Questionnaire Survey on the Occurrence of Risk factors for *Toxoplasma gondii* infection amongst Farmers in Thika District, Kenya

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A survey was conducted to determine the occurrence of risk factors for Toxoplasma gondii infection amongst farmers in Thika District, Kenya. Interviews were conducted in a total of 385 households using a structured questionnaire. The water consumed at household level originated from taps (74.3%), rivers or streams (15.1%), wells (5.4%) and boreholes (5.2%). A number of households (46.8%) consumed water without boiling or applying any form of treatment. All respondents washed vegetables before cooking, whilst 99.0% washed fruits before eating. Boiled milk was preferred by 99.5% of the farmers. The majority (85.2%) consumed beef more often, whilst 1.6% consumed pork. The majority (98.7%) consumed thoroughly cooked meat. Meat was preserved by 17% of farmers. Only four farmers (1.2%) who practised mixed farming used gloves when handling livestock manure. Five farmers (1.6%) reported the occurrence of abortion in ruminants and pigs on their farms within the last two years before the study. Almost half (44.9%) of the households owned cats, which were kept mainly as pets (79.8%) and for deterring rodents (20.2%). The majority of households (91.3%) fed the cats on leftovers, whilst 8.1% fed cats with raw offal. Sixteen households (9.2%) provided housing for cats. Only five households (2.8%) had litter boxes, but none of the households with litter boxes used gloves when cleaning them out. Disposal of cat faeces was done mainly by women (55.5%). Only one farmer (0.3%) had some knowledge about toxoplasmosis, but was not aware of the transmission mechanism. The study highlights the need for public health education to raise awareness of risk factors for toxoplasmosis.

Introduction

Toxoplasmosis is a zoonotic disease caused by the protozoan parasite *Toxoplasma gondii*. The parasite is found in humans worldwide, under a variety of climatic and socio-economic conditions. Over the last three decades, *T. gondii* infection has emerged as one of the most common opportunistic infections associated with HIV and AIDS. It is also a major cause of mortality in AIDS patients in developing countries (Carruthers 2002). Approximately 30% – 65% of the world's population is estimated to have chronic *Toxoplasma* infection (Montoya & Liesenfeld 2004).

Toxoplasma gondii infection in humans is acquired in various ways. Undercooked pork poses the biggest risk to humans, with prevalence ranging between 10% – 30% in most countries (Dubey & Jones 2008). Studies show that 30% – 60% of infections in pregnant women are due to the consumption of undercooked contaminated meat (Jacquemard 2000). Ingestion of sporulated oocysts from cat litter or soil, contaminated unwashed or unpeeled vegetables and fruits, gardening activities or contaminated water is the main transmission mechanism (Dubey & Jones 2008). Flies, such as the common housefly *Musca domestica* and the oriental blowfly *Chrysomya megacephala*, are also capable of carrying viable *Toxoplasma* oocysts from cat faeces to food for one to two days (Wallace 1971). Congenital transmission occurs when a primary maternal infection is passed transplacentally to the foetus (Jacquemard 2000). Dubey and Jones (2008) reported that blood transfusion, organ transplant or laboratory accidents also pose possible infection risks.

The majority of toxoplasmosis cases in immunocompetent people are asymptomatic (Tenter, Heckeroth & Weiss 2000) or produce only mild symptoms, but may result in life-long infection with parasites located inside tissue cysts. 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, Hsu & Meissner 1994). Reactivation of latent infection leads to severe and life-threatening disease in immunocompromised individuals (Sell, Sander & Klingerebiel 2005; Sorrentino 2005). Toxoplasmosis is the most prevalent disorder affecting the brain in HIV-patients, causing toxoplasmic meningo-encephalitis (Luft & Remington 1992). It is the second most common AIDS-related opportunistic infection and occurs in 10% – 50% of patients with AIDS who are seropositive to *Toxoplasma* and have a low CD4+T lymphocyte count

(Luft & Remington 1992). Since the introduction of highly active antiretroviral therapy (HAART) in 1996, the incidence rates of toxoplasmic meningo-encephalitis have decreased in some regions of the world (Luft & Remington 1992). HAART, which helps to decrease the viral load and improve CD4+ T-cell counts, and prophylactic treatment against reactivation of latent *T. gondii* infections, have helped to decrease the incidence of toxoplasmic meningo-encephalitis (Sukthana 2006).

