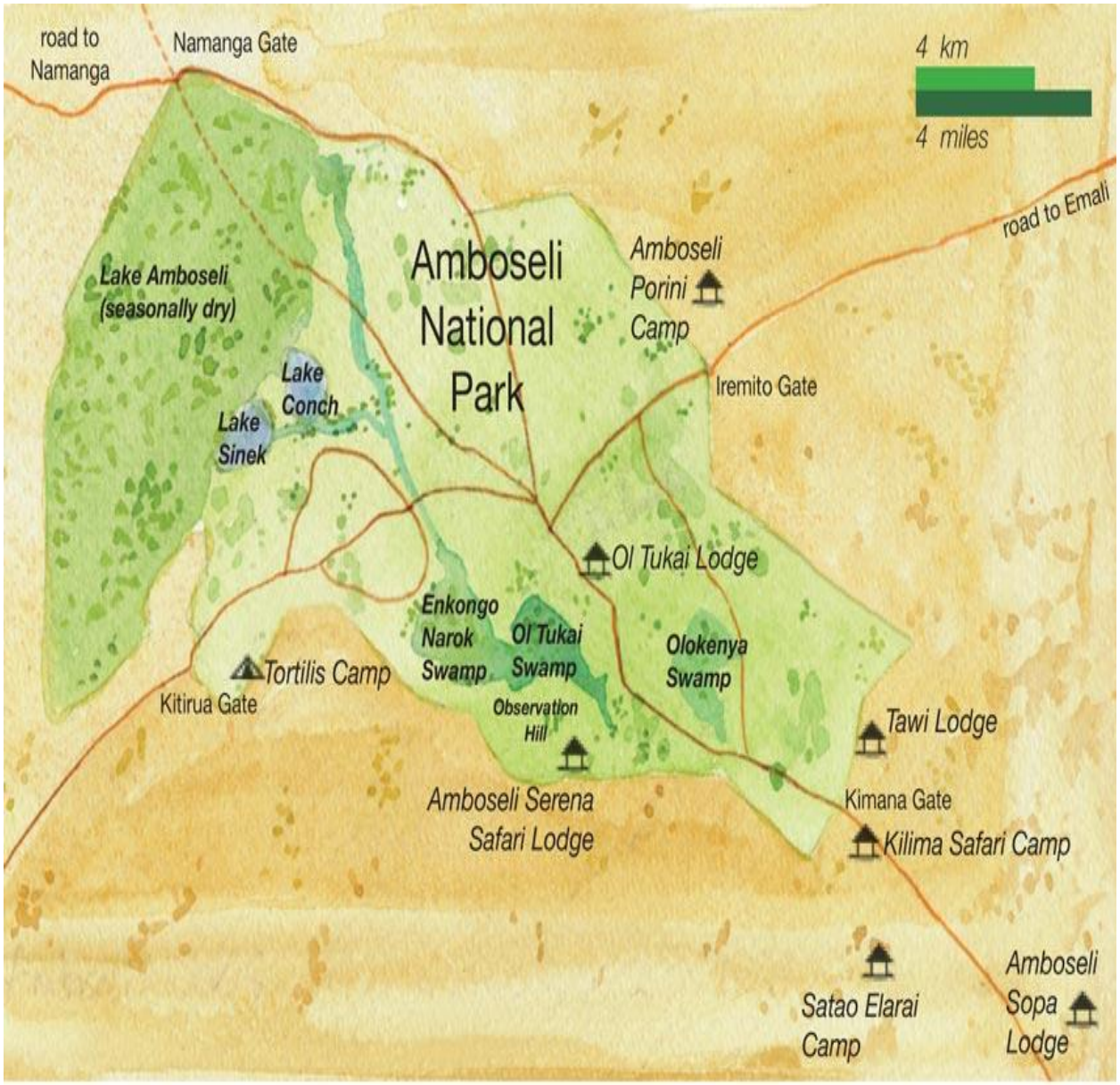

Figure 3.2: Map of Amboseli National Park and surrounding ecosystem

Two large swamps, Longinye and Enkongo Narok, transect the basin and numerous smaller swamps surface in the Central and Western parts of the park. The swamps are the life-line of Amboseli ecosystem and are home to a myriad of species of animals (Elephant voices; Kenya information guide). Thus, the Amboseli basin and Amboseli National Park constitute a dry season concentration area for migrating species of the surrounding Amboseli ecosystem, an area of approximately 3,000 sq km (Okello *et al*., 2008; Mose *et al*., 2013).

Its magnificent situation at the foot of Mount Kilimanjaro combined with its excellent opportunities to view Kenya's animals makes it one of the most visited safari parks in Kenya (Kenya information guide). Amboseli is renowned for its large herds of free-ranging African elephants, as well as huge herds of wildebeests and many other animals including giraffes, African lions, monkeys, zebras, hyenas, gazelles and antelopes (Elephant voices; Kenya information guide; Kenya safari guides; KWS).

The land outside the park is divided into group ranches occupied predominantly by the pastoral Maasai community. For years, the Maasai have occupied the extensive rangelands of Amboseli ecosystem, living and grazing alongside elephants and other migratory and non migratory herbivores. Their livelihoods are dependent on livestock keeping although other forms of land uses are emerging particularly crop farming.

3.2 Selection of the study area

The wildlife-livestock interface of Amboseli ecosystem was selected for the study because it offered a large corridor where extensive wildlife, livestock and human interaction occurs with some of the livestock and their owners accessing the Amboseli National Park for pastures and water. Wild animals also frequent human habitats for pastures. The other reason was that there

was no Q fever study done in the Amboseli ecosystem that incorporated both wild and domestic species. Such a study would therefore provide valuable information on the occurrence of Q fever in the region.

With the support of the Loitokitok sub-County Veterinary Officer, sub-locations that lie along the Park corridor were identified and five sub-locations were purposively selected. From each of these five sub-locations 4 group ranch Manyattas were selected (each ranch has more than 10 independently owned Manyattas), they include; Ilmarba, Enkong'u Narok, Esiteti, Inchakita, and Risa.

3.3 Sample size determination

In determining the sample size, the following formula described by Naing *et al*. (2006) was used to calculate the minimum number of sheep and goats for the study:

$$n = \frac{Z^2P(1-P)}{d^2}$$, where:

n = Sample size

Z = Statistic for the confidence level. A 95% confidence level was used for the study. Thus, the Z value was 1.96

P = Prevalence in sheep and goats used was 18% (p = 0.18) and 32% (p = 0.32) respectively (Knobel *et al.,* 2013).

d = the precision. A precision of 5% (d = 0.05) was used.

