

Full Length Research Paper

Seroprevalence and risk factors associated with *Toxoplasma gondii* infection among pregnant women in Ile Ife, Southwestern Nigeria

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This study investigated the seroprevalence of anti-*Toxoplasma* IgG and IgM antibodies as well as associated risk factors among pregnant women attending ante-natal care clinics in Ile Ife, Southwestern, Nigeria. A cross sectional hospital study was carried out in three major health care facilities within the study area between June and October, 2019. Three hundred and twenty pregnant women aged between 14-50 years were enrolled during the study period and their blood samples were screened for IgG and IgM antibodies against *T. gondii* using ELISA test. Structured questionnaires were also used to obtain participants' socio-demographic data and some risk factors associated with the infection. Among the 320 pregnant women studied, overall seroprevalence of *T. gondii* was 34.68%, whereas 23.75% of the pregnant women were seropositive to IgG, 10.94% showed seropositivity to IgM antibodies. The analysis of the study also revealed that the seropositivity of *T. gondii* increased as the age of the pregnant women increased and decreases with increase in level of educational attainment. Significant associations were recorded among seropositivity of *T. gondii* infection and marital status, cat ownership, habit of tasting raw/undercooked meat and presence of rodents/cockroaches in the household ($P < 0.05$). The findings of this study show that the seropositivity to anti-toxoplasma antibodies is high in the study area. Therefore, there is a need to educate pregnant women about the risk factors that can lead to toxoplasma infection in order to reduce congenital toxoplasma cases among their offspring and newborns. It is also very important to include testing for toxoplasma antibodies as part of the ante-natal investigation that will be carried out on pregnant women.

Key words: *Toxoplasma gondii*, pregnant women, seroprevalence, infection, risk factors, marital status, cat ownership.

INTRODUCTION

Toxoplasmosis is an infection caused by an obligate intracellular protozoan parasite that exhibit a heterogenous life cycle in human and other vertebrate animals (Salimaomran and Hamed, 2015). The infection

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has been widely distributed across the globe with over one billion people estimated to have been exposed to the parasite (Dubey, 2010). Infections due to *T. gondii* are considered a worldwide zoonosis of great public health importance and are influenced by cultural, hygienic and nutritional habits, climate and environmental conditions (Teneter et al., 2000). According to Dubey (2010) cats (wild and domestic) are the only animals that excrete resistant oocyst of *T. gondii* into the environment while other animals including man, serve as intermediate hosts. Vertebrates including man are infected with *T. gondii* through ingestion of undercooked meat from cattle, sheep, goats, pigs and wild life that contain tissue cysts (Jones et al., 2006) also, through ingestion of sporulated oocysts from food, vegetable, fruits and water contaminated with cats faeces (Joyner, 1982). Teneter et al. (2000) reported that infection by *T. gondii* can also occur through organ transplantation or blood transfusion. This is in addition to transplacental transmission from mother to foetus (Rorman et al., 2006). Olusi et al. (1994) postulated that consumption of infected rats may also increase the risk of direct transmission from rats or non-felines to human.

Most infections of toxoplasmosis in immunocompetent humans are asymptomatic; however, during acute infection, lymphadenopathy, fever, sore throat and flu-like illness that mimic infectious mononucleosis have been reported (Teneter et al., 2000). During pregnancy, Kafale et al. (2015) reported that the chance of sero-conversion among pregnant women is high, and would have tragic outcomes for the foetus, newborn and mother, all of which can be prevented.

Toxoplasmosis in pregnant women has been reported to be severe because the etiologic agent can cross placental barrier to infect foetal tissues consequently resulting in spontaneous abortion, miscarriages or foetal deformities (Kafale et al., 2015). The risk of toxoplasmosis transmission in women during pregnancy was reported by Dunn et al. (1999) to increase from first trimester (15-24%) to third trimester (60-90%) of pregnancy while that of congenital defect is more severe in early infections (Patton, 1993). Ante-natal serological screening of pregnant women for the parasite based on IgG and IgM is the mainstay in monitoring the risk of congenital toxoplasmosis (Kafale et al., 2015) and after the detection, maternal-foetal intervention for toxoplasmosis can be achieved through the use of drugs such as spiramycine which prevents congenital infection by more than 60% (Goldstein et al., 2008). Although, series of studies have been carried out in Nigeria to date, *T. gondii* infection has not been documented in many parts of Nigeria including the study area. Therefore, the aim of the present study was to estimate the seroprevalence and assess associated risk factors of *T. gondii* infection among pregnant women in Ile Ife, Southwestern, Nigeria.

