SEROPREVALENCE OF *TOXOPLASMA* INFECTION IN GOATS AND SHEEP AND ASSOCIATED RISK FACTORS   IN MVOMERO DISTRICT, TANZANIA

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ABSTRACT

A cross sectional study was conducted to investigate the prevalence of toxoplasmosis in small ruminants and associated human and animal exposure risk factors. Serological detection of *Toxoplasma* antibodies was done in 300 goats and 100 sheep sera by ELISA test (Interlab, São Paulo, Brazil). Information on risk factors was collected by interview method, using structured questionnaires. The overall prevalence of toxoplasmosis was 27 and 12% in sheep and goats respectively. Dairy goat had a prevalence of 10.7%, which was not significantly lower than 13.3% in indigenous goats (p=0.48). On the other hand, sheep had a prevalence of 27%, which was significantly higher than that of local goats (p=0.007). Flock prevalence was 40% in goats and 56% in sheep. Prevalence did not vary significantly across the study areas (p>0.05). Presence of cats had strong association with prevalence of toxoplasmosis (p=0.000, RR=3.4). All of the interviewed farmers were not aware of toxoplasmosis. Human practices that were considered risks for contracting toxoplasmosis are consuming undercooked meat (100%) and milk (3.8%), sharing of water source with animals (16%), access of cats (including cat faeces) to household consumables i.e. garden (10.5%) and poor disposal of animal wastes and aborted materials (100%). The present study, reports for the first time, occurrence of toxoplasmosis in goats and sheep in Mvomero district. The prevalence and risk factors reported are of public health concern. Similar study is recommended in human beings in same particular study areas in order to properly assess and quantify human burden of toxoplasmosis.

DECLARATION

I, Sabina Peter Sheppa do hereby declare to the Senate of Sokoine University of Agriculture that this dissertation is my own original work and has neither been submitted nor being concurrently submitted for a degree award in any other University.

Signed:……………………. Date…………………………..

Sabina Peter Sheppa
(MSc. Parasitology Candidate)

The above declaration is confirmed by

Signed:……………………. Date……...…………………..

Professor E.N. Kimbita
(Supervisor)

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DEDICATION

This work is dedicated to my beloved late parents, Esther Ng'wala and Peter Sheppa,

for their nurturing, guidance and care.

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LIST OF ABBREVIATIONS AND SYMBOLS

%	Percent.
°C	Centigrade.
AIDS	Acquired Immunodeficiency Syndrome.
CNS	Central Nervous System.
CVMS	College of Veterinary and Medical Sciences.
ELFA	Enzyme-Linked Fluorescent Assay.
ELISA	Enzyme Linked Immunosorbent Assay.
EPINAV	Enhancing Pro-Poor Innovations in Natural Resources and Agricultural Value Chains
IFAT	Immunofluorescent antibody test.
IgG	Immunoglobulin G.
IgM	Immunoglobulin M.
IHA	Indirect Haemaglutination Assay.
ml	milliliter.
MEIA	Microparticle Enzyme Immunoassay.
nm	Nanometer.
PBS	Phosphate buffer saline.
PBS-T	Phosphate buffer saline Tween 20.
PCR	Polymerase Chain Reaction.
rpm	Revolutions per minute.
RR	Risk ratio.
SUA	Sokoine University of Agriculture.
µl	microliter.

URT   United Republic of Tanzania.

USA   United States of America.

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background

Toxoplasmosis is a disease caused by a protozoan parasite called *Toxoplasma gondii* (Lopes *et al*., 2008)*.* The parasite affects all warm blooded animals (Carrada-Bravo, 2005) and is one of the neglected zoonotic infections (Tenter *et al*., 2000). Human beings, cattle, sheep, goats, birds, rodents and other domestic and wild animals are the intermediate hosts of *T. gondii* (Dubey and Jones, 2008). On the other hand, felidae (cat family) are the only definitive hosts of the parasite (Klevar, 2007). The biological development of the parasite occurs both in the definitive and intermediate hosts (Dzierszinski *et al*., 2004). Bradyzoites and tachyzoites are developmental stages of the parasite, which predominantly occur in the muscles of intermediate hosts (Weiss and Kim, 2011). Oocysts define the final stage of the parasite and occur in the small intestine of the feline definitive hosts (Dzierszinski *et al*., 2004).

Naturally, cats shed oocysts in feces, which contaminate the environment including water, soil and vegetations (Alvarado-Esquivel *et al*., 2010). Contaminated environment with oocysts becomes potential source of human and livestock infection (Dzierszinski *et al*., 2004). For example toxoplasmosis has been reported in human beings exposed to contaminated water (Dabritz and Conrad, 2010; Jones and Dubey, 2012) as well as vegetables and fruits (Alvarado-Esquivel *et al*., 2011a).

In addition, human beings and carnivorous animals can be infected through consumption of infected meat from intermediate hosts (Schlundt *et al.,* 2004; Sullivan *et al*., 2009; Dubey *et al*., 2011; Mohamed *et al*., 2012). The parasite is also

excreted in the milk of infected animal (Camossi *et al*., 2011). Therefore consumption of milk from the infected animal is equally an important means of human infection (Walsh *et al*., 1999). The direct risk factors include working with livestock, association with cats, drinking unboiled water, eating undercooked meat, consuming unwashed raw vegetables and fruits (Swai and Schoonman, 2009; Alvarado-Esquivel *et al*., 2011a,b; Dubey *et al*., 2011).

Infection with toxoplasmosis in animals and human beings has a worldwide distribution and varying levels of prevalence (Dubey *et al*., 2011). For example, in Tanzania, prevalence of toxoplasmosis has been reported to be 45.7% in human beings (Swai and Schoonman, 2009), 3.6% in cattle (Schoonman *et al*., 2010), 19.3% in goats (Swai and Kaaya*,* 2012) and 31.9% in sheep (Swai and Kaaya*,* 2012). In other parts of the world, a prevalence of 31% has been reported in goats in Uganda (Bisson *et al*., 2000), 28.9% Brazil (Gondim *et al*., 1999), 25.9% Ethiopia (Negash *et al*., 2008), 42.3% Pakistani (Shah *et al*., 2013), 14.1% China (Zhao *et al*., 2011) and 53.4% in USA (Dubey *et al*., 2011).

Toxoplasma gondii infection in small ruminants not only results in significant reproductive losses, but also represents an important source of human infection due to consumption of infected meat and milk (Walsh *et al*., 1999). Tanzania is one of the largest producers of small ruminants in the world. The National Livestock Census report indicates that the country has 15,154,121 goats, which accounts for 38% of the livestock population and 5,715,549 (13%) sheep (URT, 2012)). Since 19% of goats appear to be affected by toxoplasmosis (Swai and Kaaya*,* 2012), then it is very likely

that *T. gondii* is a major public health problem in Tanzania. The major concern in Tanzania is that people have habits of consuming undercooked goat/sheep meat, which is popularly known as "nyama choma" and raw milk (Swai and Schoonman, 2009). With this practice, a considerable high proportion of the population is at a great risk of contracting toxoplasmosis. Although few studies have indicated the presence of toxoplasmosis in Tanzania, more studies are needed to provide a comprehensive status of the disease in the country. In an attempt to add knowledge on the disease status, the present study was aimed at investigating occurrence and risk factors of toxoplasmosis in goats and sheep in Mvomero district, Tanzania.

1.2 Problem statement and Justification

Studies on toxoplasmosis in small ruminants in Tanzania are limited to few areas, i.e Babati and Arumeru in Northern Tanzania (Swai and Kaaya, 2012). Studies conducted in those areas indicated that toxoplasmosis is common among goats. Previous studies did not include sheep and therefore no reports of toxoplasmosis in sheep in the country. Similarly, to date there is no any study that provides information on risk factors for human toxoplasmosis in the country. Given the limited studies on toxoplasmosis in the country, there is a need to extend the investigation in other parts of the country and including other livestock species. The present study therefore sought to estimate the prevalence of *T. gondii* in goats and sheep, and the related risk factors in Mvomero district. Small ruminants are particularly given priority because they are more susceptible to disease and greatly contribute to meat consumed in many places in Tanzania. Goat and sheep meat therefore stands a great chance of being associated with transmission of

toxoplasmosis to people. An increased risk exists in the country given the widespread tendency of consuming lightly cooked meat and raw milk (Shirima *et al.*, 2003). It is hoped that the information will increase awareness of the presence of the diseases, which will form basis of devising better ways of serving goat and sheep meat.

1.3 Objectives

1.3.1 Overall objective

The overall objective of this study was to determine seroprevalence of *Toxoplasma* infection in small ruminants and to assess the risk factors associated with human infection.