From this formula, the minimum number of sheep was 200 while that of goats was 300.

3.4 Data collection

3.4.1 Evaluation of pastoralist knowledge, attitudes and practices at the wildlife-livestock interface of Amboseli ecosystem

A semi-structured questionnaire (Appendix 1) was administered in all the households of selected Manyattas to gather general household data, owner/household head data, livestock production data and on Q fever and other zoonotic disease knowledge in the study area. The questionnaire covered a range of topics that included; types of livestock kept, interactions between livestock and wildlife and the types of problems encountered, diseases of importance shared between livestock and wildlife, zoonotic diseases including tick-borne and diseases that cause abortion in animals, measures undertaken to prevent tick borne diseases and taking protective measures while handling abortus. Data on questionnaires was verified first for correctness by manually checking each questionnaire to ascertain that there was no missing data before leaving the farm every day after the visit.

3.4.2 Sheep and goats sample collection

Simple random sampling by use of random tables was employed to select sheep and goats to be bled. After physically restraining the animals, 5ml whole blood was collected through jugular venepuncture using gauge18 needles into plain vacutainer tubes after disinfecting the skin with 70% alcohol (Figure 3.3 and 3.4) . The blood was then left to stand for 1 hour in a cool box so as to clot slowly with little or no hemolysis to form clear serum. The sera was then transferred into well labelled cryo vials and stored in a refrigerator at -5°C before transport to the Department of Clinical Studies, University of Nairobi laboratory where they were stored at -20°C awaiting analysis.

Figure 3.3: Blood sampling from the jugular of a goat

Figure 3.4: Sheep and goat sampling exercise in a Manyatta

3.4.3 Impalas sample collection

Convenient sampling for impalas which are the most predominant species of antelopes that interact with livestock in the study area was employed. This was done in view of the difficulties of constructing a sampling frame in wildlife to allow random sampling which requires immense resources namely; the costs of immobilization or manpower for net capture, darting accessories and transport (poor road infrastructure). This method allowed for readily available animals of the target species to be sampled.

Two method of capture were used;

1. Physical capture

Taut nets, which remain in place when animals are captured, were used (Figure 3.5). The nets were erected near a herd of antelopes but away from animals' eyesight and also downwind. This was done late in the evening at sunset. Vehicles then approached from behind the herd and slowly drove the animals towards the nets (Figure 3.6). When the animals hit the nets, they were entangled and rangers who were hiding nearby restrained them (Figure 3.7) quickly to avoid injuries resulting from struggling. The rangers were careful not to be injured by the horns of males and hooves of both males and females. After the animals were bled, they were checked for injuries before being released.

Figure 3.5**:** Erecting taut nets by KWS rangers

Figure 3.6: Driving impalas towards nets via vehicle chase

Figure 3.7**:** A female impala captured through net capture and restrained by KWS rangers

2. Chemical capture

Animals were immobilized through darting using Etorphine hydrochloride (M99®, Verico, UK) combined with Azaperone tartarate (Kyron Laboratories, S. Africa) at dosages recommended by McKenzie (1993) depending on the animal age, degree of excitation, body condition as well as the sex and terrain.

The drugs were delivered remotely by the KWS veterinarian from a vehicle by use of projectile darts using a carbon dioxide (CO2) operated darting rifle (Dan-Inject®, Dan-Inject APS, Denmark) into parts of the body with well covered muscles such as the hindquarters (Figure 3.8). Immediately the animals went down, they were put on sternal recumbency to decrease the incidence of bloat and regurgitation as well as protect the airways by decreasing the pressure of the abdominal viscera on the diaphragm.

Figure 3.8: A male impala that has been darted in the hindquarters

Blood collection, Revival, Marking and Release

Five (5) ml whole blood was collected through jugular venepuncture (Figure 3.9) using gauge18 needles into plain vacutainer tubes after disinfecting the skin with 70% alcohol. The blood was then left to stand for 1 hour in a cool box so as to clot slowly with little or no hemolysis to form clear serum. The sera were then transferred into well labelled vacutainer cryo vials and stored in a refrigerator at -5°C before transport to the Department of Clinical Studies, University of Nairobi laboratory where they were stored at -20°C awaiting analysis.

After the sampling procedure, Etorphine hydrochloride was reversed with Diprenorphine hydrochloride (M5050®, Verico, UK) calculated at three times the amount in milligrams of Etorphine hydrochloride used for each individual animal. Azaperone tartarate was the sedative drug that was used to calm the animal in the initial central nervous system excitatory phase of Etorphine hydrochloride before its central nervous system (CNS) depressive properties took effect. It does not have an antidote and was left to be metabolized and excreted physiologically from the body

Sampled animals were marked with a coloured spray to avoid re-sampling (Figure 3.10). After blood collection, marking and revival, the animal was released and observed till it rejoined the herd. A few animals sustained minor lacerations during net capture which were managed by applying a broad spectrum antibiotic (alamycin) spray.

Figure 3.9: Bleeding through jugular venepuncture in a male impala

Figure 3.10**:** A female impala that has been marked using a coloured spray

3.5 Waste disposal

During the entire sampling exercise of the impalas, sheep and goats, both biological and sharps waste generated was put into well sealed disposable containers and transported back to the University of Nairobi, Department of Clinical Studies for disposal.

3.6 Laboratory diagnosis

Serological assay of the agent of Q fever, *C. burnetii* antibodies in the sera was done using ELISA CHEKIT Q fever test kit (IDEXX, Westbrook, Maine, USA).

The kit contained: **coated plates** containing 96 wells which are the binding sites for *C. burnetii* antibodies; **Positive and negative controls**: the controls help to normalize or standardize each plate. Controls are also used to validate the assay and to calculate sample results; **wash concentrate** which is a buffered solution containing detergent used to wash away unbound materials from the plates; **conjugate** which contains enzyme-labeled antigens that react specifically to plate-bound sample analytes. Unbound conjugate is washed away after incubation and before the addition of substrate. The optical density of the colorimetric substrate is directly proportional to the quantity of bound enzyme present; **substrate** which contains a mixture of hydrogen peroxide and a chromogen that reacts with the enzyme portion of the conjugate to produce color; **stop solution** which stops the enzyme-substrate reaction and, thereby, the color development. All the kit components were stored at 2–8°C.