MATERIALS AND METHODS

Study area

This study was carried out in Ile-Ife, Nigeria. Ile-Ife is an ancient town in Southwest Nigeria located on Latitude 7° 28' 06"N, Longitude 4° 34' 22"E and it is 244 m above sea level (Sosan et al., 2019). Ile Ife is the head quarter of Ife Central Local Government and is about 218 km to Lagos with population of 509,813 (NPC, 2006). The climate of the area is typically tropical with a characteristic dry season of about 6 months (October-March) and a wet season of about 6 months (April-September) (Akinbuwa and Adeniyi, 1996). The mean annual temperature is about 30°C (Ndifon and Ukoli, 1989), while the mean annual rainfall ranges between 1000 and 1250 mm (Oguntoyinbo, 1982). The vegetation of the area is tropical rain forest, characterized by large and tall trees. The inhabitants are majorly the Yoruba speaking people of the south-west. However, people from different ethnic groups in Nigeria are also present.

Sample size and sampling technique

Pregnant women who visited teaching hospital, private hospitals and health centers in Ile-Ife for their ante-natal clinic comprised the study population. Sampling method was carried out according to Teweldemedhin et al. (2019). The names of hospitals that care for pregnant women in Ile-Ife were written on a piece of paper and enclosed in a box. Three hospitals were selected randomly by lottery method. The total sample size was then proportionally allocated to the health facilities based on their previous records of attendant of pregnant women. Pregnant women in the age range 14-50 years, who were attending ante natal clinic in the three hospitals and also met the inclusion criteria, were recruited over the study period, using systematic random sampling method.

Study design

A cross-sectional study was carried out among 320 pregnant women that attended ante-natal clinic in three reputable hospitals, namely Obafemi Awolowo University Teaching Hospital Complex (OAUTHC), Seventh Day Adventist Hospital (SDAH) and Enuwa Primary Health Care (EPHC) in Ile Ife, Osun State, Nigeria in order to estimate the seroprevalence of anti-*Toxoplasma* IgG and IgM antibodies. During ante-natal clinic, participants were informed and enlightened about the study objectives, risks, benefits and study procedures in English Language and also interpreted in Yoruba Language, where necessary. Duly signed informed consents were obtained from all pregnant women that were interested to be part of the study. The participants were assured of confidentiality and anyone that do not want to be part of the study was allowed to opt out. One hundred and forty (43.75%), seventy one (22.19%) and one hundred and nine (34.06%) pregnant women were recruited for this study from OAUTHC, SDAH and EPHC respectively.

Questionnaire administration

A well-structured, open-ended and pre-tested questionnaire was administered to obtain the socio-demographic details of participants including age, occupation, tribe, religion and associated risk factors which include cat ownership, history of abortion, tasting of raw or undercooked meat, engaging in backyard gardening, presence of rodent and cockroaches in households.

Consent and ethical issues

Ethical clearance for the study was obtained from Ethical Committee of OAUTHC, SDAH in Ile Ife and Osun State Ministry of Health, Osogbo before the commencement of sample collection.

Collection of blood samples

Five milliliters (5 ml) of blood was collected aseptically from each participating woman by venipuncture into plain test tube using 5 ml capacity syringe and needle between the months of June and October 2019. The tubes were pre-labeled with subjects' identification numbers. Sera from the blood samples were separated by allowing the blood to clot at room temperature and centrifuged at 2000 rpm for 10 min. The clear sera were separated into vials using Pasteur pipette and stored at -20°C until serological test was carried out.

