

Full Length Research Paper

Seroepidemiology of *Toxoplasma gondii* infection among slaughtered pigs, cattle and goats for human consumption in Bobo-Dioulasso, Burkina Faso

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Received 23 April 2017; Accepted 19 June, 2017

Toxoplasmosis is a major foodborne infectious disease with substantial adverse impact on population health and economy. Human infection is usually secondary to the consumption of contaminated raw or undercooked meat. Recent studies have reported a high prevalence of the infection in slaughterhouse animals in sub-Saharan Africa but few data exist for Burkina Faso. The aim of this study was to assess the prevalence of *Toxoplasma gondii* infection in animal from Bobo-Dioulasso, Burkina Faso. A total of 962 animal (including 423 pigs, 197 cattle and 342 goats) blood samples were collected in slaughterhouses in Bobo-Dioulasso between August 2013 and May 2014. Serum samples were tested for *T. gondii* antibody detection using the modified agglutination test (MAT). The overall seroprevalence of *T. gondii* was 28.8% in pigs, 13.2% in cattle and 34.8% in goats. Females animals were more infected than males. More than 60% of older animals (higher than 5 year-old animals) were infected. The study reported a high seroprevalence of *T. gondii* infection in pigs, cattle, and goats in Bobo-Dioulasso and is, therefore, of public health concern. The consumption of raw or undercooked meat should be regarded as an important risk factor for *T. gondii* infection in the study area. However further studies are needed to design appropriate control measures.

Key words: Pigs, cattle, goats, seroprevalence, *Toxoplasma gondii*, Bobo-Dioulasso, Burkina Faso.

INTRODUCTION

Toxoplasmosis is an important food-borne parasitic disease caused by the protozoan parasite *Toxoplasma*

gondii that causes severe illness in immunocompromised individuals. The infection is found globally, affecting

almost one-third of the human population (Dubey and Jones, 2008). In human, the mode of transmission varies depending on culture behaviors, environmental factors, eating habits and/or religious practices (Dubey and Jones, 2008). The consumption of raw or undercooked meat containing viable cysts represents a major route of human contamination (Dubey, 2010, 2013; Pereira et al., 2010). *T. gondii* infection has a significant impact on animal production particularly in goats and pigs (Dubey, 2009, 2013). In fact, most infections among these animals are asymptomatic, but however can seriously affect their health. The clinical signs vary considerably and include abortions, stillbirths and fetal abnormalities (Kim et al., 2009; Dubey, 2009, 2010).

Consequently; animal reproductive performance is affected leading to heavy economic losses for livestock producers (Mahboub et al., 2013). In addition, livestock products represent an important source of nutrition around the world (Dubey, 1986, 2009; Chikweto et al., 2011).

Studies have shown that pigs, cattle and goats are at higher risk of *T. gondii* infection and the reported prevalence in slaughtered animals is higher in pigs, goats and sheep, than in cattle, and varies worldwide (Dubey, 1986; Gharekhani, 2013; Hosein et al., 2016). However, only very few studies have been conducted in Burkina Faso on the question and very little information is known about the prevalence of *T. gondii* infection in livestock in Bobo-Dioulasso (Bamba et al., 2012, 2013, 2016).

Traditional procedures of meat quality control rely on clinical examination mainly visual inspection, which leads to lot of misdiagnoses. Serological testing is considered to be an appropriate method for the diagnosis in slaughtered animals. The aim of this study was to investigate the seroprevalence of *T. gondii* infection in slaughtered pigs, cattle, and goats in Bobo-Dioulasso, Burkina Faso, using MAT.

MATERIALS AND METHODS

Study area

This study was carried out in Bobo-Dioulasso, the second largest city located in the southwest part of Burkina Faso, a sahelian landlocked country of 274,000 km² in West Africa. The country is strongly dependent on the vagaries of the climate and its economy is based on agriculture and livestock. The region of Bobo-Dioulasso has an average annual rainfall of 1000 to 1300 mm, with a temperature ranging from 16 to 45°C. These conditions are necessary for sporulation and survival of *T. gondii* oocysts when excreted with cat feces in the external environment (Dubey, 1990; Yilmaz, 1972).

Study designs and sample size

A cross-sectional study was carried out from August 2013 to May 2014 in commercial slaughterhouses across the city of Bobo-Dioulasso. Pigs, cattle, and goats of both sex and different age groups were included in the study. Blood samples were collected during animal bleeding and serum samples stored at -20°C till analysis in November, 2015. In the absence of data on *T. gondii* infection in pigs in Burkina Faso the required sample size was calculated by assuming 50% infection prevalence in pigs and goats and 14% in cattle (Bamba et al., 2013). With an expected prevalence (P), a 95% confidence interval (Z score of 1.96), and a precision (d) of 5%, the sample size was calculated as $N = (Z)^2 \frac{2P}{(1-P)d^2}$. The total sample size (N) was 423 pigs, 197 cattle and 341 goats.