1.3.2 Specific objectives

- To determine the prevalence of *Toxoplasma* infection in dairy goats, local goats and local sheep
- To assess the risk factors related to human infections

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Historical background of *T. gondii* and toxoplasmosis

Toxoplasma gondii was first isolated in North African rodent named *Ctenodactylus gundi* (Nicolle and Manceaux, 1908). The pathogenic potential of *T. gondii* was first described in 1939 when the first case of human toxoplasmosis was reported as congenital infection in an infant with associated encephalitis and retinitis (Wolf *et al*., 1939; Weiss and Dubey, 2009a). Furthermore, Sabin and Olitsky (1937) described the obligate intracellular nature of the parasite and suggested transmission by means of ingesting *Toxoplasma* contaminated tissue. The first serological test was developed by Sabin and Feldman (1948) which led to the recognition that *T. gondii* is a common parasite of all warm-blooded hosts with a worldwide distribution (Sabin and Feldman, 1948). Combined treatment with Sulphonamides and Pyrimethamine was discovered as the standard therapy in human by Eyles and Colemann (1953). Weinman and Chandler (1956) suggested that transmission might occur through ingestion of uncooked meat. The parasite was found to cause abortion storms in sheep in New Zealand in 1957 and raised questions about potential new transmission routes for the parasite (Innes, 2010). In 1983 it was identified as a potentially fatal disease in immunocompromised adults (Luft *et al*., 1983). When the life cycle was discovered in 1970 it was found that felids were the definitive hosts of the parasite and important source of infection as they shed the oocysts in the feces (Dubey, 2008).

Studies show that *T. gondii* genotypes fall into three main lineages: I, II and III based on virulence in mice (Dardé, 2004; Dardé, 2008). Lineage I is most virulent than II

and III. There are other *T. gondii* isolates that do not fall exactly to any the three lineages mentioned previously. These are designated as Atypical (also called recombinants, exotic or unusual). Atypical genotypes are much more complex because they have genetic combinations of the three lineages (Dardé, 2008). Lineages I, II and III are predominantly found in North America and Europe while atypical genotypes are found in other continents. For example, the few strains isolated from African patients possess a fixed combination of type I and III alleles which can suggest the existence of a clonal African type (Ajzen-berg *et al.*, 2004). However, studies in African toxoplasmosis are few and comprehensive information on the circulating genotypes is not available. On the other hand strains from South America are highly divergent from those of Europe or North America (Dardé, 2004). Generally therefore, different geographical distribution of *T. gondii* geneotypes determines the prevalence of toxoplamosis observed in different parts of the world.

2.2 Taxonomy of *T. gondii*

The name *Toxoplasma gondii* is derived from its morphology and its host, i.e. toxo= arc form, plasma= life, gondii= *Ctenodactylus gundi,* a rodent host where it was found (Dubey, 2009a). It is single celled organisms that contains a nucleus and has membrane bound organelles hence belong to the kingdom Protista. As a member of phylum Apicomplexa this parasite has highly developed structures at the anterior region called apical complex, which is used in host cell invasion (Lambert, 2009). *Toxoplasma gondii* is classified in a class Coccidia. Members of this class are obligate intracellular cyst forming parasites and complete the sexual stage of their life cycles within a host's intestinal tract (De Craeye, 2012). Being the member of the

family Sarcocystidae these parasites may involve more than one host in their life cycle. Sexual development occurs only in cats resulting in shedding of oocyst in faeces.

2.3 Morphology of *T. gondii*

The morphology of *T. gondii* tachyzoite is shown in Figure 1. There are three infectious forms in the life cycle of *T. gondii*, i.e. the rapidly dividing tachyzoites, slowly dividing bradyzoites contained in tissue cysts, and sporozoites contained in sporulated oocysts.

Tachyzoite: It is oval to crescent-shaped approximately 6µm long and 2µm wide with conoid anterior end and a rounded posterior end. It is surrounded by a three-layered membrane, the pellicle. In the anterior part there is an apical complex which includes the conoid and secretory organelles namely rhoptries, micronemes and dense granules. Another structure is the apicoplast, a multiple-membrane-bound plastid-like organelle which is involved in the synthesis of protein, fat acid biosynthesis and lipid metabolism (Waller and McFadden, 2005). At the posterior end of the parasite is an ovoid nucleus, which covers the entire space of the posterior end. Tachyzoites, develop from sporozoites in the intestinal tract of the intermediate host (De Craeye, 2012). They divide rapidly in cells, causing tissue destruction and spreading the infection. In pregnant women, tachyzoites are capable of infecting the fetus (Guerina *et al*., 1994).
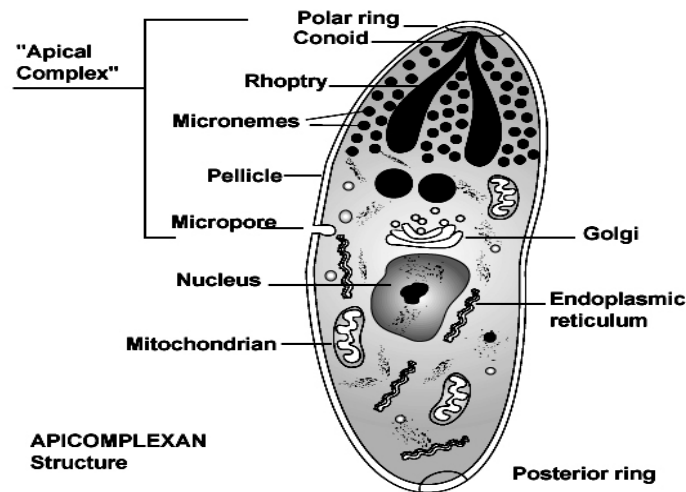
Figure 1: Morphology of *T. gondii* tachyzoite

Source: Tenter *et al.* (2000)

Bradyzoite: This differs slightly from the tachyzoites. Bradyzoites are more slender and their nucleus is located more to the posterior end compared to that of tachyzoites. Bradyzoites are found in tissue cysts. The tissue cysts are spherical in shape in the brain and in the muscle tissues they are elongated. Bradyzoites are formed when tachyzoites localize to muscle tissues and the central nervous system (CNS) where they convert to tissue cysts. This is thought to be a response to the host immune reaction (De Craeye, 2012).

Sporozoite: The sporozoites measure 2 μm by 7 μm and are found in sporulated oocysts. Oocysts are small ovoid stages of the parasite, which are shed in feces of the infected felidae. They contain two round sporocysts, each containing four elongated sporozoites, i.e. 1:2:4 configurations. Oocysts are only produced in the definitive

host, members of the family Felidae. When passed in feces and then ingested, the oocysts can infect humans and other intermediate hosts (Dubey *et al*., 1998).

2.4 Life cycle of *T. gondii*

Toxoplasma gondii is capable of infecting an unusually wide range of hosts and many different host cells (Dubey *et al*., 1998). The life cycle of *T. gondii* is facultatively heteroxenous (Figure 2). The transmission cycle is presented in Figure 3 Intermediate hosts are probably all warm-blooded animals including most livestock, and humans. Definitive hosts are members of the family Felidae, for example domestic cats (Frenkel, 2000). In intermediate hosts, *T. gondii* undergoes two phases of asexual development. In the first phase, tachyzoites (or endozoites) multiply rapidly by repeated endodyogeny in many different types of host cells. Tachyzoites of the last generation initiate the second phase of development which results in the formation of tissue cysts. Within the tissue cyst, bradyzoites (or cystozoites) multiply slowly by endodyogeny (Evans, 1999; Dubey *et al*., 1998).

Tissue cysts have a high affinity for neural and muscular tissues. They are located predominantly in the CNS, the eye as well as skeletal and cardiac muscles. However, to a lesser extent they may also be found in visceral organs, such as lungs, liver, and kidneys (Dubey *et al*., 1998). Tissue cysts are the terminal life-cycle stage in the intermediate host and are immediately infectious. In some intermediate host species, they may persist for the life of the host. The mechanism of this persistence is unknown. However, many investigators believe that tissue cysts break down

periodically, with bradyzoites transforming to tachyzoites that reinvade host cells and again transform to bradyzoites within new tissue cysts (Evans, 1999).