Laboratory analysis

Sera samples were tested in duplicates for both anti-*Toxoplasma* IgG and IgM antibodies using commercially procured kits (Microwell Toxo [IgG and IgM] Bio-Inteco Diagnostic Ltd, UK designed for the purpose. Thaw serum samples and reagents were brought to room temperature and tests were performed according to the manufacturer's instructions. The mean value for each sample was calculated by dividing the mean absorbance values of duplicate wells of each sample by the cut off calibrator mean value. Results were interpreted as follows

Toxo IgG Index	Interpretation
≥1.00	Positive
0.91-0.99	Equivocal (sample should be retested)
≤ 0.90	Negative

Toxo IgM Index	Interpretation
<0.90	Negative for IgM to <i>Toxoplasma gondii</i>
0.91-0.99	Equivocal, sample should be retested
1.00-2.00	Low Positive
2.01-2.50	Moderate Positive
>2.50	High Positive

Statistical analysis

All statistical analyses were carried out using SPSS for windows version 21.0. Significance of differences in seroprevalences of *Toxoplasma* infection among subgroups was determined by chi-square and statistical difference was assigned at $p \leq 0.05$.

RESULTS

A total of 320 pregnant women aged between 14-50 years were screened for anti-*Toxoplasma* IgG and IgM antibodies. The overall seroprevalence of infection was 34.69% with IgG and IgM antibodies constituting 23.75 and 10.94% respectively. Table 1 showed the distribution

of seropositivity to *T. gondii* antibodies according to socio-demographic information of the pregnant women. From the result in Table 1, women aged 20 years and below had the least anti-*Toxoplasma* IgG (14.29%) and IgM (9.52%) antibodies while the highest IgG (25.66%) and IgM (10.41%) prevalence were recorded in age groups 31-40 years and 21-30 years respectively. Generally, it was observed that seropositivity to *Toxoplasma* infection increased along with increases in ages of the pregnant women. It was also observed that none of the pregnant women aged between 41 and 50 years was seropositive to anti-*Toxoplasma* antibodies. No significant relationship was recorded between age distribution of anti-*Toxoplasma* IgG and IgM seropositivity among the pregnant women ($P > 0.05$). Statistical analysis revealed that there was no significant difference ($P > 0.05$) in the seropositivity among the different educational classification of the pregnant women studied. However, a decrease in seropositivity of *T. gondii* was observed with increase in educational level (Table 1).

Based on the different occupational classification of the pregnant women, a non-significantly higher seroprevalence of anti *T. gondii* IgM (15.22%) and IgG (26.09%) antibodies was recorded among pregnant women that were artisan and trader respectively ($P > 0.05$) (Table 1). Considering the effect of marital status among the pregnant women, higher seroprevalence (41.79%) of Toxoplasmosis was recorded among pregnant women that were single in the category. A significant relationship was observed between marital status and seropositivity of anti *T. gondii* antibodies among the pregnant women ($P < 0.05$) (Table 1). With regards to proximity to cats, none of the pregnant women who own cats were seropositive to anti-*Toxoplasma* antibodies. However, a significant association ($P > 0.05$) was observed among those that do not own cats and seroprevalence of anti-*T. gondii* antibodies among the pregnant women (Table 2).

Considering the influence of backyard gardening on the seropositivity of *Toxoplasma* infection among pregnant women, the results showed a non-significantly higher prevalence of infection among pregnant women that engaged in backyard gardening (37.59%) ($P > 0.05$) in comparison with those without backyard soil contact (Table 2). Pregnant women in the habit of tasting raw or undercooked meat while cooking had higher seroprevalence of both IgG (27.05%) and IgM (14.98%) antibodies than their counterparts that eat meat only when it is properly cooked. There was a significant association between raw meat tasting and seroprevalence of anti-*Toxoplasma* antibodies among the pregnant women ($P < 0.05$) (Table 2). High seroprevalence of anti-*T. gondii* antibodies was recorded in all women of gestational ages in this study, but the highest (13.95%) IgM antibodies were recorded among those in their 1st trimester of pregnancy. There was no significant relationship between seropositivity and gestational age of

Table 1. Distribution of anti-*Toxoplasma* IgG and IgM in relation to social demographic information among pregnant women in Ile Ife Osun State.