Sample collection

Blood samples were aseptically collected from ear vein (pigs), coccygeal (cattle) and jugular (goats) venipuncture using sterile vacuum tubes without anticoagulant, the blood samples were labeled and immediately stored in ice boxes with ice packs and transferred to the laboratory of Institute for Research in Health Sciences in Bobo-Dioulasso. Samples were centrifuged at 3200 RPM for 10 min and the separated sera were transferred to Eppendorf tubes and kept at -20°C until analysis.

Serological test

The modified agglutination test (MAT) was used to detect *T. gondii* antibodies in animal serum samples. *T. gondii* tachyzoites were fixed using formalin for the quantitative assessment of IgG. The mercaptoethanol was used to inhibit the IgM-like antibodies that interfere with the assay specificity. All sera were serially diluted to 1:25, 1:50, 1:100, 1:1600 and 1: 3200 before testing. Positive sample antibodies titers threshold was set at 25 (Dubey and Desmonts, 1987).

Data analysis

The EpiData software version 3.1 and the Epi Info software version 6.04 were used for data entry and analysis respectively. The Chi² test was used for statistical comparison and a *p* value <0.05 was considered statistically significant. The seroprevalence of *T. gondii* infection was assessed regarding the animal age and gender. The national ethics committee of health research has approved the study (protocol 07/2013) before the implementation of the study related activities.

RESULTS AND DISCUSSION

Overall seroprevalence

A total of 962 animals including 423 pigs, 197 cattle and 342, goats were assessed and serum samples collected

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Table 1. Seroprevalence of *Toxoplasma gondii* infection in pigs, cattle and goats from Bobo-Dioulasso as determined by the MAT.

Animal	No. tested	No. positive	Seroprevalence (%)	CI 95%
Pigs	423	122	28.8	24.6 - 33.3
Cattle	197	26	13.9	8.9 - 18.4
Goat	342	119	34.8	29.8 -39.9

for IgG anti-*T. gondii* analysis. Two hundred and sixty seven animals composed of 122 pigs, 26 cattles and 119 goats respectively were positive.

The overall seroprevalence of anti-*Toxoplasma gondii* IgG antibodies in animals studied was 27.7% (95%CI: 29.4 to 30.6%) using MAT. Regarding the technical approach, MAT was chosen for the serological assay because of its good sensitivity and specificity for the diagnosis of latent infections compared to other serological tests (Dubey et al., 1995). Previous studies conducted in slaughtered animals in Bobo-Dioulasso have reported a seroprevalence of *T. gondii* antibodies of 58% in sheep (Bamba et al., 2012), and 14.3% in cattle (Bamba et al., 2013) using high direct sensitivity agglutination assay (HDSA) analogous to the modified agglutination test (MAT). Moreover, previous findings have shown that the MAT is a specific technique for searching *T. gondii* antibodies in pigs and no cross-reaction with other parasites was reported when using this test (Dubey, 1997).

The seroprevalence of 28.8% (95%CI: 24.6 - 33.3%) in pigs, 13.2% (95%CI: 8.9 - 18.4%) in cattle and 34.8% (95%CI: 29.8 - 39.9%) in goats were reported. Comparing the seroprevalence rate in each group, it appears to be lower in cattles than pigs and in goats (Table 1). This might be due to the difference in feeding habits of these animals. In fact, goats are more likely to be infected from the pasture as they graze close to the ground than cattle which prefer browsing. In other hand, pigs could be contaminated by *T. gondii* oocysts eliminated with the cat faeces in the environment (soil, grass, and water). These finding show that *T. gondii* is present among these animals slaughtered in Bobo-Dioulasso depicting the widespread environmental contamination by *T. gondii* oocysts in this country.

The current seroprevalence (28.8%) indicate the major role of pigs as intermediate hosts of *T. gondii*. The high seroprevalence of *T. gondii* in the present study might be associated with farm management systems and access to free roaming cats on pig farms. Furthermore, the prevalence was 13.2% (Table 1) in cattle group using MAT. According to previous studies, *T. gondii* has been rarely isolated from naturally infected cattle (Dubey et al., 1986; Dubey, 2010). Indeed, calves and cows infected with high doses of oocysts by oral inoculation indicate that *T. gondii* is eliminated or greatly reduced in cattle

tissues, some animals being negative during infection (Dubey and Thulliez, 1993). Therefore, cattle are considered a poor intermediate host of *T. gondii*.

Otherwise, our findings noted that over 30% of goats from Bobo-Dioulasso were seropositive for *T. gondii* antibodies (Table 1). Toxoplasmosis is a significant cause of reproductive failure in small ruminants such as goats. However, there is no available data on *T. gondii*-associated abortions in Burkina Faso. The high prevalence of *T. gondii* antibodies among goats in our study (34.8%) shows high levels of exposure to the parasite. In addition, it is known that tissue cysts persist throughout the host life (Dubey 1998). Therefore, this could be a potential risk to public health. However, in Burkina Faso, meat is consumed cooked or overcooked usually a common practice which minimizes the risk of *T. gondii* transmission.