If ingested by a definitive host, the bradyzoites initiate another asexual phase of proliferation which consists of initial multiplication by endodyogeny followed by repeated endopolygeny in epithelial cells of the small intestine. The terminal stages of this asexual multiplication initiate the sexual phase of the life cycle. Gamogony and oocyst formation also take place in the epithelium of the small intestine. Unsporulated oocysts are released into the intestinal lumen and passed into the environment with the faeces. Sporogony occurs outside the host and leads to the development of infectious oocysts which contain two sporocysts, each containing four sporozoites (Dubey *et al.*, 1998).
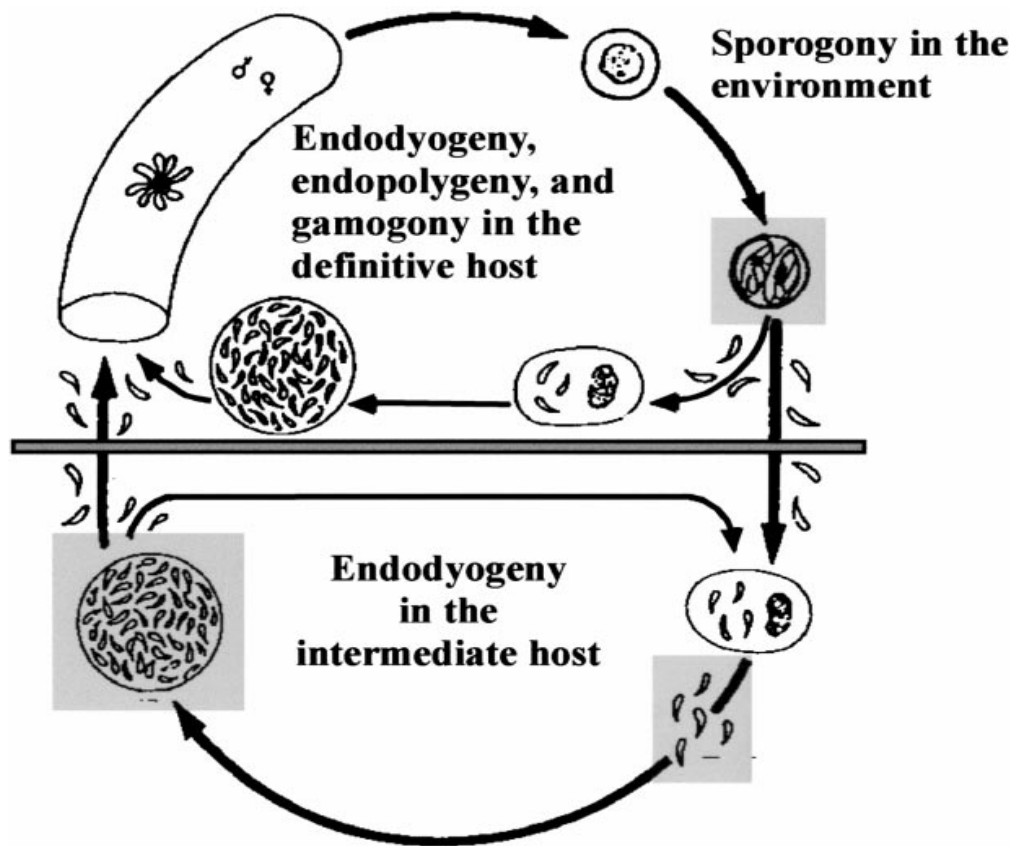
Figure 2: Life cycle of *T. gondii*

Source: Tenter *et al*. (2000)
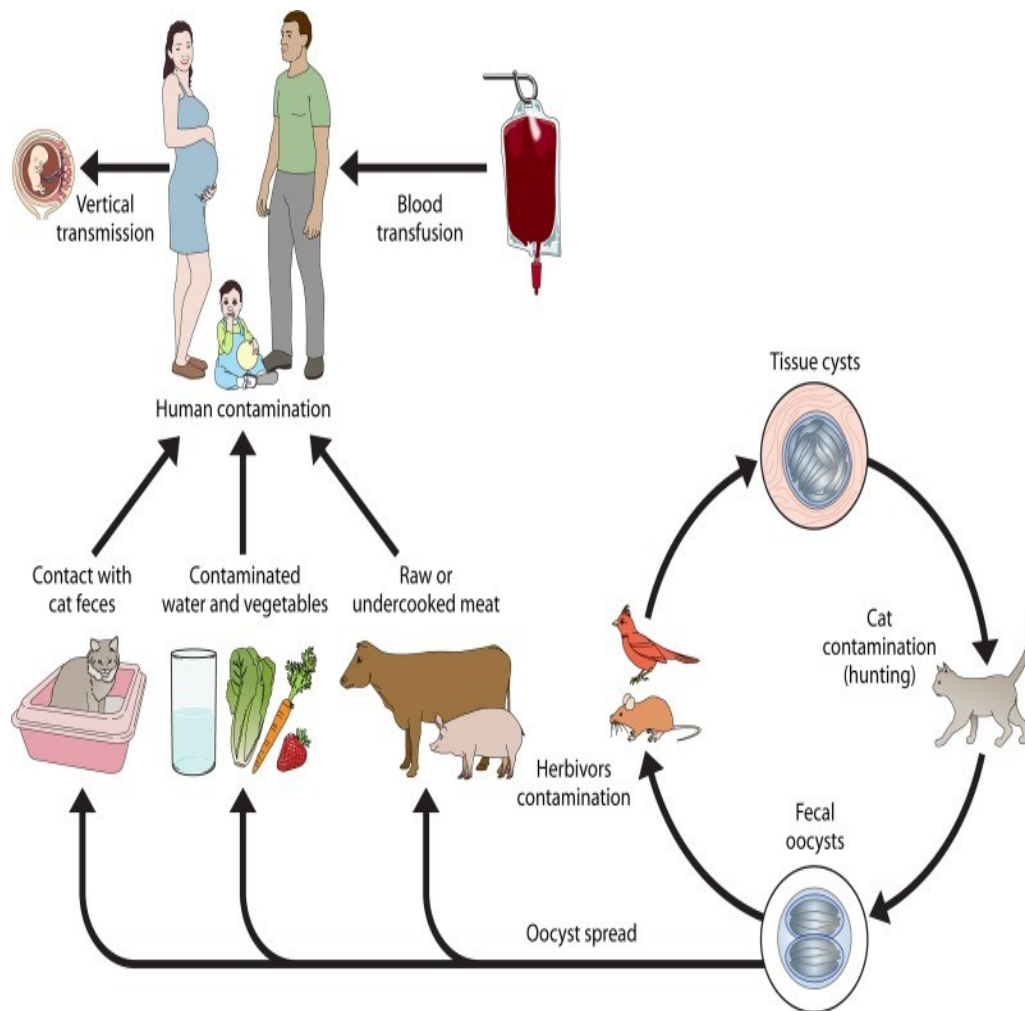
Figure 3: Transmission cycle of *T. gondii*
Source: Esch and Petersen (2013)

2.5 Epidemiology of toxoplasmosis

2.5.1 Occurrence of toxoplasmosis

It is generally agreed that approximately one third of the world's human population is infected by *Toxoplasma* (Montoya and Liesenfeld, 2004). Actually the prevalence varies widely between and within countries (10 to 80%). Low seroprevalences (10 to 30%) are reported in North America, in South East Asia, in Northern Europe, and in Sub-Saharan countries of Africa. Moderate prevalence (30 to 50%) has been found in countries of Southern Europe, and high prevalence has been found in Latin America and in tropical African countries (Pappas *et al.*, 2009). The estimated worldwide prevalence of toxoplasmosis in livestock has been reported as 30% in sheep, 15% in goats and 9% in cattle (Dubey, 2009b). Among livestock, pigs, sheep, and goats have the highest rates of chronic *T. gondii* infection and therefore considered to be major sources of human infection (Tenter *et al.*, 2000).

The rate of infection in meat-producing animals has been shown to be influenced by varying farming and management practices and the presence of cats in an area (Dubey and Jones, 2008). Environmental conditions are important for oocyst survival. Moist conditions can increase oocyst survival during long periods of heat, which likely accounts for the high prevalence in tropical countries of South America and Africa (Dubey and Jones, 2008).