Social Demographic Information	Number Examined	No (%) Positive		Total	χ^2	P-value
		IgG	IgM			
Age (years)						
≤ 20	21	3(14.29)	2(9.52)	5(23.81)	0.343	0.952
21-30	184	44(23.91)	21(11.41)	65(35.33)		
31-40	113	29(25.66)	12(10.62)	41(36.28)		
41-50	2	0(0.00)	0(0.00)	0(0.00)		
Total	320	76(23.75)	35(10.94)	111(34.69)		
Educational status						
Primary	162	39(24.07)	23(14.20)	62(38.27)	3.073	0.282
Secondary	137	29(21.17)	11(8.03)	40(29.19)		
Tertiary	2	1(50.00)	0(0.00)	1(50.00)		
No education	19	7(36.84)	1(5.26)	8(42.11)		
Total	320	76(23.75)	35(10.94)	111(34.69)		
Occupational category						
Civil servant	62	13(20.97)	7(11.29)	20(32.26)	3.307	0.045
Trader	135	37(27.41)	17(12.59)	54(40.00)		
Artisan	46	12(26.09)	7(15.22)	19(41.30)		
Farmer	3	0(0.00)	0(0.00)	0(0.00)		
Student	41	8(19.51)	2(4.88)	10(24.39)		
Applicant	33	6(18.18)	2(6.06)	8(24.24)		
Total	320	76(23.75)	35(10.94)	111(34.69)		
Marital status						
Single	67	19(28.36)	9(13.43)	28(41.79)	0.343	0.009
Married	188	51(27.13)	24(12.77)	75(39.89)		
Divorced	50	4(8.00)	2(4.00)	6(12.00)		
Widowed	15	2(13.33)	0(0.00)	2(13.33)		
Total	320	76(23.75)	35(10.94)	111(34.69)		

 χ^2 =Chi square.

Source: Author

Table 2. Distribution of anti-*Toxoplasma* IgG and IgM in relation to behavioral parameters of pregnant women in Ile Ife Osun State.

Cat Ownership	Number Examined	No (%) Positive		Total	χ^2	P-value
		IgG	IgM			
Cat owners	9	0(0.00)	0(0.00)	0(0.00)	2.884	0.026
Non cat owners	311	76(24.44)	35(11.25)	111(35.69)		
Total	320	76(23.75)	35(10.94)	111(34.69)		
Backyard gardening						
Yes	141	36(25.53)	17(12.06)	53(37.59)	0.442	0.506
No	179	40(22.35)	18(10.06)	58(32.40)		
Tasting of raw/undercooked meat						
Taster	207	56(27.05)	31(14.97)	87(42.03)	3.532	0.001
Non-taster	113	20(17.69)	4(3.54)	24(21.24)		
Total	320	76(23.75)	35(10.94)	111(34.69)		

 χ^2 =Chi square.

Source: Author

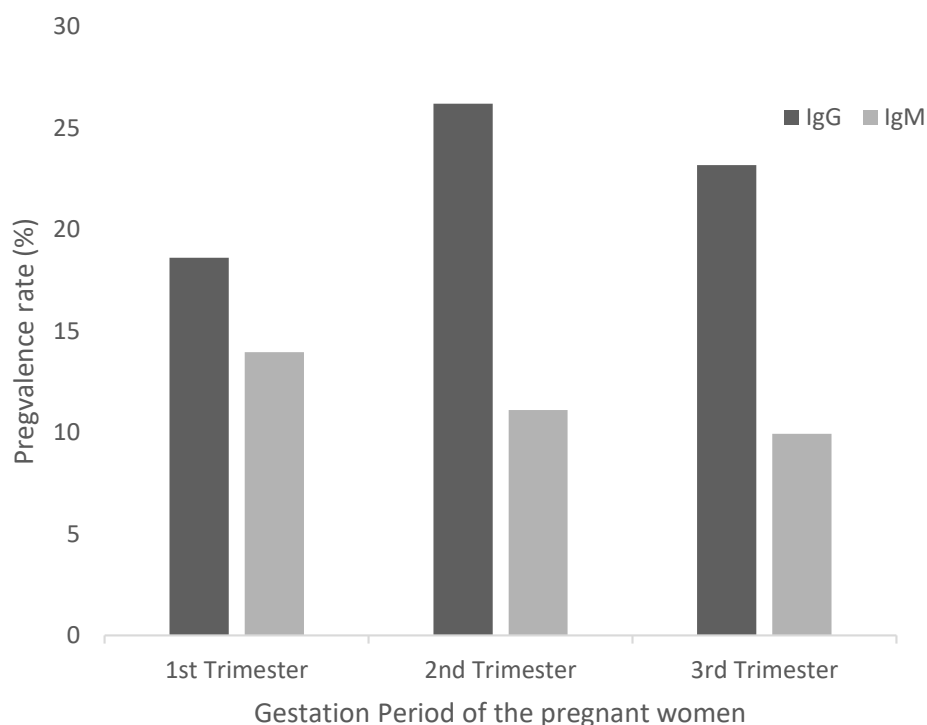


Figure 1. Seroprevalence of *T. gondii* according to gestation age of the pregnant women from Ile Ife.