Toxoplasmosis is a major public health problem in most countries, including Kenya. A study in Kenya showed that 54% of HIV infected patients had *Toxoplasma*-specific IgG antibodies in contrast to 1% of the HIV negative group (Brindle *et al.* 1991). Sera from blood donors in four areas of Kenya showed an antibody prevalence of 54%. These reports, along with clinical ones (Lodenyo & Susan 2007), show that there is widespread distribution of *T. gondii* infection in Kenya. Statistics from the Kenya National Aids Strategic Plan for 2009/2010 indicate that the Thika district has one of the highest prevalence rates (9.1%) of HIV-related cases in the country, higher than the national prevalence (6.3%). Previous studies reported that 60% of the hospital beds were occupied by patients with HIV or AIDS related diseases (Nyambura 2009).

Risk factors associated with *T. gondii* infection in humans have not been investigated in most African countries, including Kenya. Thika is a district where all possible risk factors for toxoplasmosis are in abundance: keeping of livestock in both rural and urban environments and lack of functional sewage and waste disposal systems. This suggests that the *T. gondii* infection burden in the human population could be high. This study investigated the socio-economic conditions of farmers in the Thika District and possible risk factors for toxoplasmosis.

Information on the potential risk factors for *T. gondii* in the study area can therefore be valuable in enabling informed decision making by the relevant bodies involved in disease control. It will also empower farmers by giving them knowledge about how to curb transmission from animals to man.

Materials and Methods

Study area

Thika District; (1° 4′ 60 S 37° 4′ 60 E), is an administrative region located in the southern part of the Central Province of Kenya. The district is made up of six administrative divisions, namely Kamwangi, Gatundu, Kakuzi, Gatanga, Thika municipality and Ruiru. It covers 1960.2 km² and according to the 2009 Kenya Population and Housing census, has an estimated human population of 864,509. The district has a tropical climate with an annual rainfall ranging between 500 mm – 1500 mm (Government of Kenya 2005; Jaetzold & Schmidt 1983). The majority of farmers in the Thika district are smallholders, practicing mixed agriculture, including livestock production, food and cash crops. Urban farming is common in Thika and Ruiru municipalities and is

critical in providing milk, eggs and meat for the increasing urban population.

Selection of study sites and farms

A cross-sectional study was undertaken on farms in Thika District between June 2011 and August 2011. In order to calculate the sample size of farmers to be recruited into the study, an expected prevalence of 50% was considered to maximise the sample size, and a minimum confidence interval of 95% and statistical error of 5% were established. Thus, the working sample size (*n*) was as follows (Martin, Meek & Willeberg 1987):

$$n = \frac{Z^2 \frac{a}{2} (PQ)}{L^2}$$

$$n = \frac{1.96^2 (0.5) (1-0.5)}{0.05}$$

$$n = 385$$
[Eqn 1]

Where *n* is the maximum sample size required, Z is the Z score for a given confidence level, *P* is a known or estimated prevalence, Q = (1-P), and L is statistical error.

Multistage and systematic sampling methods were employed. Sampling points comprised the five sampling sites in Thika District (Thika, Ruiru, Gatundu, Kakuzi and Gatanga). Georeferenced maps used during the 2009 Kenya Population and Housing Census survey were obtained from the District offices so as to determine the distribution of households. In order to avoid spatial clustering of respondents, households were systematically chosen by initially randomising a starting point. A skip interval of nine households was used to choose the next household until the desired sample size of 385 was achieved.

Questionnaire survey

A structured questionnaire was prepared and pre-tested before administration to the selected farmer households. 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, consumption of raw vegetables or fruits, kitchen hygiene and personal hygiene habits, 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 (if the head of the household was unavailable). This was done using the Kiswahili language, which is common in the area. For farmers who were unable to understand Kiswahili, the research assistants administered the questionnaire in the Kikuyu language.

Data entry and analysis

All of the responses to the questionnaires were entered into a computer spread sheet, Microsoft Excel® (Microsoft

Corporation, USA) followed by descriptive analysis using SPSS Version 16.0 (SPSS, Inc., Chicago, IL, USA). Data with quantitative variables were expressed as the mean +/-standard deviation (+/- s.d.) and range, whereas qualitative variables were estimated and presented as frequencies and percentages. Percentages and the corresponding 95% confidence intervals (95% CI) were calculated using the method described by Collett (2002).