2.5.2 Transmission of *T. gondii*

Cats play an important role in the spread of toxoplasmosis through fecal oral route. Felids are the only animals capable of shedding oocysts in their feces and transmitting the parasite by this means. In other host species, the ingestion of infective oocysts from cat feces or contaminated soil, water, or other materials can lead to the formation of tissue cysts that are infective via the secondary consumption of infected tissues (Dubey *et al*., 2009; Elmore *et al*., 2010). For examples, livestock acquire infection through ingestion of oocysts from contaminated environment with cat feces during grazing in pastures. Oocysts are shed in large numbers by acutely infected cats once for approximately two weeks, except in cases of feline immunosuppression, such as coinfection with feline immunodeficiency virus (FIV) or feline leukemia virus (FeLV), which can result in secondary shedding (Malmasi *et al*., 2009). After shedding, parasite sporulation takes place in one to five days, providing infective oocysts (Dubey, 2009a; Dubey and Frenkel, 1976). *Toxoplasma gondii* rapidly excysts within the environment of the intestine, dependent upon temperature, pH, bile salts, and trypsin, developing into the highly infective tachyzoite form (Yoshida *et al*., 2011; Carruthers and Boothroyd, 2007). Cellular infection is rapidly established, resulting in bradyzoite-containing tissue cysts (Yoshida *et al*., 2011; Carruthers and Boothroyd, 2007). The consumption of infected tissue or fecal material by naïve, primarily young, felines results in their infection and subsequent shedding of infectious oocysts (Dubey, 2006).

On the other hand, carnivorous mode of transmission of *Toxoplasma gondii* occurs when felids become infected by eating infected rodents, birds, or other small animals.

On the other hand, people become infected through the accidental consumption of undercooked meat containing infective cysts (Dubey and Jones, 2008; Innes *et al.*, 2009). Congenital cases of human toxoplasmosis can also occur transplacentally from mother to fetus. A pregnant woman who acquires toxoplasmosis infection can pass the organism to the developing fetus through the placenta.

2.5.3 Toxoplasmosis in human beings

Livestock plays major role in transmission of *T. gondii* to human beings. Cat faeces are potential sources of oocysts that can contaminate water, soil, vegetables and household equipments (Ogendi *et al.*, 2013). On the other hands, consumption of undercooked meat and raw milk is reported to be the most common risk practice for contracting toxoplasmosis (Tenter *et al.*, 2000). Similarly, handling cat faeces during gardening without gloves and eating without washing hands are likely to transmit toxoplasmosis in human beings. Toxoplasmosis is considered to be an occupational disease, where people who are involved with animals tend to have high incidences of toxoplasmosis. Related to occupational risk is the age. Toxoplasmosis occurs more in adults than young people (Neto *et al.*, 2008).

Toxoplasmosis in immunocompetent individuals is typically mild or asymptomatic and usually results in life-long immunity (Tenter *et al.*, 2000; Montoya and Liesenfeld, 2004). The disease develops as mild flu-like illness characterized by fever, headaches and sore throat or no illness at all. Toxoplasmosis is known to be an opportunistic infection in people with a weakened immune system, such as those

who are spleenectomized or infected with HIV and pregnant women (Negash *et al*., 2008).

In pregnant women, *Toxoplasma* is an important cause of abortion and stillbirth after primary infection. The organism can also cross the placenta, thereby infecting the developing foetus (Guerina *et al*., 1994). The parasite causes blindness as a result of retinochoroiditis and mental retardation in congenitally infected children. Retinochoroiditis and toxoplasmic encephalitis are the main symptoms in immunocompromised patients, the later being the leading cause of morbidity and mortality in AIDS patients. Recent studies show that *T. gondii* has an influence in human behaviour when parasitizing the brain. Women who are infected with *T .gondii* have increased intelligence and increased rule conscious behaviors, which is vice versa in men (Lafferty, 2006).

2.5.4 Prevention and control of toxoplasmosis in human beings

Toxoplasmosis can be prevented through a number of ways. Considering the risk factors such as accidental consumption of oocysts from the contaminated soil or ingestion of tissue cysts from infected meat, the most obvious preventive measures should focus on proper hygiene and thorough cooking of meat. People should wash hands after touching soil and disengage in areas where cats defecate. Also, milk should be boiled thoroughly and all fruit and vegetables must be washed before they are

to be consumed (Tenter *et al.*, 2000). This is particularly important for pregnant women and immunodeficient individuals.

2.5.5 Treatment of toxoplasmosis in human beings

Toxoplasmosis is particularly serious in immunocompromised people and pregnant women, and therefore treatment is mandatory for these individuals. However, treatment for immunocompetent individuals is usually not necessary. Sulfadiazine plus pyrimethamine is the most commonly recommended therapy for congenital and ocular toxoplasmosis, as well as infection in immunocompromised individuals (Montoya and Liesenfeld, 2004). Alternative treatments for patients intolerant to sulphonamides are also used. These include clindamycin plus pyrimethamine, clarithromycin plus pyrimethamine, and atovaquone (Montoya and Liesenfeld, 2004). Co-trimoxazole (trimethoprim plus sulfamethoxazole) is used in place of sulfadiazine and pyrimethamine.

2.6 Toxoplasmosis in small ruminants

Goats and sheep infected by *T. gondii* represent important sources of human infection due to consumption of meat and milk from infected animals (Dubey, 2004). However presence of cats is the main risk factor for goats and sheep to be infected. Small ruminants acquire infection through ingestion of oocysts in water or pastures contaminated with cat feces during grazing. Infection with *Toxoplasma* is more common in female and older goats and sheep (Ahmad *et al*., 2015). Toxoplasmosis is one among the causes of reproductive losses in small ruminants through inducing abortion, still birth and infertility especially in sheep (Dubey, 2004). The incidence

of infection with *T. gondii* is much higher in warmer and moist areas than in cold and dry areas. Moist conditions favour viability of oocysts (Van der Puije et *al*., 2000). Extensive and semi intensive management systems allow goats and sheep to get contact with oocysts-contaminated environment (Cavalcante *et al*., 2008). In goat, the main clinical outcome of toxoplasmosis is abortion. Other clinical signs include early embryonic death, fetal resorption, fetal death, mummification, still birth and neonatal death (Dubey, 2004).

2.7 *Toxoplasma* detection methods

Several serological (Dubey *et al*., 2011) and molecular detection (Vitale, 2013) methods are available for toxoplasmosis diagnosis. The most common serological methods include enzyme linked immunosorbent assay (ELISA) (Montoya and Liesenfeld, 2004), Microparticle Enzyme Immunoassay (MEIA), Enzyme-Linked Fluorescent Assay (ELFA) and Immune-Fluorescent Antibody Technique (IFAT) (Rodrigues *et al*., 2009). Other tests include Sabin Feldman dye test, indirect haemagglutination test, Immunosorbent agglutination assay and direct agglutination test e.g MAT (Gamble *et al.*, 2005). These techniques have their own sensitivity and specificity, depending on tissue or sample to be analyzed. Generally ELISA tests are considered to be more sensitive and specific. For example, Hill *et al*. (2006) reported a 100% sensitivity of the serum-based ELISA in both experimentally and naturally infected pigs, a sensitivity which is higher compared to other serological tests. On the other hands, a previous study (Dubey *et al*., 1995), reported a higher sensitivity with the MAT as compared with an ELISA using naturally infected sows. With regard to congenital human toxoplasmosis, Rodrigues *et al*. (2009) reported that specific IgA

anti-*T. gondii* was reactive in 25%) of the infected newborns, presenting less sensitivity (57.1%) than MEIA (60.9%), ELFA (60.9%) and IFAT (59.6%).

Polymerase chain reaction is molecular method, which is considered to be most specific and sensitive detection of *T. gondii* DNA in clinical settings and for epidemiological surveys (Gross *et al*., 1992; Wastling *et al*., 1993; Pujol-Rique *et al*., 1999; Jauregui *et al*., 2001; Contini *et al*., 2005). However, the sensitivity and reliability of PCR is subject to influence of type of sample. For example given the random distribution of tissue cysts, it becomes difficult to know the suitable organ which can yield maximum results for PCR. The low sensitivities in conventional PCR can be improved by doing Nested or real time PCR (Vitale, 2013).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study area

The study was conducted in Mvomero district in Eastern Tanzania, which is located between 5^o58' and 10^o00' S and 35^o25' and 38^o30' E. Data for dairy goats was collected from the following villages: Nyandira and Mtamba in Nyandira ward, Ndunguti in Ndunguti ward, and Kibagala, and Tchenzema in Tchenzema ward (Table 1). All these villages are found in Mgeta division. Mgeta is a moutaneous humid area located at 1800m above sea level with annual rainfall of 1800mm. Annual temperature and relative humidity range from 9-21°C and 75-94% respectively (Kusiluka *et al*., 1995). On the other hand, data related to local goats and sheep were collected from Maasai flocks at Wami Dakawa, Melela, Dakawa and Makunganya villages (Table 1). The Maasai villages are low lands with tropical climate. The estimated altitude ranging from 370-495m and annual rainfall of 975mm. Annual temperature is 25°C. The villages were randomly selected. Sampling at farm and flock levels was done conveniently based on the availability of animals, risk factors such as keeping cats and accessibility.