As per the manufactures instructions the following procedure was followed: All the kit components and the sera stored at -20°C were thawed to room temperature (18–25°C) before

beginning the procedure. The reagents were mixed gently by inverting or swirling and the plate position recorded.

One hundred (100) µl of positive and negative controls were dispensed into duplicate wells each, followed by dispensing of 100µl of sera into appropriate wells; the plate was tapped gently to mix the contents. The microplate was covered and placed inside a humid chamber (to avoid evaporation) and then incubated for 60 minutes at 37°C. After incubation, excess solution in the wells was removed and each well washed with approximately 300 µl of wash solution 3 times while avoiding plate drying between the washings.

The plate was then tapped onto an absorbent material after the final washing to remove any residual fluid. 100µl of the conjugate was then dispensed into each well, the plate covered, placed in a humid chamber and incubated for 60 minutes at 37°C. The washing procedure was repeated prior to dispensing 100µl of TMB substrate into each well and incubated at room temperature (18–26°C) for 15 minutes away from direct sunlight. Afterwards, 100µl of stop solution was dispensed into each well.

Results were read using a photometer at a wavelength of 450 nm. Results were obtained using the following formulas:

$$\textbf{Negative control } (\mathbf{NC\bar{X}}) = \frac{\mathbf{NC1A\ (450) + NC2A\ (450)}}{\mathbf{2}}$$

$NC\bar{X}$ = Plate mean negative control OD

NC1A = Plate OD value of the first negative control

NC2A= Plate OD value of the second negative control

Positive control (P $C\bar{X}$) = $\frac{\mathbf{PC1A\ (450)+PC2A\ (450)}}{\mathbf{2}}$

P $C\bar{X}$ = Plate mean positive control OD

PC1A = Plate OD value of the first positive control

PC2A = Plate OD value of the second positive control

Sample (%) =100* $\frac{\mathbf{Sample\ A\ (450)-NC\bar{X}A(450)}}{\mathbf{PC\bar{X}-NC\bar{X}}}$

Sample A = Plate OD value of sample to be calculated

NC$\bar{X}$ = Plate mean negative control OD

P C$\bar{X}$ = Plate mean positive control OD

A sample was considered positive if the percentage result was ≥40%.

3.7 Evaluation of potential risk factors to Q fever

The potential risk factors that could predispose local residents of the Amboseli ecosystem wildlife-livestock interface and their animals to Q fever (Appendix 3) were derived from the findings of the questionnaire (Appendix 1) and the animal Biodata forms (Appendix 2).

3.8 Data handling and analysis

All data collected from questionnaires and from blood analysis were entered into Microsoft office Excel 2007 spreadsheet file. The data on Microsoft Excel spreadsheet file was then exported to the statistical packages, SPSS 16.0 and STATA for statistical analysis. The packages were used for data editing and all statistical procedures. ELISA results were converted to a binary outcome where any animal that tested ELISA positive (1) was considered as infection-positive while those testing ELISA negative (0) were considered infection-negative.

The sero-proportion of Q fever in impalas, sheep and goats was determined based on serological results at 95% confidence interval. Frequency tables showing the ELISA results (positive or negative) versus the risk factors were generated and using Mixed Logistic Regression, association between sero-positivity and potential risk factors was determined. Fisher's Exact Test was used to test for association between the various risk factors.

CHAPTER 4: RESULTS

4.1 Sero-proportion of Q fever in sheep, goats and impalas at the Amboseli ecosystem wildlife-livestock interface in 2016

The sero-proportion of Q fever in, sheep, goats and impalas at the wildlife-livestock interface of Amboseli ecosystem was, 6% [2.7% , 9.3% at 95% CI], 21.7% [17% , 26.4% at 95% CI] and 25% [6% , 44% at 95% CI] respectively (Table 4.1).

Table 4.1: Sero-proportion of Q fever in impalas, sheep and goats at the Amboseli ecosystem, wildlife-livestock interface, Kajiado County in 2016

Species	ELISA positive	Total	% Sero-proportion	95% Confidence Interval	
				Lower	Upper
Impala	5	20	25	6	44
Sheep	12	200	6	2.7	9.3
Goat	65	300	21.7	17	26.4

4.2 Percentage sero-positivity in sheep, goats and impalas at the Amboseli ecosystem wildlife-livestock interface in 2016

4.2.1 Sero-positivity to Q fever in impalas by sex

The sero-positivity of Q fever in impalas according to sex at the wildlife-livestock interface of Amboseli ecosystem was 10% (2/20) in males and 15% (3/20) in females (Table 4.2).

Table 4.2: Sero-positivity to Q fever in impalas by sex at the Amboseli ecosystem wildlife-livestock interface, Kajiado County in 2016

Sex	ELISA Positive	% Sero-proportion
Male	2	10
Female	3	15
Total	5	25

4.2.2 Sero-positivity to Q fever in sheep and goats by sex

The sero-positivity of Q fever in sheep at the wildlife-livestock interface of Amboseli ecosystem was 0.5% (1/200) and 5.5% (11/200) in males and females respectively; in goats, the sero-positivity was 3.7% (11/300) and 18% (54/300) in males and females respectively. Both sheep and goats had a combined sero-positivity of 15.4% (77/500) where, 2.4% (12/500) were males while13% (65/500) were females (Table 4.3).

Table 4.3: Sero-positivity to Q fever in sheep and goats by sex at the Amboseli ecosystem wildlife-livestock interface, Kajiado County in 2016

Species	Sex	ELISA positive	% Sero-proportion
Sheep	Male	1	0.5
	Female	11	5.5
	Total	12	**6**
Goat	Male	11	3.7
	Female	54	18
	Total	65	**21.7**
Sheep & Goats	Male	12	2.4
	Female	65	13
	Total	77	**15.4**

4.2.3 Sero-positivity to Q fever in sheep and goats by sub-location in Amboseli location

The sero-positivity of Q fever in sheep and goats as per the five selected sub-locations of Amboseli location was 4.6% (23/500) in Ilmarba and Risa, 2.8% (14/500), 2.0% (10/500) and 1.4% (7/500) in Inchakita, Esiteti and Enkong'u Narok respectively (Table 4.4). Manyatta 21 had the highest number of sero-positive cases at 2.6% (13/500) while Manyatta 9 did not have any sero-positive cases (Figure 4.11)

Ilmarba had the highest number of sero-positive cases in sheep which accounted for 2.0% (4/200) of the 6.0% (12/200) total seropositive sheep cases. Enkong'u Narok and Inchakita had 1.0% (2/200) sero-positive cases each, while Risa and Esiteti had 1.5% (3/200) and 0.5% (1/200) sero-positive cases respectively. Risa had the highest number of sero-positive cases in goats which accounted for 6.7% (20/300) of the 21.7% (65/300) total sero-positive goat cases. Ilmarba followed closely with 6.3% (19/300) sero-positive cases, while Inchakita, Esiteti and Enkong'u Narok had 4.0% (12/300), 3.0% (9/300) and 1.7% (5/300) sero-positive cases respectively (Table 4.5).