Characteristics of animals

According to the sex, a predominance of female animals in each animal group was noted in this study (Table 2). The male to female sex ratio was 0.9 (197/226), 0.7 (86/111) and 0.6 (128/214) respectively for the pigs, cattle and goats (Table 2). This would be linked to the religious practices in Burkina Faso that make the majority of the male animals kept in the farms, sold, slaughtered or sacrificed outside the slaughterhouse during religious or traditional ceremonies. This justifies the high proportion of the female animals on slaughter chains in our study.

Table 3 shows age group repartition of animals with a predominance of older animal (5 years old) in all groups. The average age was 2.6 years (± 1.3) for pigs, 2.1 years (± 1.1) for cattle and 1.7 years (± 0.12) years for goats. The animals more than 5 years old were the most represented (> 60%) in each group (Table 3). This observation could be justified by the fact that the consumption of piglet, calf, and little goat is not part of culinary habits in Burkina Faso.

Serological data according to characteristics of animals

According to animal sex, females were more infected

Table 2. Seroprevalence of *T. gondii* infection regarding age and MAT titer.

Animal	Age (Year)	No. tested	No. positive (%)	No. positive with MAT titers of							
				25	50	100	200	400	800	1600	3.200
Pigs	< 2	184	33 (17.9)	17	4	5	5	1	0	1	0
	3-5	218	76 (34.8)	11	26	7	12	6	11	2	1
	> 5	21	13 (61.9)	2	1	2	1	1	4	2	0
Cattle	< 2	31	7 (22.6)	5	2	0	0	0	0	0	0
	3-5	154	11(7.1)	8	3	0	0	0	0	0	0
	> 5	12	8 (66.7)	6	2	0	0	0	0	0	0
Goats	< 2	257	83 (32.3)	17	13	16	15	13	7	2	0
	3-5	58	17 (29.1)	2	5	3	4	1	1	1	0
	> 5	27	19 (70.4)	4	2	3	1	6	1	2	0

Male vs female Pigs: Fisher's exact test: The two-tailed P value equals $P = < 0.0001$ was considered to be statistically significant; Male vs female Cattle: Fisher's exact test: The two-tailed P value equals $P = 0.065$ was considered not statistically significant. Male vs Female cattle: Male vs Female goats: Fisher's exact test: The two-tailed P value equals $P = < 0.0001$ was considered to be statistically significant.

Table 3. Seroprevalence of *T. gondii* infection in pigs, cattle and goats from Bobo-Dioulasso (Burkina Faso) according to sex.

Pigs					Cattle				Goats			
Sex	No. tested (%)	No. positive (N=122)	Sero-prevalence	P value	No. tested (%)	No. positive (N=26)	Sero-prevalence	P value	No. tested (%)	No. positive (N=119)	Sero-prevalence	P value
Male	197 (46.6%)	27	13.7% CI 95% (9.4-19.1%)	$P = < 0.0001$	86 (43.7%)	7	8.1% CI 95% (3.6-15.4%)	$P = 0.065$	128 (37.4)	22	17.2% CI 95% (11.3-24.4%)	$P = < 0.0001$
Female	226 (53.4%)	95	42.1% CI 95% (35.7-48.5%)		111 (56.3%)	19	17.11% CI 95% 10.9%-24.9%		214 (62.6%)	97	45.3% CI 95% 37.8%-51.1%	

than male. About 42.1% (CI 95%: 35.7-48.5%) of female pigs, and 45.3% (CI 95%: 37.8-51.1%) female goats were infected with p value $P = < 0.0001$ in each group (Table 2). The high susceptibility of female animals reported in the current study corroborates with previous study reports and is related to a higher female animal's

susceptibility to protozoan infections (Alexander and Stinson, 1988).

In addition, the seroprevalence of *T. gondii* increased according to the age with more than 60% of older animals infected in each group (Table 3). These findings suggest postnatal acquisition of *T. gondii*.

Regarding the level of dilution, positive MAT tests at the titers of 1:200 or higher were reported in 21.3% of pig and 45.4% of goats. None of the cattle serum samples was positive at the 1:200 dilutions (Table 3). Our results noted that none of the cattle was positive in 1:200 serum dilutions using MAT assay. The low MAT titers among

cattle indicate not the persistent infection but an exposure of *T. gondii*.

Furthermore, authors suggest that a titer of 1:25 in the MAT is considered specific for the detection of *T. gondii* in pigs, goats, sheep, and other livestock (Dubey, 2010), unlike cattle.

Conclusion

An epidemiological study was carried out to evaluate the seroprevalence of *T. gondii* in pigs, cattle, and goats slaughtered for human consumption in Bobo - Dioulasso. In Burkina Faso, livestock represents the second most important source of revenue in the country's primary sector.

Breeding in the study area is generally characterized by a traditional and extensive management system. Majority of slaughtered animals are destined for local human consumption. This study reported a high seroprevalence of *T. gondii* in pigs, cattle, and goats in Bobo-Dioulasso, and is, therefore, of public health concern. The consumption of raw or undercooked meat should be regarded as an important source of infection to people in the study area. However further studies are needed to design appropriate control strategies in Burkina Faso. Moreover, consumer knowledge should be strengthened in order to reduce the impact of the disease.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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