3.2 Study design

A descriptive cross-sectional study was conducted to determine the prevalence of toxoplasmosis in goats and sheep; and risk practices (here referred to as risk factors) in human beings.

3.3 Study subjects and variables

The study subjects consisted of randomly selected dairy goats, local goats and sheep. Also heads of households were selected for questionnaire interview. Consumption of raw milk, handling of raw goat and sheep meat, consumption of undercooked goat and sheep meat (nyama choma in Swahili language), management system and contact with cats were considered to be the most important study variables used to assess the risk factors for human toxoplasmosis.

3.4 Sample size and sampling procedures

Three hundred (300) goats and 100 sheep were sampled in this study. The sample size was based on the reported prevalence (p) of 26% in goats (pooled from Tenter *et al.*, 2000; Kamani *et al.*, 2010 and Swai and Kaaya, 2012), precision (d) of 5%, significant level (α) of 5%, and Z-score of 1.96 and calculated using the following formula:

$$n= z^2 p (1-p)/d^2$$

Samples were collected from 11 villages as shown in Table 1.

Table 1: Number of goats and sheep sampled from respective village

Village	Flock size	Number sampled	
		Goats	Sheep
Nyandira	46	30	-
Mtamba	42	30	-
Nduguti	30	15	-
Kibagala	41	20	-
Tchenzema	53	37	-
Nyasungu	8	8	-
Vidigisi	18	10	-
Wami Dakawa	188	40	25
Melela	199	40	25
Dakawa	177	40	25
Makunganya	142	30	25

Table 2: Number of animals sampled based on age and sex

Background information	Number sampled for each animal species/type			
	Dairy goats	Local Goats	Local Sheep	Total
Age				
Number of adults (≥12 months)	90	95	35	220
Number of young (≤12 months)	60	55	65	180
Sex				
Male	25	40	30	95
Female	125	110	70	305

3.5 Assessment of knowledge and risk factors

Awareness of toxoplasmosis and risk factors was assessed using Swahili-translated structured questionnaire presented in Appendix 2. An interview was conducted privately with each consenting participant, from which the following information was obtained: resident status, sex, age, socio-economic data, ownership and handling of cats, household water source, handling of raw meat and consumption of raw or undercooked meat products, consumption of raw milk, livestock kept on the farm, production system and management practices. The questionnaire was administered to the head of the household or any other mature household member (incase the head of the household was not available).

3.6 Sampling of blood and serological analysis

Blood samples (5ml) were collected from the jugular vein of each animal using plain vacutainer tubes. The blood was left for one hour to clot at room temperature. After clotting, the samples were centrifuged at 3000 rpm for 15 min and the obtained sera were decanted and stored at -20°C before analysis. An ELISA was carried out for the detection of IgG and IgM antibodies to *T. gondii* as described by Mineo *et al*. (1980), with some modifications. Briefly, polystyrene microtiter plates (Interlab, São Paulo, Brazil) previously coated with 1×10^5 *Toxoplasma* tachyzoites/well were washed three times in PBS containing 0.1% Tween 20 (PBS-T) followed by addition of 50

μl/well goat serum and incubated at 37°C for 45 minutes. Positive and negative serum controls previously determined by conventional serological tests (i.e., IHA and IFAT) were included on each plate. Washings in PBS-T were made between the steps of the reaction. Further, peroxidase- labeled rabbit IgG anti-goat IgG (prepared as described by Wilson & Nakane 1978) was added in 100 μl/well at 1:3,000 dilution with PBS-T and incubated for 1 h at 37°C. Next, enzyme substrate consisting of 0.03% hydrogen peroxide and ortophenylenediamine (OPD - Merck, Germany) in 0.1M citrate- phosphate buffer, pH 5.0 was added (50 μl/ well) and incubated for 15 min at room temperature. The reaction was stopped by adding (25μl/well) 2N H_2SO_4 and the absorbance was determined in ELISA reader (Titertek Multiskan Plus, Flow Laboratories, USA) at 492 nm. The cut off was established as the mean absorbance values of negative controls plus three standard deviations. Serum samples that showed reactivity at dilution greater than 1:64 were considered positive samples (Nieto and Melendez, 1998).

3.7 Data handling and statistical analysis

Raw data were entered in Microsoft excel worksheet. Prevalence of toxoplasmosis was determined by using Epi-info statistical software version 6.0. The prevalence of local and dairy goats was compared by using Mantel-Haenszel chi-square test. The risk factors associated with human toxoplasmosis were summarized as frequencies of their occurrences and determination of risk ratio.

CHAPTER FOUR

4.0 RESULTS

4.1 Demographic characteristics and economic activities of interviewed farmers

The present study interviewed 43 adult farmers. Forty percent of dairy farmers were females and 60% were males. On the other hand all (100%) of the interviewed farmers with local animals were males. Livestock keeping and crop production was found to be the major economic activities in Mgeta Division. This was reported by all (100%) of the interviewed farmers. Dairy goats, pigs and chickens were the main livestock species kept at Mgeta. Other animals kept in the farms are presented in Table 2. Goats were largely managed under tethering method, where farmers used communal grazing spaces to feed the goats. Chickens were mainly local ecotypes and were kept under free range system. The goat houses were poorly constructed using woods in all farms that were visited. Through visual observation, it was concluded that the design of the houses was not good enough to allow thorough cleanliness (Plate 1). In addition, because of poor animal houses, cats had access to animals (Plate 1). In other areas where Maasai live; livestock farming was the main economic activity where goats, sheep and cattle were managed extensively through communal grazing. Few (12%) of farmers practiced mixed farming by subsidizing livestock farming with crop production.

Table 3: Farm animals that were found in the study areas

Animals	Management system	Location
Dairy goats	Teethering	Mgeta
Local goats	Free grazing	Maasai farms*
Local sheep	Free grazing	Maasai farms
Pigs	Indoor	Mgeta
Dogs	Free ranging	Mgeta and maasai farms
Cats	Free ranging	Mgeta and maasai farms
Rabbits	indoor	Mgeta
Chickens	Free ranging	Mgeta and maasai farms
Ducks	Free ranging	Mgeta

*Maasai farm is used to represent all farms other than Mgeta where local goats and

sheep were sampled.

Plate 1: Design of dairy goat houses in Mgeta.

Note the poorly constructed house and presence of a cat inside the house.

4.2 Prevalence of toxoplasmosis

The results of seroprevalence of toxoplasmosis are presented in Tables 3 and 4. Comparative values of seroprevalence between species of animal, sex and breeds are shown in Figure 4. All positive cases were IgG positive and there were no IgM positive cases. Table 4 shows that the overall prevalence of toxoplasmosis in small ruminants was 12% in goat and 27% in sheep respectively. Flock prevalence was 40% (10/25) in dairy goat and 56% (14/25) in local farms (p=0.26). Among dairy goats, prevalence of toxoplasmosis was higher in Mtamba village than other villages in Mgeta (Table 5). On the other hand, toxoplasmosis was not detected in Ndugutu and Nyasungu villages in Mgeta. With regard to local small ruminants, toxoplasmosis was more prevalent in Dakawa and Melela villages than other villages that have similar animals (Table 5). These differences of toxoplasmosis between locations were however not statistically significant (p>0.05).

Table 4: Prevalence of toxoplasmosis in goats and sheep

Animal	Number sampled	Number positive	Percentage positive
Dairy goats	150	16	10.7
Local goats	150	20	13.3
Local sheep	100	27	27.0
Overall sheep	100	27	27.0
Overall goats	300	36	12.0

Table 5: Prevalence of toxoplasmosis based on study locations

Village	Flock size	Sample size	Positive samples	Percentage positive
Nyandira	46	30	2	6.7
Mtamba	42	30	6	20.0
Nduguti	30	15	0	0.0
Kibagala	41	20	3	15.0
Tchenzema	53	37	4	10.8
Nyasungu	8	8	0	0.0
Vidigisi	18	10	1	10.0
Wami Dakawa	188	65	10	15.4
Melela	199	65	13	20.0
Dakawa	177	65	15	23.1
Makunganya	142	55	9	16.4