Table 4.4: Sero-positivity of Q fever in sheep and goats per sub-locations by Manyatta in Amboseli location, Kajiado County in 2016

Study area	Manyatta	ELISA positive	% Sero-proportion
Ilmarba	1	9	1.8
	2	4	0.8
	3	7	1.4
	4	3	0.6
	Total	23	**4.6**
Enkong'u Narok	5	1	0.2
	6	1	0.2
	7	2	0.4
	8	3	0.6
	Total	7	**1.4**
Esiteti	9	0	0.0
	10	5	1.0
	11	4	0.8
	12	1	0.2
	Total	10	**2.0**
Inchakita	13	3	0.6
	14	4	0.8
	15	6	1.2
	16	1	0.2
	Total	14	**2.8**
Risa	18	5	1.0
	19	1	0.2
	20	4	0.8
	21	13	2.6
	Total	23	**4.6**

Figure 4.11: Percentage sheep and goat sero-positive cases by Manyatta in Amboseli location, Kajiado County in 2016

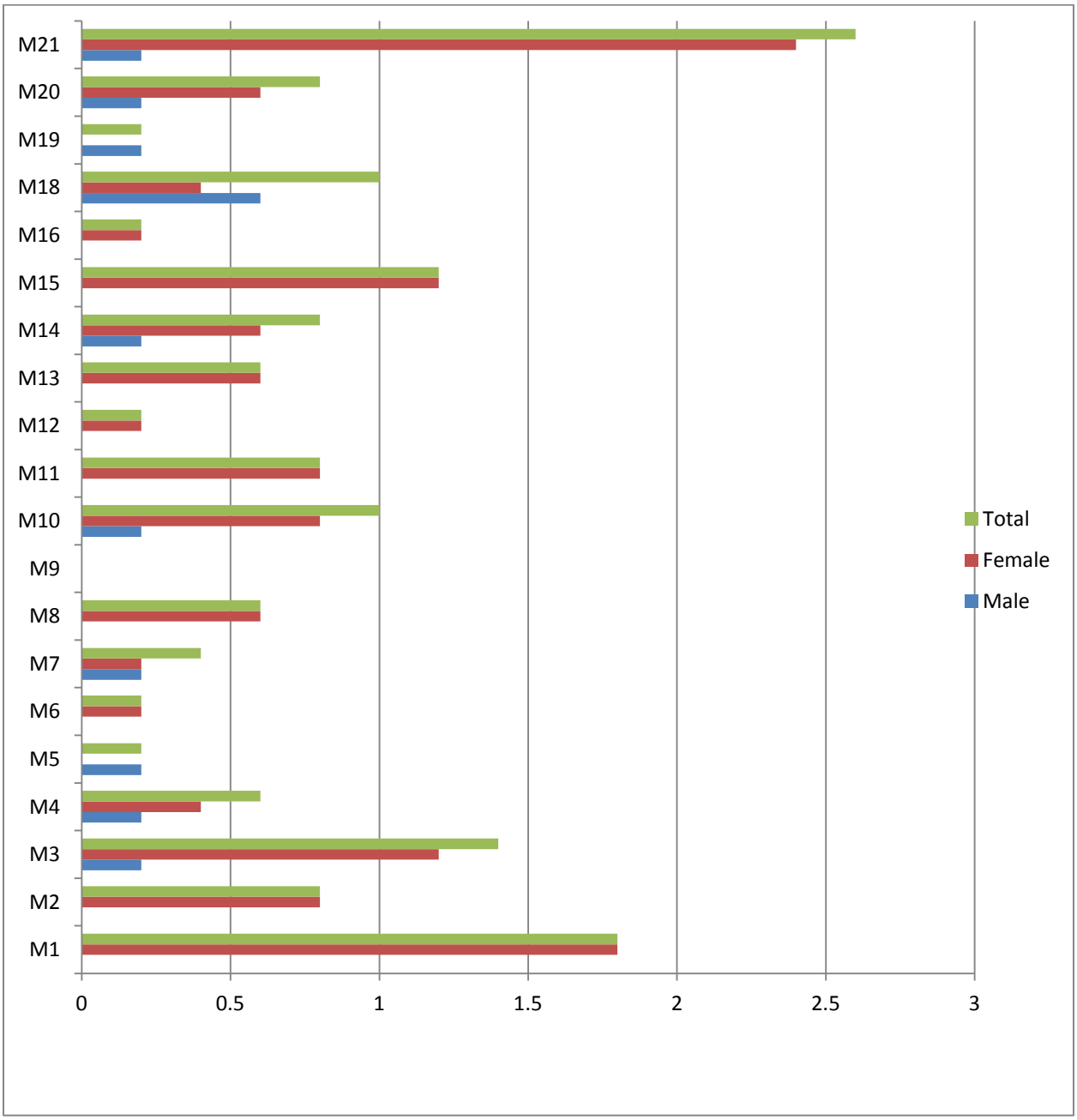

Table 4.5: Sero-positivity to Q fever in sheep and goats per sub-locations by sex in Amboseli location, Kajiado County in 2016

Study area	Species	Sex	ELISA positive	%Sero-proportion
Ilmarba	Sheep	Male	1	0.5
		Female	3	1.5
		Total	4	**2.0**
	Goat	Male	1	0.3
		Female	18	6.0
		Total	19	**6.3**
Enkong'u Narok	Sheep	Male	0	0.0
		Female	2	1.0
		Total	2	**1.0**
	Goat	Male	2	0.7
		Female	3	1.0
		Total	5	**1.7**
Esiteti	Sheep	Male	0	0.0
		Female	1	0.5
		Total	1	**0.5**
	Goat	Male	1	0.3
		Female	8	2.7
		Total	9	**3.0**
Inchakita	Sheep	Male	0	0.0
		Female	2	1.0
		Total	2	**1.0**
	Goat	Male	1	0.3
		Female	11	3.7
		Total	12	**4.0**
Risa	Sheep	Male	0	0.0
		Female	3	1.5
		Total	3	**1.5**
	Goat	Male	6	2.0
		Female	14	4.7
		Total	20	**6.7**

4.3 Risk factors associated with Q fever in sheep and goats at the Amboseli ecosystem wildlife-livestock interface in 2016

4.3.1 Risk factors to Q fever in sheep and goats

The potential risk factors to Q fever associated with sheep and goat sero-positive cases at the wildlife-livestock interface of Amboseli ecosystem identified were; game park access for pasture and watering, presence of ticks on animals, the type of tick control method, acaricide used species and sex.