Results

Socio-demographic characteristics of farmers

The general socio-demographic characteristics of farmers are shown in Table 1. Overall, 143 males (37.1%, 95% CI: 32.3-42.2) and 242 females (62.9%, 95% CI: 57.8-67.7) were interviewed. Male-headed households (87.5%, 95% CI: 83.8-90.7) were more common than those headed by females (12.5%, 95% CI: 9.3-16.2). The mean age of the farmers was 38.7 years (s.d. 14.1 years; range 20-88). The mean number of children per household was 3.2 (s.d. 1.9; range 0-11). However, 20 farmers (5.2%, 95% CI: 3.2-7.9) could not remember their age at the time of the study. Most farmers (73.3%, 95% CI: 68.2-78.0) had acquired land through inheritance, whilst (23.1%, 95% CI: 18.7-28.0) had bought their land. With respect to house size, 238 farmers (61.8%, 95% CI: 56.8-66.7) had houses with two to three rooms, 113 farmers (29.4%, 95% CI: 24.8-34.2) had houses with more than three rooms, whilst 34 farmers (8.8%, 95% CI: 6.2-12.1) lived in one-roomed houses.

Potential risk factors for toxoplasmosis

As shown in Table 2, a total of 173 households owned cats (44.9%). The majority, (91.3%, 95% CI: 86.1-95.1) reported feeding their cats with leftovers, whilst 14 farmers (8.1%, 95% CI: 4.5-13.2) reported feeding their cats with raw offal. Only one farmer (0.6%, 95% CI: 0.0-3.2) indicated that the cat was not given any food and it was assumed that the cat lived off rodents. Sixteen households (9.2%) provided housing for the cats, whilst the other cats roamed free. With regard to litter boxes, only five households (2.9%) reported having litter boxes for their cats. None of those with litter boxes used gloves when cleaning them out. All of the cat owners disposed of cat litter on the farm. In 96 of the households, the responsibility of disposing of cat faeces was left to women (55.5%) (see Table 2). However, 66 cat owners (38.2%) reported that they were not aware of the site where the cats defaecated. Since most cats were not housed cats, they roamed in the grazing pastures, barns, pens and where domestic animal feed was stored.

A considerable proportion of farmers 286 (74.3%) reported tap water as their main source of water, whilst 58 (15.1%) obtained water from rivers or streams, 21 (5.5%) obtained water from wells and 20 (5.2%) used water from boreholes. With regard to water treatment or boiling before consumption, 205 farmers (53.2%) boiled or treated the water (see Table 2). All of the farmers interviewed ate raw food that included carrots, sweet potatoes and fruits. Of the 381 farmers who responded that they ate raw food, the majority (99.0%, 95% CI: 97.4–99.7)

reported that they washed fruits before eating. All of the 385 farmers said that they washed vegetables before cooking. The majority of farmers who engaged in mixed farming 326/331 (98.5%) used livestock manure for cropping. Of these 326, 322 (98.8%) handled the manure with bare hands, whilst only 4 (1.2%) used gloves. This is an important factor, as cat faeces might get mixed with livestock manure, thus posing a high risk of transmission of *T. gondii* oocysts.

TABLE 1: Socio-demographic characteristics of respondents.

Proportion of	Variable	Р	Proportion of farmers			
farmers		n^{\dagger}	%	95%	CI	
Age group (years)	20–29	126	34.5	29.6	39.6	
	30–39	95	26.0	21.6	30.8	
	40–49	69	18.9	15.0	23.3	
	50–59	28	7.7	5.2	10.9	
	60–69	34	9.3	6.5	12.8	
	\geq 70	13	3.6	6.5	1.9	
Education	None	28	7.4	5.0	10.5	
	Primary	134	35.3	30.5	40.3	
	Secondary	177	46.6	41.5	51.7	
	Tertiary	41	10.8	7.9	14.4	
Occupation	Full time farmer	224	58.2	53.1	63.2	
	Formal employment	53	13.8	10.5	17.6	
	Business person	26	6.8	4.5	9.7	
	Other	82	21.3	17.3	25.7	
Average income	Low (\leq \$ 37.5)	62	16.8	13.2	21.1	
	Medium (\$ 38 – \$ 124)	204	55.4	50.2	60.6	
	High (≤\$ 125)	102	27.7	23.2	34.2	
Residence	Kakuzi	112	29.1	24.6	34.0	
	Gatundu	85	22.1	18.0	26.	
	Gatanga	66	17.1	13.5	6	
	Thika	65	16.9	13.3	21.3	
	Ruiru	57	14.8	11.4	21.0	

 $\dagger,$ The total for each category may not equal the overall sample size because of nonresponse CI, Confidence Interval.