In Figure 4, it appears that sheep had significantly higher prevalence of toxoplasmosis than goats ($p=0.007$). On the other hand, occurrence of toxoplasmosis did not differ significantly between local and dairy goats. Sexual differences were not apparent when considering goat and sheep together. However when sheep and goat were considered separately, a significantly ($p<0.05$) higher prevalence was observed in female than male sheep (Figure 5). There were no significant differences between prevalence of toxoplasmosis in male and female goats ($p>0.05$). Age differences on toxoplasma infection are presented in Figure 6. The result indicates that adult goats and sheep has significantly higher prevalence of toxoplasmosis than young animals ($p<0.05$).
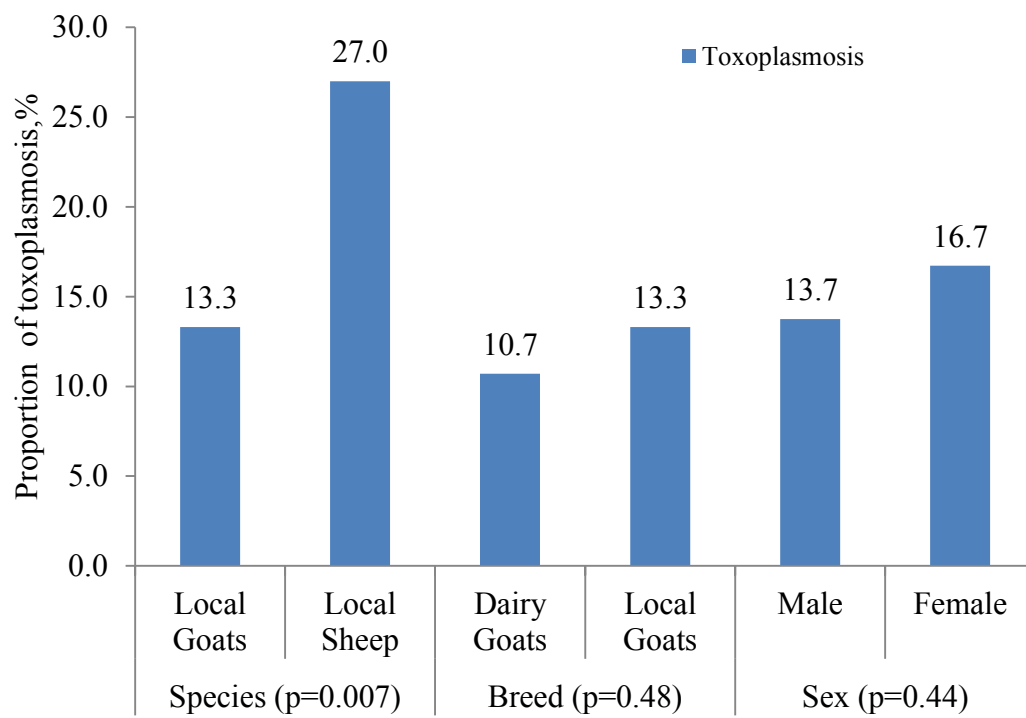
Figure 4: Differences on prevalence between species, breeds and sex of animals
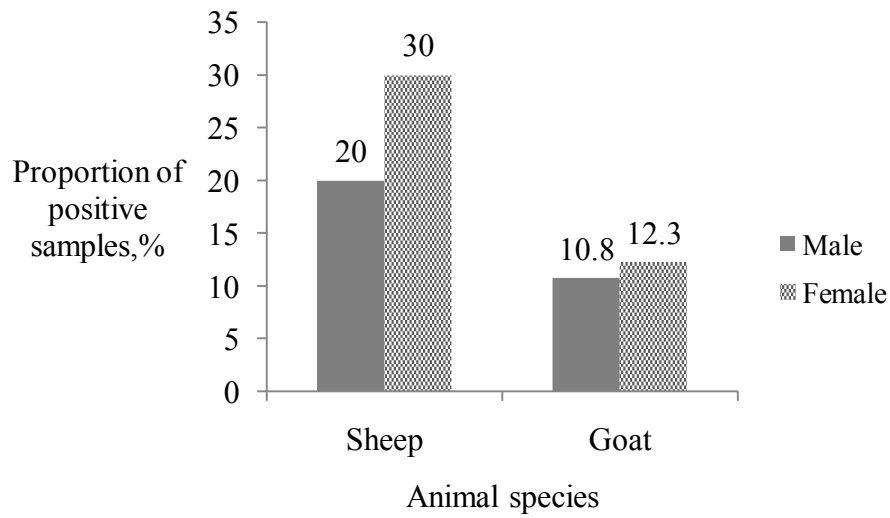
Figure 5: Comparative proportion of *Toxoplasma* infection between male and female animals stratified for sheep and goat
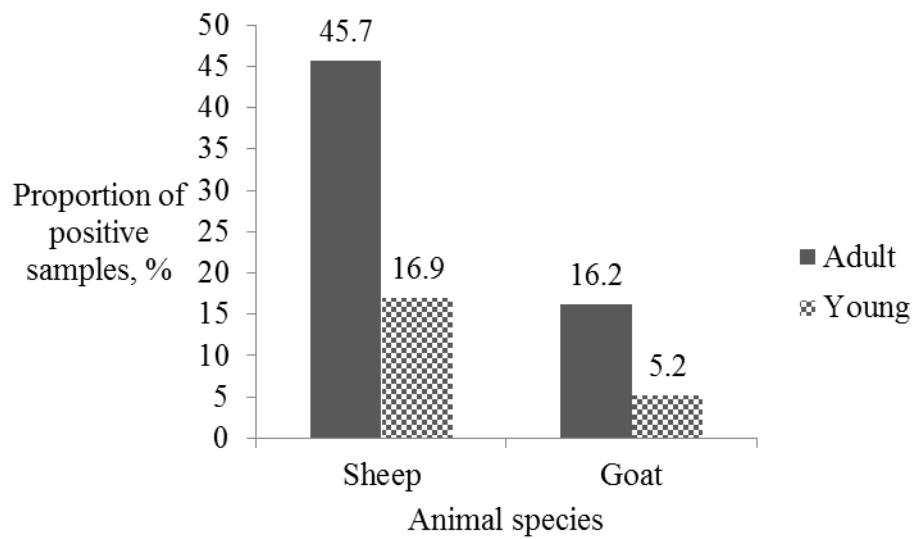
Figure 6: Comparative proportion of *Toxoplasma* infection between young and adult animals stratified for sheep and goat

4.3 Disease knowledge and risk factors for human toxoplasmosis

Table 6 shows various risk factors investigated in the present study. It shows that, of the 43 interviewed farmers, none (0%) of them was aware of toxoplasmosis. Presence of abortions in the farms was reported by 53.5% of farmers. However, although aborted animals had slightly higher prevalence of toxoplasmosis (Figure 7), this did not differ significantly from non-aborting animals, and thus no association could be established (p=0.82, RR=1.1). On the other hand, 44.2% of farmers were keeping cats. As it is seen in Table 5, majority of interviewed farmers did not know the sites where cats defecate, while few reported their cats to defecate either inside the house (21.1%) or in the gardens (10.5%). Presence of cats in the farms was strongly associated with occurrence of toxoplasmosis. This association is presented in Figure 7, where the risk of toxoplasmosis in farms with cats to farms without cats is shown to be 3.4 at p-value of 0.0000. Other identified risk factors were contact with animal wastes, consumption of raw milk and sharing water between animals and human beings.

Table 6: Awareness and risk factors for toxoplasmosis in animals and farmers

Knowledge/risk variable	Respondents	Response	Percentage response
Know toxoplasmosis	43	0	0.0
Abortion seen	43	23	53.5
Keep cats	43	19	44.2
Cat defecates on garden	19	2	10.5
Cat defecates inside	19	4	21.1
Don't know where cats defecates	19	13	68.4
Touch cat faces	19	0	0.0
Touch manure without protective gears	43	43	100.0
Consume raw milk	26	1	3.8
Consume raw/undercooked meat	17	17	100.0
Shared water source	43	7	16.3
Drinking unboiled water	43	0	0.0
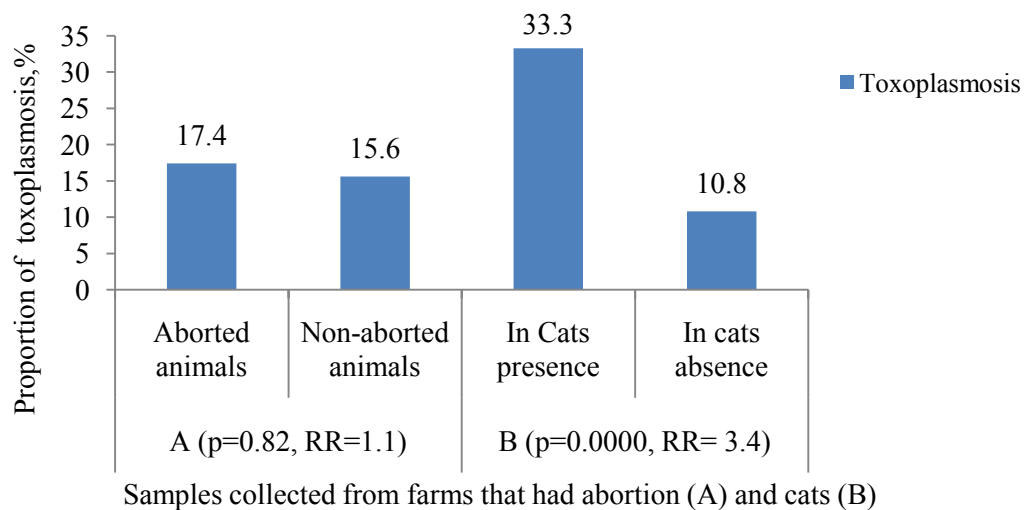
Figure 7: Occurrence of toxoplasmosis in farms that experienced abortions and farms that had cats.