From the 15.4% (77/500) sero-positive sheep and goat cases, 13.2% (66/500) of the animals had access to the game park, 11.4% (57/500) usually had ticks on them, 13.4% (67/500) of the animals were sprayed as a method of tick control and in 7.4% (37/500), a combination of acaricides were used (Table 4.6).

Table 4.6: Risk factors associated with sheep and goat sero-positive cases at the Amboseli ecosystem wildlife-livestock interface, Kajiado County in 2016

Risk Factor	Category	ELISA positive	% Sero-positivity
Game park access	Yes	66	13.2
	No	11	2.2
Ticks on animals	Yes	57	11.4
	No	20	4.0
Tick control method	spraying	67	13.4
	Dipping	7	1.4
	Both	3	0.6
Acaricide	Ectomin	7	1.4
	Triatix	15	3.0
	Dominex	18	3.6
	Combination	37	7.4

4.3.2 Mixed Logistic Regression test for significant association between risk factors and Q fever sero-positivity

Since data was hierarchical in nature with different animals nested under Manyattas which were nested under specific regions, some level of clustering was anticipated and thus mixed models were the best choice for the analysis. The average optical density (OD) was 1.023056 with a standard deviation (SD) of .6354237, data was slightly right skewed. There was a bimodal appearance in the distribution of the optical readings.

Mixed Logistic Regression was used to test for statistically significant association between the various risk factors and sero-positive goat and sheep cases, there was significant association only in species (p=0.000) (Table 4.7).

Table 4.7: Mixed Logistic Regression test for association between risk factors and sero-positive goat and sheep cases at the Amboseli ecosystem wildlife-livestock interface, Kajiado County in 2016

Variable	Estimate	Z	P value	95 % CI
Sex (male)	-.452097	-1.28	0.200	-1.143188 .238994
Species (Sheep)	-1.590605	-4.63	0.000	-2.26363 -.9175802
Ticks on the animals	-.1963314	-0.40	0.692	-1.166359 .7736966
Tick control mechanisms (Spraying)	-1.166359	-0.91	0.364	-2.914222 1.068642
Spraying, Dipping	-1.100529	-0.85	0.395	-3.635688 1.43463
Overall			0.6085	
Type of acaricide used (Dominex)	-.1052825	-0.19	0.851	-1.205895 .9953304
Ectomin	.7184603	0.68	0.499	-1.366624 2.803544
Triatix	-.3479011	-0.58	0.560	-1.517235 .8214327
Combination	ref			
Overall			0.7153	
Park access (Yes)	-.1451128	-0.23	0.820	-1.395192 1.104967

4.3.3 Fishers Exact Test for interaction between risk factors

Fishers Exact Test was used to test for interaction between the various risk factors. Most of the Manyattas 15/20 (75%) whose animals had park access had more animals with ticks on them. Moreover of all the Manyattas whose animals had park access, most 13/17 (76.47%) had ticks on them. Although these differences were evident, there was no statistical evidence that the animals in Manyattas without park access weren't equally infested with ticks with a p value of 0.7.

Similarly, although there was more tick infestation on animals in Manyattas 8/20 (40.0%) where combination methods of tick control were used, this difference was not statistically significant, the fishers Exact Test provided a $p = 0.327$.

Additionally the tick control method had no significant effect on the infestation of animals by ticks. Spraying appeared to be the least effective method with 14/18 (77.78%) of Manyattas that practiced spraying as a tick control method having animals infested by ticks. There was however no statistical evidence that this was the case with Fishers Exact Test $p = 0.447$.

4.4 Household demographics, simple shoat management factors and pastoralist zoonotic knowledge at the Amboseli ecosystem wildlife-livestock interface in 2016

4.4.1 Pastoralist knowledge on zoonotic diseases

The results on animal owners' knowledge on zoonotic diseases at the wildlife-livestock interface of Amboseli ecosystem were as follows: 95.24% were aware of foot and mouth disease, rabies, poxvirus and CCPP, 57.14% said they or a member of their family had at one time been diagnosed with a zoonotic disease; the diseases stated were FMD, tuberculosis and rabies.

All the respondents said they took preventive measures to avoid zoonotic diseases such as avoiding sharing sleeping quarters with animals and boiling milk before drinking (Table 4.8).

Table 4.8: Respondents' knowledge on zoonotic diseases at the Amboseli ecosystem wildlife-livestock interface, Kajiado County in 2016

Factor	Category	Frequency (n=21)	Percentage
Zoonotic knowledge	Yes	20	95.24
	No	1	4.76
Zoonotic diseases known	FMD	14	66.67
	Rabies	1	4.76
	Poxvirus	2	9.52
	CCPP	1	4.76
	FMD, Poxvirus	2	9.52
	Don't know	1	4.76
Family zoonotic infection	Yes	12	57.14
	No	9	42.86
Zoonotic disease affecting family	FMD	9	42.86
	Tuberculosis	2	9.52
	Rabies	1	4.76
	None	9	42.86
Sharing sleeping quarters	Yes	0	0.00
	No	21	100.00
Animal product disease transmission knowledge	Yes	5	23.81
	No	16	76.19
Boil milk?	Yes	21	100.00
	No	0	0.00
Zoonosis education	Yes	3	14.29
	No	18	85.71
Where learnt?	NGO workshops	2	9.52
	Wildlife experts	1	4.76
	Not learnt	18	85.72

4.4.2 Pastoralist knowledge on tick borne diseases

The results on animal owners' knowledge on tick borne diseases at the wildlife-livestock interface of Amboseli ecosystem were as follows; 90.48% of the respondents were aware of ECF, anaplasmosis and heartwater disease (Table 4.9). All the respondents were aware that tick borne diseases are transmitted through tick bites and they used spraying and dipping as methods of tick control, the acaricides used included triatix, dominex and Ectomin (Table 4.10).