TABLE	2:	Potential	risk	factors	for	toxop	lasmo	osis
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Variable	Р	Proportion of farmers				
	n	%	95%	CI		
Reason for keeping cat	35	79.8	73.0	85.5		
	138	20.2	14.5	27.0		
Housing for cat	16	9.2	5.4	85.5		
	157	90.8	85.4	27.0		
Litter boxes	5	2.9	0.9	6.6		
	168	97.1	85.4	94.6		
Responsibility for disposing cat litter	96	55.5	47.8	63.0		
	4	2.3	0.6	5.8		
	7	4.0	1.6	8.2		
	66	38.2	30.9	45.8		
Household water source	286	55.5	69.6	78.6		
	58	2.3	11.6	19.0		
	21	4.0	3.4	8.2		
	20	38.2	3.2	7.9		
Boiling or Treatment of water	205	53.2	48.1	58.3		
	180	46.8	41	51.9		
Milk consumption	383	0.5	98.1	100.0		
	2	99.5	0.0	1.8		
Beef consumption	374	98.	96.9	99.6		
	5	7	0.4	3.1		
Manure handling	322	98.8	96.9	99.6		
	4	1.2	0.3	3.1		

CI, Confidence Interval.

Livestock reared by the farmers included cattle, sheep, goats, pigs, and poultry (see Table 3). With respect to cattle rearing systems, 154 out of 229 farmers (67.2%, 95% CI: 60.8-73.3) reported using zero grazing, whilst 36 (15.7%, 95% CI: 11.3-21.1) reported using free range rearing systems. Of the 43 farmers who reared sheep, 17 (39.5%, 95% CI: 25.0-56.0) reported using free range systems, whilst 11 (25.6%, 95% CI: 13.5-41.2) used tethering. Out of the 164 farmers who kept goats, 67 (40.9%, 95% CI: 33.2-48.8) indicated using zero grazing as the rearing system, whilst 41 (25.0%, 95% CI: 18.8-32.3) reported using tethering. With respect to the rearing systems for chickens, 165 out of 275 respondents (60.0%, 95% CI: 53.9-65.8) reported using free range, whilst 110 (40.0%, 95% CI: 34.2-46.1) reported using housed systems. All of the eight respondents who reared pigs had enclosed systems. Out of the 348 farmers who kept livestock, 332 (95.4%, 95% CI: 92.6-97.3) reported having regular access to veterinary services.

Of the 315 respondents with large livestock (cattle, goats, sheep, pigs), 5 (1.6%, 95% CI: 0.5–3.7) reported a history of abortion in the last two years before this study was conducted. All of the farmers whose livestock had aborted did not wear gloves when handling the aborted foetuses and foetal membranes. The aborted material was disposed of by burying.

At a household level, 43 farmers (11.2%, 95% CI: 8.2–14.7), had household member(s) with immunodeficiency disease(s), mainly HIV-related disease. Sixteen farmers (4.2%, 95% CI: 2.4–6.7) reported a history of congenital disease(s) in the household, for example: children who experienced seizures or fits. Moreover, 11 farmers (2.9%, 95% CI: 1.4–5.1) reported having a household member with a psychiatric disorder manifested by loss of memory, and in some cases the affected persons were reported to be violent. Only one farmer reported having some knowledge of toxoplasmosis because his pregnant wife had been ill and the doctors had advised her to stay away from cats, but he was not aware of the transmission mechanism.

Discussion

This study is the first to report the potential risk factors for *T. gondii* infection in farmers in Kenya. As in other African societies, in this study, men head most of the households (Lupala 2002). Most farmers had resided on their farms for prolonged periods of more than ten years. This suggests that there is some stability in the environmental exposure to any parasite over extended periods of time. A considerable

TABLE 3: Livestock reared b	y farmers within	the study sites.
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Livestock reared	Number of farmers	Mean number of animals	s.d.	Range
Chicken	275	16.0	45.5	1-400
Cattle	229	2.2	1.7	1-14
Goats	164	3.2	2.5	1-14
Sheep	43	2.8	1.7	1-10
Pigs	8	7.3	9.5	1-30

s.d., standard deviation.

proportion of farmers had an educational level ranging from primary to tertiary, which might make it easier to educate them on improved household and farm management practices to prevent transmission from livestock to humans.

In the present study, a substantial proportion of farmers report the presence of cats around the farms, which is very important, as cats are reservoirs for animal and human toxoplasmosis (Dubey 1995). The presence of cats has been considered a risk factor for toxoplasmosis in goats (Steven et al. 2000). In some studies, the higher prevalence verified in goat herds was associated with the presence of a highdensity 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 toxoplasmosis infection. Cats in the study area could access grazing pastures, barns, pens and feed stores, defaecate and cause oocyst transmission to livestock. In general, 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 foetuses (Frenkel 1990; Steven et al. 2000). A number of farmers report free range as the rearing system for their livestock. Freerange rearing systems permit contact between livestock and oocysts excreted in the faeces of feral or domestic felines, thus increasing the probability of infection.

