

Sero-prevalence and associated risk factors of *Toxoplasma gondii* infection among pregnant women attending Omdurman friendship hospital, Omdurman

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Abstract

Background: Toxoplasmosis in pregnancy is associated with spontaneous abortions, low birth weight babies, congenital deformities and intrauterine deaths. In many developed countries, pregnant mothers are screened for infection with *Toxoplasma gondii*, the causative parasite, and treatment is offered early in order to prevent these complications. In most developing countries, including Khartoum, the burden and risk factors of *T. gondii* infection among pregnant women, thus making it difficult to plan and implement control measures.

Study Objectives: This study aimed at determining the prevalence of *T. gondii* infections and their risk factors among pregnant women attending antenatal care clinics in Kigali, Rwanda.

Methods: This was a cross-sectional descriptive study involving 384 pregnant women aged 18 years and above who were attending antenatal Omdurman friend ship hospital, between April 2015- May 2017.

Venous blood samples were collected from study participants and screened for IgG and IgM antibodies against *T. gondii* using the ELISA technique. The participants were also interviewed about selected behaviors that predispose individuals to infections with *T. gondii*.

Results: Out of the 300 surveyed pregnant women, 65(21.7%) serum samples were found to have IgG while IgM was showed only 3(1%) IgM. Surveyed populations were divided into four age groups: (up to 20 years), (21-30), (31-40 years) and (more than 40 years). The positive cases within each age group showed IgM in one (0.3%), one (0.3%), one (0.3%) and 0(0%) respectively. On the other hand IgG was detected in 9(3%), 36(12%), 19(6.3%) and one (0.3%) respectively. Exposure to cats has been considered a major risk factor for acquisition of infection [1]. In the present study, it appears that cats have no direct role in transmission of the disease and there is no statistically association between cat contact and infection (P= 0.88 and 0.1 with IgM and IgG respectively). Also meat consumption has no role in disease transmission as no statistically association women with previous history of miscarriage in the study group showed *T. gondii* sero- prevalence rate of 42 and 3 using ELISA IgG and IgM respectively compared to 23 and 0 among others with no past history of miscarriage with no significant difference between them (P =0.76 and 0.18) respectively.

Conclusion: The seroprevalence of *T. gondii* antibodies is relatively low among the pregnant women. Undercooked meat consumption and cat contact are not significantly associated with sero-prevalence of anti-*T. gondii* IgG and IgM among the pregnant women.

Introduction

T. gondii is an obligate intracellular protozoan parasite that causes toxoplasmosis [2]. It is estimated that about one third of the world's population is infected with *T. gondii* [3]. In Africa, overall seroprevalence rate as high as 92.5% has been reported [4]. However, the prevalence of infection varies widely between countries (ranging between 10 and 80%) and often within a given country or between different communities in the same region [3]. However, in most developing countries including Sudan, the prevalence of toxoplasmosis is largely unknown. In the immunocompetent *Toxoplasma* infection is often asymptomatic, and frequently results in the chronic persistence of cysts within host tissues, that probably lie dormant for life [5,6]. High prevalence of the infection have been reported among pregnant women and women of childbearing age from different foci in Latin America, parts of Eastern/Central Europe, the Middle East, parts of south-east Asia and Africa [3]. This can lead to congenital toxoplasmosis, which is a serious disease that is associated with severe congenital deformities and intrauterine

deaths especially when the infection is acquired early in pregnancy [7]. Control of congenital toxoplasmosis is therefore important and, in the developed countries, includes screening of pregnant mothers for infection. Most studies on prevalence of *T. gondii* infections have relied on antibody detection. Although, antibody detection is associated with false positives. A more reliable way of diagnosing active infection is the use of molecular techniques such as PCR for diagnosing active infection using blood, brain biopsy, and liver biopsy [8-10]. However, this technique is very expensive and not easily available in most developing

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countries making its use in routine diagnosis not feasible. It is for this reason that diagnosis of toxoplasmosis in most health facilities in these countries still largely rely on serology, and this is the method of diagnosis that was used in this study for purposes of being realistic with what is done in the field. In line with this, the present study sought to determine the seroprevalence of *T. gondii* infections among pregnant women attending selected antenatal clinics in omdurman, using ELISA tests and risk factors associated with the acquisition of toxoplasmosis.

Material and methods

The Study area and population

The study was conducted in Khartoum state targeting the pregnant women attending Omdurman friendship hospital. The targeted women were categorized according to age groups (15- 20, 21-30, 31-40 and 40-45 year) and stage of pregnancy (1st, 2nd and 3rd trimesters).

Study design: This was a cross-sectional descriptive study involving pregnant women attending antenatal care at selected health centers. Each participant was interviewed in private to promote confidentiality. Furthermore, no names were used on any data collection forms. Instead, each study participant was given a unique identifying number. Venous blood samples were drawn from each study participant.

Sample size: The minimum sample size for the study was calculated using the following formula given by Fischer *et al.* [11]. Sample size calculation according to Fischer's formula.

Data collection and analysis: After obtaining informed consent, the study participants were interviewed by administration of a standard questionnaire to obtain the socio-demographic and economic status information as well as epidemiological risk factors. The influential risk factors considered in the study included maternal age, gestation period, contact with cats, and cooking and eating habits such as: eating raw or undercooked meat. Laboratory results from the tests were entered using the same number as the one on the questionnaires. All the data were then entered into Excel spreadsheet and later exported into SPSS for statistical analysis.