Table 4.9: Respondents' knowledge on tick borne diseases at the Amboseli ecosystem wildlife-livestock interface, Kajiado County in 2016

Factor	Category	Frequency (n=21)	Percentage
Tick on animals	Yes	17	80.95
	No	4	19.05
Tick control	Yes	21	100.00
	No	0	0.00
Tick borne disease knowledge	Yes	19	90.48
	No	2	9.52
Tick borne diseases known	ECF	15	71.43
	Anaplasmosis	1	4.76
	Heartwater disease	1	4.76
	ECF, Heartwater	1	4.76
	Don't remember	3	14.29
Route of tick disease transmission	Bites	21	100.00

Table 4.10: Acaricide and methods of tick control practised by pastoralists in Amboseli ecosystem wildlife-livestock interface, Kajiado County in 2016

Factor	Category	Frequency (n=21)	Percentage
Tick control method	Spraying	19	90.48
	Dipping	1	4.76
	Spraying , Dipping	1	4.76
Acaricide	Triatix, Dominex	9	42.86
	Dominex	6	28.57
	Triatix	5	23.81
	Ectomin	1	4.76

4.4.3 Pastoralist knowledge on abortion causing diseases

The results on animal owners' knowledge on diseases that cause abortions at the wildlife-livestock interface of Amboseli ecosystem were as follows; 95.24% of the respondents were aware of foot and mouth disease, contagious caprine pleuropneumonia, rift valley fever, contagious bovine pleuropneumonia, pox virus and lumpy skin disease. All the respondents that handle aborted material said they did not take any preventive measures and they give aborted material to their dogs (Table 4.11).

Table 4.11: Respondents' knowledge on abortion causing diseases at the Amboseli ecosystem wildlife-livestock interface, Kajiado County in 2016

Factor	Category	Frequency (n=21)	Percentage
Abortion causing diseases knowledge	Yes	20	95.24
	No	1	4.76
Abortion causing diseases known	FMD	8	38.10
	CCPP	2	9.52
	RVF	1	4.76
	FMD, CCPP	3	14.30
	CBPP, CCPP	2	9.52
	FMD, CCPP, Pox	1	4.76
	FMD, CCPP, LSD	1	4.76
	FMD, CCPP, CBPP	2	9.52
	Don't know	1	4.76
Abortions in livestock	Yes	21	100.00
	No	0	0.00
Species aborting	Cattle	1	4.76
	Goats	5	23.81
	Sheep & goats	7	33.33
	Cattle, Sheep , goats	8	38.10
Number affected	<10	19	90.48
	>10	2	9.52
Abortion investigation	Yes	5	23.81
	No	16	76.19
Handling of aborting animals	Called AHP	3	14.29
	Nothing	18	85.71
Abortus handling	Yes	15	71.43
	No	6	28.57
Gloves for abortus handling	Yes	0	0.00
	No	15	100.00
Abortus disposal	Give to dogs	20	95.24
	Give to dogs/ leave in the field	1	4.76

4.4.4 Pastoralist animal management system

The results of animal owners' farm management system at the wildlife-livestock interface of Amboseli ecosystem were as follows; in all the households, all the respondents' said they kept cattle, sheep, goats and donkeys, pets kept were cats and dogs and they interacted with livestock. All the animals were grazed and 85.71% had access to the game park for pasture and water (Table 4.12).

All the animals interacted with wild animals during grazing and watering, these wild animals included; antelopes, zebras, giraffes, elephants, buffaloes, warthogs and sometimes hippopotamus. Wildlife-livestock interaction led to various problems that were outlined by pastoralists as; predation, diseases, parasites and competition for water and pastures (Table 4.13).

Table 4.12: Respondents' animal management system at the Amboseli ecosystem wildlife-livestock interface, Kajiado County in 2016

Factor	Category	Frequency (n=21)	Percentage
Livestock kept	Cattle, sheep, goats, donkey	21	100.00
Graze animals	Yes	21	100.00
	No	0	0.00
Game park access	Yes	18	85.71
	No	3	14.29
Wildlife interaction	Yes	21	100.00
	No	0	0.00
Interaction type	Seasonal	0	0.00
	All the time	21	100.00
Animal health consultant	Buy drugs	11	52.38
	AHP	1	4.76
	AHP , Buy drugs	9	42.86
Qualified vet consultation	Sometimes	20	95.24
	Never	1	4.76
Consultation reason	Treatment ,Vaccination	10	47.61
	Vaccination	8	38.10
	Treatment	3	14.29
Pets	Yes	21	100.00
	No	0	0.00
Pets kept	Cats, Dogs	20	95.24
	Dogs	1	4.76
Pet-livestock interaction	Always	20	95.24
	Sometimes	1	4.76

Table 4.13: Problems as a result of wildlife-livestock interaction and wild animals in contact with livestock at the Amboseli ecosystem wildlife-livestock interface, Kajiado County in 2016

Factor	Category	Frequency (n=21)	Percentage
Wild animals in contact	Antelope, Zebra, Giraffe Elephant	6	28.58
	Antelope, Zebra, Giraffe	4	19.05
	Antelope, Zebra, Giraffe Elephant, Buffalo, Warthog	3	14.29
	Antelope, Zebra, Elephant	2	9.52
	Antelope, Zebra, Buffalo, Giraffe Elephant	2	9.52
	Antelope, Zebra, Giraffe Elephant, Warthog	1	4.76
	Antelope, Zebra, Giraffe Elephant, Warthog, hippo	1	4.76
	Antelope, Elephant	1	4.76
	Antelope, Elephant, Giraffe	1	4.76
Problems due to interaction	Predation	9	42.86
	Diseases	6	28.58
	Predation , Diseases	2	9
	Parasites	1	4.76
	Predation ,Competition	1	4.76
	Predation , Parasites	1	4.76
	Parasites , Diseases	1	4.76

CHAPTER 5:

DISCUSSION

In this study, all species investigated had sero-positive cases. However, the sero-proportion rate of Q fever in sheep and goat in this study was lower compared to other reports in Kenya. Knobel *et.al* (2013) reported 32% (95% CI: 27.3–37%) in goats and 18.2% (95% CI: 12.6–25.1%) in sheep in Western Kenya while DePuy *et al*. (2014) reported sero-prevalence of up to 3-4% in cattle, 13-20% in sheep, 31-40% in goats and 5-46% in camels across five ranches in Laikipia County.

The potential risk factors to Q fever in sheep and goats identified were access to the game park for pasture and watering, presence of ticks on animals, the type of tick control method, acaricide used, species and sex. However, only species had statistically significant association with sero-positivity.