Source: Author

pregnancies of the pregnant women ($P > 0.05$) Figure 1. Majority of the pregnant women screened in this study admitted to harbouring rodents and cockroaches in their households, consequently higher seroprevalences of 25.08% and 11.37% in IgG and IgM antibodies were recorded among these categories of women respectively. There was a significant relationship between seropositivity to *T. gondii* infection and presence of rodent/cockroaches in households ($P < 0.05$) Figure 2.

DISCUSSION

The determination of *T. gondii* serostatus of pregnant women in all stages of pregnancy is very important and is key to developing prevention and control programme for congenital toxoplasmosis. In a situation where a woman seroconverted for the first time during pregnancy, there is high possibility of passing the pathogen to the fetuses, a situation that can ultimately lead to serious clinical complication and even foetal damage (Gelaye et al., 2015). This study is very important in the study area because to the best of our knowledge there is no information about seroprevalence of anti-*Toxoplasma* antibodies and associated factors among pregnant women in Ile Ife, Osun State Nigeria. The overall

seroprevalence rate of 34.68% recorded among the pregnant women studied is not significantly different from 34.10, 38.80 and 37.80% reported by Elnahas et al. (2003), Almushait et al. (2014) and Hajsolaimani et al. (2012) in Sudan, Saudi Arabia and Iran respectively. However, the present result is lower than 83.60 and 85.40% reported among pregnant women in Jimma town, Ethiopia (Zemene et al., 2012) and Addis Ababa (Gelaye et al., 2015) respectively. The result was higher than 18.4% reported in South Africa (Bessong and Mathomu, 2010), 19.1% in Italy (Thaller et al., 2011), 29.9% reported in Zaria (Deji-Agboola et al., 2011) and 11.7% reported in Jos (Akubuilu et al., 2020). The variation might be attributable to differences in geographical location, behavioral and feeding culture and serological methodology employed (Almushait et al., 2014).

In this study, the trend of seropositivity of anti-*Toxoplasma* antibodies among the pregnant women appears to be age-dependent. This is because seropositivity of anti-*Toxoplasma* antibodies increased as the age group among the pregnant women increases; which could be owing to extended exposure to the risk factors associated with *T. gondii* infection (Peyron et al., 2016; Olariu et al., 2020). Similar findings have been documented in the reports of other authors (Varella et al., 2003; Deji-Agboola et al., 2011; Mwanbe et al., 2013;

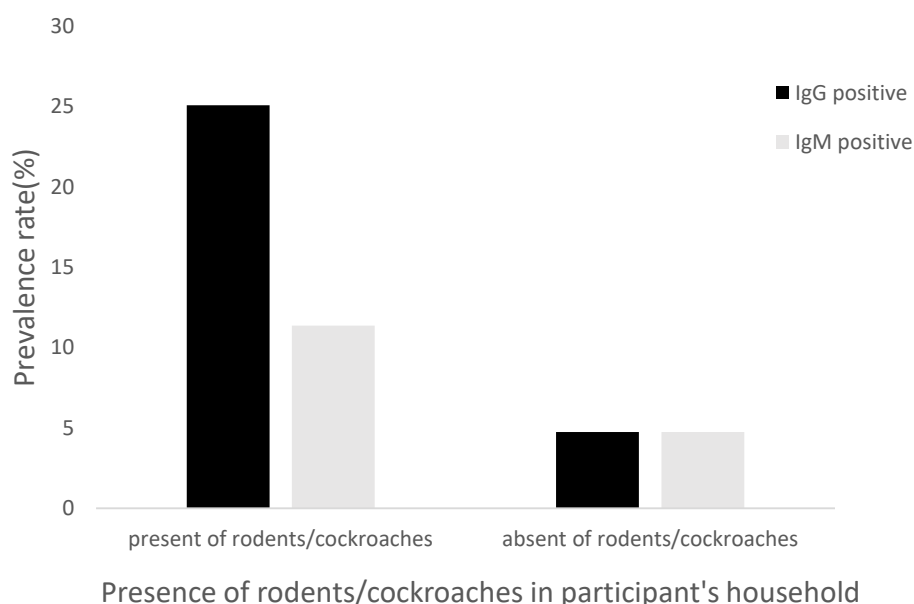


Figure 2. Seropositivity of *T. gondii* among participants with respect to presence of rodents/cockroaches in the household.