Collection of sample: About 5mL of venous blood were collected aseptically from each of the 300 pregnant women. All specimens were transported to the laboratory where serum was separated from the blood by centrifugation at 3,000 rpm for 5 minutes. Serum samples were numbered and tested for anti-*T. gondii* antibodies (IgM and IgG) using ELISA assays.

Methods

TOXO IgG- IgM: Ten microliters of plasma sample was diluted using 1000 µl of sample diluent. The diluted specimen was then incubated in *T. gondii* antigen-coated wells at 37°C for 1 hour. The wells were then washed with washing solution to remove residual test specimen. Enzyme-labeled antibodies to human IgG -IgM (conjugate) were then added. After another washing using washing solution to eliminate unbound material an enzyme substrate solution containing a chromogen was added. This solution developed a blue colour if the sample contained anti-*T. gondii* IgG and anti-*T. gondii* IgM. The blue colour then changed to yellow after blocking the reaction with sulphuric acid. The intensity of the colour which is proportional to the amount of anti-*T. gondii* IgG-IgM in the test specimen was read using an ELISA reader at 450 nm wavelength. The concentration of antibodies in the sample was then determined by mean of a calibration curve.

Result and discussion

Few studies have looked at sero-prevalence of anti-*T. gondii* in Sudan. The present study showed an overall seroprevalence of anti-*T. gondii* antibody among pregnant women in Omdurman friendship, to be 22.6%. (Table 1). The result however, agreed with Musa (2014) who showed that 20.2% of pregnant women were positive for IgG. Some showed a high prevalence rate than the results obtained for example in Turkey 77% was recorded, and in Togo 75% were reported by Deniau *et al.* also the results obtained by Satti, 2011 showed that the prevalence was 38.9% by ELISA IgG in Khartoum state; also Khalil (2014) was found 73.1% by using ELISA IgG in rural areas in Sudan. It may also disagree with result obtained in 2006 by Maha *et al.* who showed that the prevalence using ELISA was 35.1% positive IgG antibodies to *T. gondii* in Sudanese pregnant women. Regional variation of sero-prevalence of *T. gondii* infection has previously been described [1]. The reason for this may not be clear but could include several factors such as socio-economic status, education, sanitary conditions and differences in dietary habits. The fact that the study population had some individuals with IgM (1%) antibodies implies that transmission of *T. gondii* infection had either taken place during the pregnancy or the women had become pregnant shortly after acquiring *T. gondii* infection. Although the prevalence of acute infection was low at 1%, this is still an important finding since there is a risk of these women transmitting the infection to their unborn child thereby resulting in congenital toxoplasmosis, which is a more severe disease that may not only lead to abortion but also to congenital malformations in the new born, the results indicated that the higher rate was encountered in the age group of 21-30 years, followed by the next group 31-40 years. approximately, corresponds with the age group 26-30 years previously reported by Khalil *et al.* [12] and disagree with the results revealed by AL-Dahmoshi *et al.* [13] in Iraq that the age group ranged from 14-24 years was more infected with toxoplasmosis (Table 2). In the present study, it appears that cats have no direct role in transmission of the disease and there is no statistically association between cat contact and infection (P= 0.88 and 0.1 with IgM and IgG respectively). However, several studies have concluded that exposure to cats increased the risk of *T.gondii* seropositivity, other studies found no association [14-19]. Exposure to cats is not sufficient for transmission of *T. gondii* to women; infection in cats and personal hygiene should be evaluated. In Sudan the contact between people and cats is not as in Europe (Table 3). The role of consumption of raw meat in the acquisition of *T. gondii* infection has not always been clear. Several studies have found no association, although some studies have identified an association between eating raw meat and *T. gondii* seropositivity [1,16,20-26]. In this study, meat consumption has no role in disease transmission as no

Table 1. Prevalence according to IgG and IgM specific antibody for *T. gondii* using ELISA

Type of immunoglobulin by ELISA	Total	Positive cases	Percentage (%)
IgG	300	65	21.7%
IgM	300	3	1%

Table 2. prevalence of *T.gondii* immunoglobulin according to age groups

Immunoglobulin by ELISA	Age group(years)				Total
	15- 20	21-30	31-40	41-45	
IgM	1	1	1	0	3
IgG	9	36	19	1	65

Table 3. Antitoxoplasma antibodies in pregnant women who had contact with cats

Cat contact	Number examined	IgG positive	IgM positive
Yes	153	39(25.5%)	3(2%)
No	147	26(17.7%)	0(0%)

Table 4. Anti Toxoplasma antibodies (IgM, IgG) in pregnant women who consume raw meat using ELISA

Consuming raw meat	Number examined	IgG positive	IgM positive	p value
Consume	123	32 (26%)	1 (0.8%)	0.127
Not Consume	177	33 (18.6%)	2 (1.1%)	0.786

statistically association that may due to people recently stop that habit of eating raw or uncooked meat (Table 4).

Conclusion

As a general conclusion, it can be stated that the frequency of specific IgM and IgG antibodies using ELISA is considered high among pregnant women investigated for Toxoplasmosis. The absence of a statistically significant relationship between the prevalence of *Toxoplasma* infection among pregnant women and contact with cats and consumption of raw meat.

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