These findings show that Q fever is a significant yet under-diagnosed cause of abortion or infertility in sheep and goats (CDC, 2013), this view is further supported by the fact that all the interviewed pastoralists confirmed that abortions in their livestock was a common occurrence.

Despite a very small sample size of impalas, Q fever was present at a sero-proportion of 25%. It is therefore important to use a larger sample size and understand the role of wildlife in the epidemiology of infectious pathogens including *C. burnetii*, the agent of Q fever (Dorko *et al*., 2009; Barandika *et al*., 2007; Ndeereh, 2016). The infection dynamics and route by which transmission of infection from wild animals to livestock may occur is unclear (Kruse *et al*., 2004;

Barandika *et al*., 2007), and greater understanding of this is necessary to determine the factors involved.

This study therefore confirms that wildlife species have the potential to contribute significantly as reservoirs of Q-fever infection (Marrie, 2009) for both livestock (Porter *et al*., 2011) and humans (Medeannikov *et al*., 2012), and wildlife surveillance may be a useful tool in monitoring patterns of infection and potential disease risk. In wildlife, several reports outside Kenya exist on sero-epidemiology of Q fever in different species that include various mammals, birds, reptiles and fish (Maurin and Raoult, 1999; McQuiston *et al*., 2002; Barandika *et al*. 2007; Hernandez *et al*., 2007; Dorko *et al*., 2009; Kersh *et al*., 2012).

This was the first study conducted to investigate the sero-epidemiology of Q fever in impalas, sheep and goats at the Amboseli ecosystem wildlife-livestock interface in Kenya. Q fever should be of public health concern (Wardrop *et al.,* 2016) at the Amboseli ecosystem which has unique human-livestock-wildlife interfaces that can potentially facilitate transmission of infectious pathogens across different species. However, the disease remains unreported in wildlife, livestock and humans in the entire sub-Saharan Africa (Knobel *et al.,* 2013).

The pastoralists interviewed expressed knowledge of several zoonotic diseases mostly foot and mouth disease, rabies, poxvirus and tuberculosis among others. A high percentage was also aware of tick-borne diseases infecting livestock such as ECF, anaplasmosis and heartwater disease. However, none of the respondents expressed any knowledge of Q fever. Disease transmission at the wildlife-livestock interface was identified as a major problem encountered by the pastoralists.

Generally within Kenya, there seems to be a low level of knowledge towards many zoonotic diseases including Q fever amongst pastoral communities (DePuy *et.al*, 2014; Ndeereh, 2016) which is consistent with the findings of this study raising concerns about the potential risks of Q fever amongst local populations. Education for proper diagnosis, treatment and prevention of Q fever is needed. This will require interdisciplinary and cross-cultural work to understand how this and other disease cycles in the region could be embedded in livestock management practices.

The low level of knowledge on Q fever amongst the respondents' at the Amboseli ecosystem raise concerns about the potential risks posed by the diseases in local residents. These findings also suggest that the diseases could be circulating unnoticed in the area. Therefore, the diseases could be amongst the 'fevers of unknown origin' recorded in most medical facilities (DePuy *et.al*, 2014; Ndeereh, 2016; Knobel *et al.*, 2013). A recent study conducted by Wardrop *et al* (2016) in parts of Western and Nyanza regions, Western Kenya, in cattle and humans showed a sero-prevalence of Q fever of 2.5% in humans and 10.5% in cattle.

The study further identified certain practices which could predispose the local residents to zoonotic transmission of the diseases. These included; sharing of habitats and other resources such as water between humans, livestock and wildlife (Mediannikov *et al.,* 2010; Marrie, 2009), own treatment of livestock by most pastoralists through buying drugs, lack of livestock abortion investigation, handling of abortus without any protective gear (CDC, 2013; Mediannikov *et al.,* 2010) and giving aborted material to dogs or leaving the material in the field (OIE, 2012). Close contact of pastoralist to wild animals can expose them to tick bites (Medeannikov *et al*., 2010).

Q fever could be a possible cause of acute lower respiratory illness among the pastoralists (Knobel *et al.,* 2013) in the study area and could also be a common infection to visitors

(Potasman *et al.,* 2000) who frequent the Amboseli ecosystem. Inhalation of aerosols contaminated by the parturient fluids of infected animals is the main mode of human infection with Q fever (OIE, 2012; CDC, 2013).

Further investigation on of the role of domestic dogs and cats (Knobel *et al.,* 2013) is required as all the households visited keep them as pets. Extensive epidemiological surveillance is needed to fully understand the complex ecology of Q fever.

Q fever is listed in the OIE *Terrestrial Animal Health Code* and Member Countries and Territories are obligated to report occurrences of the disease to the OIE (OIE, 2012).

CONCLUSIONS

- Q fever was detected in impalas, sheep and goats indicating that the disease is present at the Amboseli ecosystem.
- Sheep and goats data had a statistically significant association with sero-positive cases according to Mixed Logistic Regression analysis.
- Presence of *C. burnetii*, the agent of Q fever antibodies in sheep, goats and impalas suggests that the disease could be an important cause of lower respiratory illness among pastoral communities living in Amboseli ecosystem since it's zoonotic.
- Lack of knowledge of Q fever was observed amongst the questionnaire respondents' in the local pastoral community.

RECOMMENDATIONS

1) Further Q fever sero-epidemiology studies in the Amboseli ecosystem especially in wildlife, cattle, donkeys, cats and dogs so as to understand their role in the transmission of the disease to humans.
2) The county government of Kajiado should put in place long term wildlife and livestock surveillance for Q fever and other zoonotics of importance.
3) The veterinary personnel in charge of the Amboseli ecosystem should investigate all abortion cases and include Q fever in their list of differential diagnosis of abortion causing diseases.
4) The human health sector should initiate serological survey of Q fever among the local resident of Amboseli ecosystem in hospitals especially in patients presenting with flu like symptoms, fever or/and respiratory distress so as to avoid misdiagnosis.
5) There should be a one health approach to Q fever by the wildlife, livestock and human health sectors to better understand the epidemiology of the disease at the Amboseli ecosystem.

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CHAPTER 7: APPENDICES

Appendix 1: Questionnaire on Knowledge of Q fever and other zoonotic diseases

BIODATA

Study area: _____

Location/Ward: _____

Village/Manyatta: _____

Enumerator: _____

Date: _____

Respondent: Name: _____ Age: _____

Sex: _____

Position of pastoralist in household: Head: ☐ Spouse: ☐ Son: ☐

Daughter: ☐ Employee: ☐ Other: Specify: _____

QUESTIONS

PASTORALIST KNOWLEDGE ON Q FEVER AND OTHER ZOONOTICS

1. Do you know of any disease that you can acquire from animals? Yes: ☐ No: ☐

If yes, which one(s)?