Source: Author

Singh et al., 2014; Akubuilu et al., 2020). Also, a non-significant association between age and seropositivity to *T. gondii* was observed among the pregnant women studied. This result agrees with those of Gelaye et al. (2015), Endrias et al. (2013) and Al-Harathi et al. (2006), but contradicts the report by Zemene et al. (2012) in Jimma Town in Ethiopia.

Considering seropositivity to *Toxoplasma* infection with level of education among pregnant women, a non-significant relationship was observed with those without formal education having the highest prevalence. Women without any formal or low level of education had higher rate of toxoplasmosis in this study. Similar observations were reported in previous studies by Hajisoleimani et al. (2012) in Iran, Nasir et al. (2015) in Maiduguri, Fallah et al. (2003) in Iran, Ertug et al. (2005) in Turkey, Neguessie et al. (2017) in East Ethiopia and Akubuilu et al. (2020) in Jos, Nigeria. Based on marital categorization of pregnant women, single pregnant women recorded higher seroprevalence of *T. gondii* in this study. This is in agreement with the previous report of Singh et al. (2014) in India and the reason is unknown.

Surprisingly, pregnant women that own cats were seronegative in this study, while high seropositivity was recorded among those that do not own cats. This agrees with Walker et al. (2008) who found no direct association between cat ownership and risk of toxoplasmosis contraction in people. The result however contradicts those of Lin et al. (2008) and Zemene et al. (2012) who reported high seroprevalence of *T. gondii* infection

among people that kept cats as pet as opposed to those that did not.

The present study recorded a non-significant association between pregnant women that engaged in gardening behind their residences and seropositivity to *T. gondii* antibodies. This agrees with the earlier reports of Nissapatron et al. (2011) in Thailand, Agmas et al. (2015) in Ethiopia but contradict the studies by Jumaian (2005) in Jordan and Mandour et al. (2017) in Egypt. Dubey (2000) stated that the association of cat and human toxoplasmosis is difficult to assess, but constant direct association with soil will increase the risk of transmission because the oocysts are found buried along with faeces of cats in the soil. This might be the reason why high prevalence of infection was recorded among pregnant women that engaged in backyard gardening because stray cats might have buried their faeces in the soil within their garden.

Tasting of raw or undercooked meat has been recognized as one of the important means of transmission; however, a significantly higher relationship in *Toxoplasma* seropositivity was observed among the pregnant women with respect to tasting of raw or undercooked meat. According to Dubey (2000), eating or tasting of raw/undercooked meat, contact with soil and travelling outside one's country were the major factors that exposes one to high risk of infection. Majority of the seropositive pregnant women in this study alluded to the presence of rodent and cockroaches in their households and this is in agreement with the study of Moura et al.

(2013) in Brazil. Rodents and cockroaches have been epidemiologically implicated in mechanical transmission of *T. gondii* oocyst since they are potential facilitators of the contamination of food and water.

A non-significant association was observed between the gestation age and the seropositivity of *T. gondii* antibodies in this study. High seroprevalence of acute toxoplasmosis (IgM antibodies) was recorded among pregnant women in their first trimester. The results were in accordance with the report of Alayande et al. (2013) in Sokoto, Northwestern Nigeria. Foetal implication with acute toxoplasmosis during the first trimester is very hazardous to the developing foetus because this trimester is the formative stage and according to Patton (1993), the risk of congenital defect in the foetus is more severe with such early infection.

Conclusion

This study provided the current status of anti-toxoplasma antibodies among pregnant women in Ile-Ife, Osun State, Southwestern Nigeria. Our findings showed that the possible risk factors to *T. gondii* infection in the study area are marital status, tasting of raw or undercooked meat and presence of rodent or cockroaches in the household. Based on the findings of this study, it is therefore recommended that testing for toxoplasma antibodies should be included as part of the ante-natal investigation that will be carried out on pregnant women. Also, there is a need to educate pregnant women on the need to improve on personal hygiene and risk factors that can lead to toxoplasma infection in order to reduce congenital toxoplasma cases among their offspring and newborns.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interest.

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