Exposure to cats' faeces by owning cat(s) and/or having cats in the immediate surroundings and, in addition, by handling cats' sandboxes containing their faeces, has been found to be associated with Toxoplasma infection (Dubey & Beattie 1988). In this study, the responsibility of the disposal of cat faeces or cleaning of litter boxes in most households is left to women. A previous study found that Toxoplasma antibodies were more prevalent in pregnant women with cats at home than in pregnant women who did not possess cats (Al-Hamdani & Mahdi 1997). 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. In Brazil and Mexico, cats are fed leftovers and raw viscera, which have been identified as risk factors for human T. gondii infection. Moreover, cats are rarely trained to defaecate in litter boxes, making the contaminated environment a major risk factor in the human population (Cavalcante et al. 2008). 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 & Lappin 2006). 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.

In this study, a considerable proportion of households consume water without boiling or applying any form of treatment to it. There have been reported cases of outbreaks in some areas that have implicated drinking unfiltered water with oocyst contamination (Bahia-Oliveira et al. 2003; Isaac-Renton et al. 1998). Consumption of raw vegetables is common amongst the surveyed farmers, and in some cases is accompanied by lack of washing or peeling of this type of food. Such practices could be a major source of toxoplasmosis (Kapperud et al. 1996). Consumption of undercooked meat is also reported by some of the farmers. Classically, consumption of undercooked meat, particularly pork and lamb, has been cited as the major risk factor for human T. gondii infection (Dubey & Jones 2008). However, some studies have demonstrated that the parasite may remain viable and virulent for a period of over one year in cattle, suggesting that the risk of transmission via beef products should not be underestimated (Dubey & Thulliez 1994). Improved animal husbandry practices as well as increased awareness of the risks of consuming undercooked meat have resulted in a decreased prevalence of toxoplasmosis worldwide (Tenter, Heckeroth & Weiss 2000) This should be implemented in the study area through public and veterinary health awareness measures from the relevant stakeholders within the government.

A number of factors are associated with the acquisition of T. gondii oocysts by animals (Kapperud et al. 1996; Tenter, Heckeroth & Weiss 2000), such as hygienic standards in breeding, density of cats and environmental conditions. In this study, the majority of the farmers practise mixed farming and the use of manure for crops is reported by almost all of the farmers. An important advantage of using animal manure is its relative availability, low cost and its improvement in urban food production, thereby increasing access to food by the urban population, since rapid growth and urbanisation have negative effects on food security (Baumgartner & Belevi 2001). There is also a high probability of livestock manure mixing with cat faeces, considering that a large proportion of farmers did not know where their cats defaecated and all farmers with cat litter boxes disposed of the litter on the farm. In the present study, most farmers kept free range chickens and studies have shown that the prevalence of toxoplasmosis in free-ranging chickens as intermediate hosts of T. gondii is a good indicator of environmental contamination (Dubey & Beattie 1988).

Conclusions

The potential risk factors for transmission of toxoplasmosis identified in the study area include the keeping of freerange cats, improper handling and disposal of cat faeces and livestock manure, and consumption of untreated water. This suggests that farmers from the Thika District are at increased risk for *T. gondii* infection. However, the lack of awareness about the disease suggests that toxoplasmosis is a neglected disease in Kenya. Further epidemiological investigations to determine the occurrence of toxoplasmosis in livestock and humans in the study area are needed. These findings will be helpful in providing more information to enable informed decision making about disease control and to empower farmers with knowledge to curb transmission from animals to man. Given the numerous challenges that are faced in the management of health problems in Kenya, the findings of this study could be of considerable use for public and veterinary health. However, the problem needs concerted effort from various ministries within the government, notably the Ministries of Public Health & Sanitation and Livestock Development. Health education of the populace is an important intervention, especially for farmers. This can be conducted during the frequent *barazas* (meetings) held by veterinary personnel and farmers. The farmers should be informed about better animal rearing and production methods designed to reduce *T. gondii* transmission. Moreover, education programmes for high-risk groups such as pregnant mothers and immunodeficient patients should be created through the district hospitals and health centres.

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Competing interests

The authors declare that they have no financial or personal relationship(s) that may have inappropriately influenced them in writing this article.

Authors' contributions

S.M.K. (Jomo Kenyatta University of Agriculture and Technology, JKUAT) was the project leader, E. O.(JKUAT), N.W.M. (JKUAT), J.M.K. (JKUAT), M.N. (Institute of Primate Research) and G.G.M. (Kenya Medical Research Institute) were responsible for project design and implementation. All authors participated in the writing of the manuscript.

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