CHAPTER FIVE

5.0 DISCUSSION

The present study has, for the first time established occurrence of toxoplasmosis in small ruminants in selected areas of Mvomero district in Morogoro, Tanzania. In all of the interviewed households, men were found to head the families. This is expected as is in most of African traditions. The interviewed households in Mgeta division were engaged in mixed farming, equally practicing livestock and crop production. As shown in the results; pigs, dairy goats and local chickens were the main livestock animals kept in Mgeta. The poorly constructed animal houses allow access of cats to animal houses and because of the close vicinity of houses of different farm animals there is direct communication of wastes from one house to another. Poor management of animals therefore creates favorable conditions for cross contamination and infections among animals and human beings. On the other hand, unlike in Mgeta, interviewed farmers in other locations were predominantly pastoralists, with large herds and flocks of indigenous cattle, sheep and goats. A substantial proportion of farmers reported the presence of cats around the farms (either through ownership or stray cats), which is very important as cats are reservoirs of toxoplasmosis for human and animals.

The prevalence of toxoplasmosis identified in the present study, which is 12% in goat and 27% in sheep seems far less than the estimated global average of 31% (Kamani *et al.*, 2010). The world seroprevalence of toxoplasmosis in goats and sheep has great variability and can range from 0-77% (Tenter *et al.*, 2000, Ahmad *et al.*,

2015). Variations could be contributed by climatic conditions, animal susceptibility, presence and interactions with multiple hosts and husbandry practices.

Nevertheless, however prevalence reported in this study is of significant public health importance that needs attention. The findings of this study compliment previous reports on toxoplasmosis in Northern Tanzania by Swai and Kaaya (2012), where the average prevalence of toxoplasmosis in goats was found to be 17%. In addition, it appears from the results that occurrence of toxoplasmosis is influenced by sex, species, breed and the age of the animals; where females, sheep, local breeds and older animals are more infected that the corresponding counterparts. These findings are in agreement with studies by Teshale *et al*. (2007), Kamani *et al*. (2001) and Swai and Kaaya (2012). For example Swai and Kaaya (2012) reported prevalence in local goats to be 14.3% compared to 12.1% in pure breeds. On the other hand, Teshale *et al*. (2007) reported a prevalence of 88.8% in older goats compared with 57.4% in young goats. Similarly, the same study reported a prevalence of 80.5% in females and 74.5% in males. Occurrence of toxoplasmosis in sheep is within the global prevalence, which range from 17 to 84% (Teshale *et al.,* 2007). It is however less than the prevalence of 69% reported in Great Britain in 2009 (Teshale *et al.,* 2007). As mentioned earlier, variations in occurrence of toxoplasmosis is influenced by geographical conditions and management factors.

Therefore it is tempting to confirm that, the incidence of toxoplasmosis is influenced by the animal factors mentioned above. The influence of sex and age on the occurrence of toxoplasmosis is probably linked to the cumulative exposure to the

parasite. Females are usually kept for longer time in the farms than males for breeding purposes. This contributes to their over-stay in the farm and hence more exposure to the parasite. On the other hand, older animals have more time in the farm to get more exposure to the parasite than young animals.

The study also indentified important risk factors for toxoplasmosis in small ruminants and farmers. This was evidenced by the significant association of cats with occurrence of *Toxoplasma* seropositive animals reported in the results of the present study, which clearly compliments the finding in human beings (Al-Hamdan and Mahdi, 1997). A previous report found that *Toxoplasma* antibodies were more prevalent in pregnant women with cats at home than in pregnant women who did not possess cats (Al-Hamdan and Mahdi, 1997). Presence of 44.2% of farmers that keep cats is considered to be a potential risk for toxoplasmosis in animals identified. It appears that keeping cats is a common practice among livestock keepers. This had previously been reported by Ogendi *et al*. (2013) in Kenya where about 45% of livestock farmers were keeping cats.

In some studies, the higher prevalence verified in goat herds was associated with the presence of a high-density population of cats, pets, rodents and humans, providing high availability of definitive and intermediate hosts for the parasite, thus increasing the chances of infection (Bisson *et al.* 2000; Cavalcante *et al.* 2008; Gondim *et al.* 1999). This also suggests that livestock farmers in the study area are at an increased risk to *Toxoplasma* infection. The risk of keeping cats in the farms is connected to their defecation behavior. As reported in the present study, most of cats (68%) do

defecate in sites unknown to farmers, while others defecated inside the farmers' house or on the gardens. Cats therefore stand a great chance of disseminating oocysts and contamination of household environment, farm and grazing areas. Similarly, farm cats have a major influence on the epidemiology of the disease due to their predatory habits and their diet, which includes wild birds, rodents and *Toxoplasma*-infected placentas and stillborn fetuses (Frenkel, 2000).

The other equally important risk practices for toxoplasmosis were poor knowledge, sharing water sources between animals and farmers, poor handling of manure, consumption of undercooked meat and feeding of cats with uncooked offal. Knowledge of disease is important for prevention of disease by farmers. Knowing how the disease is transmitted farmers can work out on available prevention methods and changing practices to avoid risks associated with the infection. Fecal contamination of water with oocysts from cats is important for both animal and human exposure to toxoplasmosis. In addition, farmers can unknowingly get into contact with oocyst in cats faces or handling of soil with unprotected hands that are then used for eating without washing hands. This was confirmed in this study where all of the interviewed farmers did not use gloves when handling farm activities or soil. Feeding cats on raw offal, as reported by some farmers in this study, is a notable infection risk factor for the cats, and by extension, for the farm household members. To prevent cats from becoming infected, they must be fed on well-cooked meat and, if possible, kept indoors to prevent them from hunting or scavenging (Dubey, 2004). In the present study, all of the farmers whose livestock had aborted handled the aborted material with bare hands and disposal was by burying. This exposes them to

the parasite directly and indirectly since cats can also pick up the infection by eating aborted material. The risk factors described in this study are comparable to the risk practices and factors described by Ogendi *et al*. (2013), which include presence of cats in the farm, poor disposal of cat feces, poor knowledge, lack of protective gears during handling of cat faeces, consumption of unboiled water and undercooked meat.

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATION

The results of the present study provide the following conclusive observations. The study confirms that toxoplasmosis is prevalent in small ruminants in Mvomero district, Morogoro. Cats and malpractices related to sharing water sources, consumption of unboiled water, poor disposal of wastes and aborted fetuses were identified as the most important risk factors for toxoplasmosis in the study area. Likewise knowledge of toxoplasmosis is low among livestock farmers. It is therefore important to create awareness and design knowledge package suited for livestock farmers, which will help them change behaviour and prevent risk practices related to *Toxoplasma* infection. In addition, existence of toxoplasmosis in animals in the study area is an indication of doing a study on farmers' side in order to establish whether or not famers are exposed to the infection, thus quantifying the burden and public health impact of disease.

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APPENDICES

Appendix 1: Survey questionnaire-English

TOXOPLASMOSIS RESEARCH

QUESTIONNAIRE FOR ASSESSING RISK FACTORS

Basic information

1. Date of interview…………………………………………………………………

2. Questionnaire number……………………………………………………………

3. Name of interviewer………………………………………………………………

4. Name of interviewee
   ……………………………………………..………………………………...

5. Residence
   …………………………………………………………………………

Respondents' personal information

6. Age ……………………………………………………………………

7. Sex ……………………………………………………………..………

8. Level of education
   ……………………………………………………………………………...

9. Family role (s)……………………………………………………. ……

10. Farming activities: (i) Livestock (ii) cropping (iii) Mixed farming (iv) vegetable garden

Questions regarding farm animals and vectors

11. Which animals do you keep? : (i) Goats (ii) Cattle (iii) Sheep (iv)Dogs (v)

Cats (vi) Chickens (vii)

Other…………………………………………………………………...