(a)Brucellosis

(b)Rift valley fever

(c)Q fever

(d)Anthrax

(e)Rabies

(f)Other: _____

2. Have you or your family member ever been infected by a zoonotic disease(s)? Yes: ☐

No: ☐ If yes, which one(s)?

(a)Rabies

(b)Q fever

(c)Brucellosis

(d)Anthrax

(e)RVF

(f) Tuberculosis

(g)Other: _____

3. What type of animals do you keep? Cattle: ☐ Sheep: ☐ Goats: ☐

Chicken: ☐ Donkey: ☐ Other: Specify_____

4. Do you graze your animals? Yes: ☐ No: ☐

Do your animals access the game park? Yes: ☐ No: ☐

5. Do livestock mix (direct contact) with wildlife during either grazing or watering?

Yes: ☐ No: ☐

If yes, is the interaction seasonal: ☐ OR all the time? ☐

Which wild animals do your livestock graze with?

(a)Antelopes

(b)Zebras

(c)Hippos

(d)Buffaloes

(f)Giraffes

(g)Elephants

(h)Warthogs

(i)Other: _____

6. Do you use any method(s) of tick control? Yes: ☐ No: ☐ If yes, which one(s)?

(a)Spraying

(b)Dipping

(c)Pour on

(d)Hand application

(e)Traditional methods

(f)Other:

Which acaricide do you use?

(a)Triatix

(b)Mostraz

(c)Norotraz

(d)Dominex

(e)Steladone

(f)Ectomin

(g) Other: _____

7. Do you know any diseases that cause abortions in cattle, sheep and goats? Ye ☐ N ☐

If yes, which one(s)?

(a)RVF

(b)Q fever

(c)Brucellosis

(d) Trichomoniasis

(e)BVD

(f)Other: _____

8. Have you ever had abortions in your livestock? Yes ☐ ☐ If yes, which animals?

Sheep: ☐ Goats: ☐ Cattle: ☐ Other: specify:

How many animals were affected?

Did you try and find out the cause of abortion? Yes: ☐ No: ☐

If yes, how did you handle the aborting animals?

(a)Called a Veterinary surgeon

(b)Called an Animal health practitioner

(c)Called a Traditional healer

(d)Nothing

(e)Other: _____

9. Do you handle aborted material? Yes: ☐ No: ☐

If yes, do you use protective clothing and/ glove? Yes: ☐ No: ☐

How do you dispose the aborted material?

(a)Burning

(b)Burial

(c)Throw in a pit latrine

(d)Leave it in the field

(e) Give to dogs

(f)Other: _____

10. Do your animals usually have ticks? Yes: ☐ No: ☐

11. Do you know whether animal can get diseases from ticks? Yes: ☐ No: ☐

If yes, which diseases do you know?

(a)Q fever

(b)Anaplasmosis

(c)Babesiosis

(d)ECF

(e)Heartwater disease

(f)Other: _____

12. Do you know how animals get diseases from ticks?

(a)Direct contact

(b)Ingestion

(c) Bites

(d)Other: _____

13. Does anyone in the family share sleeping quarters with the animals? Yes: ☐ No: ☐

If yes, which family member? Father: ☐ Mother: ☐ Girls: ☐ Boys: ☐

Employee: ☐ Other:

What problems has he/she encountered?

(a)Parasites; Ticks; ☐ Mites; ☐ Fleas; ☐ Lice; ☐

(b)Diseases

(c) Allergies

(d)Trauma

(e)Other: _____

14. What problems do you encounter as a result of livestock mixing with wildlife?

(a)Diseases

(b)Parasites

(c)Competition for pastures and water

(d)Predation

(e)Other: _____

If its diseases, which one(s)?

(a)Anaplasmosis

(b)FMD

(c)Tuberculosis

(d)Q fever

(e)MCF

(f)ECF

(g) Rabies

(h) Other: _____

15. Who do you consult concerning your animals health?

(a)Veterinary surgeon

(b)Animal health practitioners

(c)Traditional healers

(d)Buy drugs

(e)Other: _____

16. How often do you consult a qualified veterinarian concerning your animal's health?

Always: ☐ Sometimes: ☐ Never: ☐

What are the reasons for consultation?

(a)Treatment

(b)Vaccination

(c)Routine practice e.g. dehorning, PD

(d) Nutritional advice

(e)Animal movement

(f)Other: _____

17. Do you keep any pet animals? Yes: ☐ No: ☐ If yes, which ones?

(a) Cats

(b) Dogs

(c) Rabbits

(d)Other: _____

How often do they interact with the livestock? Always: ☐ Sometimes: ☐

Never: ☐

18. Do you know that handling animal products can result in disease transmission?

Yes: ☐ No: ☐

19. Do you boil your milk before drinking? Yes: ☐ No: ☐

Have you been taught about the dangers of disease transmission between livestock, wildlife and humans? Yes: ☐ No: ☐

Where did you learn?

(a)Workshops

(b) Ministry of health

(c)Livestock experts

(d)Wildlife experts

(e)Neighbors

(f)Other: _____

Appendix 2: Animal Biodata Form

Date (day/month/year): _____

Farmer name: _____

Study area: _____

Manyatta: _____

Animal ID: _____

Species: _____

Sex: _____

Appendix 3: Potential risk factors that could predispose humans to Q fever infection in the Amboseli ecosystem in 2016

1. The sharing of habitats and other resources such as water between humans, livestock and wildlife
2. Own treatment of livestock by most pastoralists through buying of drugs
3. Lack of livestock abortion investigation
4. Handling of abortus without protective measures
5. Giving aborted material to dogs or leaving the material in the field

Appendix 4: Potential risk factors to Q fever in sheep and goats at the Amboseli ecosystem in 2016

Factor	Category
Sharing sleeping quarters	Yes
	No
Tick on animals	Yes
	No
Tick control	Yes
	No
Tick control method	Spraying
	Dipping
	Spraying , Dipping
Acaricide	Combination
	Dominex
	Triatix
	Ectomin
Abortus disposal	Give to dogs
	Leave in the field
Graze animals	Yes
	No
Game park access	Yes
	No
Wildlife interaction	Yes
	No
Pets	Yes
	No
Sex	Male
	Female
Species	Sheep
	Goat