Animal	goats	cattle	sheep	dogs	cats	chickens	other
management							

-Type of management: (i) Free range (ii) Stall feeding (iii) intensive (iv) extensive

12. How many cats do you have (or were you having

previously)..

13. Do the cats have their own house?

....

14. Can you tell me where your cats defecate?..

15. Is there any stray cats visiting your site?..

16. Does the cat access the area where animals feed or graze? Yes/No

……………………….

17. Do these cats access the water sources where goats drink water?

Yes/No…………………

18. Do you also see rats (and other Rodents) roaming around your

house?...............................

Specific questions for goats

19. How many goats do you have?...

20. Is there any history of abortions in your herd? Yes/No……………

21. If yes, when did abortion occur?………………………...

22. How many times did abortion occur? ……………………...

Risk practices

23. Are you at one time involved in animal slaughtering activities?

   Yes/No………………….

24. Do you drink milk? Yes/No………………………………………….

25. Do you boil the milk before drinking? Yes/No…………………….

26. Mention types of meat you always eat: (i) beef, (ii) pork, (III) goat, (iv)

   sheep, (iv) chicken, (v) Other……………………………………………

27. Does the meat you consume cook thoroughly?

   Yes/no………………………………......

28. Have you ever eaten undercooked goat meat such as nyama choma? Yes/No

   ……………

29. Can you mention sources of domestic water you always use?

   (i)Tap water, (ii) River/stream, (iii) wells, (iv) boreholes, (v)

   other……………………....

30. How do you treat water before consumption?

   (i) Boiling, (ii) apply disinfectant (…………….), (iii) Not boiling/not treating

31. Do you think, animals such as cats, goats e.t.c. have access to water you

   consume? Yes/no………..

32. Is there any chance where cats defecate on household material, e.g. feeds

   (maize bran~sometimes cats defecate) Yes/no………………..have you ever

   observed?…….

33. Have you ever consumed unwashed fruits and raw vegetables?

Yes/no…………………..

34. Do you think, animals such as cats, goats e.t.c. have access to your vegetable garden or fruits you consume? Yes/no………..

35. Can you tell methods you use when you dispose animal wastes? During cropping/mixed farming…………………………………… or during doing cleaning home surroundings……………………………………….

36. Are you or any member of your family use protective gears when touching cat faeces or other animal wastes? Yes/no…………….mention the gear…………………..

37. Who is doing cleanliness, especially disposing animal wastes?

    (i) Father (ii) Mother (iii) Children (include sex)……………sex…………..

Disease knowledge

38. Are you aware of any disease, which can be transmitted from animals to human?........

39. Mention the diseases……………………………………………………

40. Regarding toxoplasmosis, do you know this disease? Yes/no.........................

41. Are you aware that toxoplasmosis can affect human? yes /no…………….

42. If yes, how the disease is transmitted from animals to human

43. Can you mention the clinical signs from a person suffering from toxoplasmosis? Yes/no……….

<u>THANK YOU FOR YOUR COOPERATION</u>

Appendix 2: Survey questionnaire-Swahili translated

DODOSO LA KUCHUNGUZA VIASHIRIA HATARISH VYA UGONJWA WA TOXOPLASMOSIS

Taarifa muhimu za jumla

44. Tarehe ya usaili……………………………………………………………

45. Namba ya dodoso….………………………………………………………

46. Jina la anayesaili……………………………………………………………

47. Jina la anayesiliwa …………………………………………..…………

48. Makazi …………………………………………………………………

Taaarifa za anayesailiwa

49. Umri …………………………………………………………………

50. Jinsia ……………………………………………………………...…

51. Kiwango cha elimu ……………………………………………..………

52. Majukumu katika kaya……………………………………………….
…………………………..

53. Kazi za shamba: (i) ufugaji (ii) kilimo cha mimea (iii) kilimo mseto (iv) bustani za mboga/matunda

Maswali kuhusu wanyama

1. Unafuga wanyama gani? : (i) mbuzi (ii)ng'ombe (iii) kondoo (iv)mbwa (v) paka (vi) kuku (vii) wengine………………………………………………

-njia ya ufugaji: (i) huru (ii) ndani kwa wakati (iii) ndani moja kwa moja (iv)

Mnyama	mbuzi	ng'ombe	kondoo	mbwa	paka	Kuku	wengine
njia ya ufugaji							

2. Una paka wangapi? (au ulikuwa napaka wangapi hapo zamani)...

3. Hawa paka wana kaa katika nyumba yao wenyewe?
....

4. Unaweza eleza ni wapi paka anajisaidia haja ndogo/kubwa?...

5. Je kuna paka wanaozurura hapa kwako kutoka maeneo mengine?..

6. je paka wanashirikiana sehemu ya kula/kunywa maji na wanyama wengine? ndiyo/hapana ……………………….

7. je paka wanashirikiana sehemu ya kunywa maji na mbuzi ? ndiyo/hapana …………………

8. Je kuna panya au wanyama wengine wadogodogo wanafika katika mazingira yako hapa?...............................

Maswali maalumu kuhusu mbuzi

9. Je una mbuzi wangapi?..

10. Je ulishakuwa na tatizo la mbuzi wako kutoa mimba? ndiyo/hapana……………………………………….

11. Kama jibu ni ndiyo, ni lini uliona muzi wako akitoa mimba? ……………………………………………………………...

12. Ni mara ngapi kwa wastani umeona mbuzi wako wakitoa mimba? ……………………………………………………...

Vitendo hatarishi vya maambukuzi

13. Je Unakunywa maziwa ya mbuzi……………………………………

14. Je maziwa unayokunywa yanachemshwa kabla ya kunywa………………

15. Je huwa unajihusisha na shughuli ya kuchinja wanyama? ndiyo/hapana …………………

16. Kwa kawaida huwa unakula nyama ya aina gani: (i) ng'ombe, (ii) nguruwe (III) mbuzi (iv) kondoo (iv) kuku (v) nyinginezo……………………………

17. Je nyama unayokula inapikwa na kuiva vizuri? ndiyo/hapana ……………………………….....

18. Ulishawahi kula nyama isiyoiva vizuri? ndiyo/hapana … nyama choma? ndiyo/hapana ……………

19. Maji unayotumia unayatoa wapi? (i)bombani, (ii) mtoni, (iii) bwawani, (iv) shimoni (v) nyinginezo……………………....

20. Maji unayotumia unayatibu vipi? (i) kuchemsha (ii) kuweka dawa (……………), (iii) siyafanyi chochote

21. Unadhani wanyama kama paka, mbuzi n.k wanafika katika maji unayotumia? ndiyo/hapana ………..

22. Je kuna uwezekano wowote paka akajisaidia katika vyakula ndani (mfano katika unga), ndiyo/hapana ……………….je ulisha wahi kumuona?…….

23. Ulisha wahi kula matunda yasiyooshwa ? ndiyo/hapana …………………..

24. Je kuna uwezekano wowote paka akajisaidia katika katika bustani ya mboga/matunda unalyokula?? ndiyo/hapana ………..

25. Je unaweza eleza njia unazotumia kutupa taka za vinyesi vya wanyama (wakati wa kilimo mseto)……………………………………………………au wakati wa kufanya usafi wa mazingira…………………………………………………………………… ….

26. Je wewe au mmoja wa familia yako mnavaa vifaa/nguo za kujikinga na uchafu wakati wa kutupa taka za vinyesi vya wanyama? ndiyo/hapana ……………..taja vifaa unavyotumia…………………..

27. Je ni nani anafanya usafi, hasa katika kutupa taka za vinyesi vya wanyama?
(ii) baba(ii) mama(iii) watoto (taja jinsia)… jinsia…………..

Ufahamu wa ugonjwa

28. Je unafahamu ugonjwa wowote unaoambukiwa kutoka kwa wanyama kwenda kwa wanadamu?........

29. Taja magonjwa unayojua………………………………………………………

30. Kuhusiana na toxoplasmosis, unaufahamu huu ugonjwa? ndiyo/hapana………………………

31. Je unafamu kwamba toxoplasmosis unaambukiza binadamu? ndiyo/hapana ……………….

32. Kama jibu ni ndiyo, je binadamu anaambukizwaje na wanayama?

33. Je unafamu dalili za ungonjwa wa toxoplasmosis? ndiyo/hapana ……….

34. Taja dalili hizo……………………………………………………………………… ……………………………………………………………………………………… ……………………………………………………………………………………… …………………………………………….

ASANTE KWA USHIRIKIANO WAKO