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### **Original article**

### Seroprevalence of anti-Toxoplasma IgG and IgM among pregnant women attending antenatal clinic in Jos-North, Plateau State, Nigeria

Eberechukwu Esther Nwachukwu, Ocheme Julius Okojokwu<sup>\*</sup>, Murna Ahmed Ali, Yusuf Amuda Agabi

Department of Microbiology, Faculty of Natural Sciences, University of Jos, Jos, Plateau State, Nigeria .

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#### ABSTRACT

Background: Toxoplasmosis, one of the TORCH infections in pregnancy is caused by Toxoplasma gondii (T. gondii) an obligate intracellular protozoan parasite which can cause severe complications for an infected mother if the primary infection was acquired during pregnancy. Aim: This study aimed to determine the seroprevalence of anti-Toxoplasma IgG and IgM antibodies and the associated risk factors in pregnant women attending antenatal clinics in Jos-North, Plateau State, Nigeria. Methods: Descriptive cross-sectional study was undertaken involving a total of 158 consenting pregnant women visiting health facilities in Jos from March to June 2020. Data regarding socio-demographics and associated factors were gathered using questionnaire. Enzyme-linked immunosorbent assay (ELISA) was deployed to test for anti-T. gondii IgG and IgM antibodies. Results: The seroprevalence of anti-Toxoplasma IgG was 26.6% while that of IgM was 5.7%. Seven (4.4%) women were seropositive for both IgG and IgM anti-Toxoplasma antibodies. Amongst all the socio-demographics assessed, only occupation was significantly associated with prevalence of anti-Toxoplasma antibodies (p =0.035). Amongst the risk factors, regular contact with soil and source of drinking water were significantly associated with T. gondii infection (p = 0.028 and p =0.002 respectively). Conclusion: Serological evidence showed that pregnant women in the current study had moderate exposure to Toxoplasma gondii while 72.2% of the study subjects have not any previous exposure are at risk of toxoplasmosis. Hence, regular screening for T. gondii infection is recommended during pregnancy and antenatal care.

#### Introduction

*Toxoplasma gondii (T. gondii)* is an obligate apicomplexan intracellular parasitic protozoan that causes toxoplasmosis, a zoonosis [1,2]. It is reported that a third of the world's population, especially in developing and low-income countries harbours or have had the parasite [3]. The rate of infection varies greatly between and

even within countries and difference could be as high as between 20% to 60% [4-6]. Infection rates are highest in regions with hot, humid weather and lower altitudes [7]. Almost all warm-blooded species, including birds, are infected by the parasite [8]. The feline (cat) family is the only known definitive host and consequently the infection's

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<sup>\*</sup> Corresponding author: Ocheme Julius Okojokwu

E-mail address: okojokwuo@unijos.edu.ng

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primary reservoir [9]. To all hosts, *T. gondii* has three infectious stages: bradyzoites, tachyzoites, and sporozoites in oocysts. Food-borne (undercooked or raw meat containing tissue cysts, improperly washed vegetables, and fruits), drinking water contaminated with oocysts shed by cats [10], and transplacentally, where an infected pregnant woman transmits tachyzoites crossing the placental barrier to the foetus, are the most common ways for humans to become infected [11]. Other transmission routes, however, are possible.

Even though *T. gondii* infection in pregnant women is linked to spontaneous abortion, abnormal foetal organ development, foetal death shortly before and after birth, the infection is neglected in Nigeria, where screening is not routinely performed during antenatal care and follow-up is not available [12]. Therefore, it is the aim of this study to determine the seroprevalence of anti-*T. gondii* antibodies and associated risk factors among pregnant women receiving antenatal care in Jos-North, Plateau State, Nigeria.

#### **Materials and Methods**

A descriptive cross-sectional study was conducted between March and June 2020 in Jos– North, Plateau State, Nigeria. Permission was obtained from the Ministry of Health Monitoring and Evaluation Committee of the Primary and Health Care Development, Jos–North, Plateau State. The ethical committee of Faith Alive Foundation (FAF) and Bingham University Teaching Hospital (BUTH) also approved conduct of this study (reference number NHREC/21/05/2005/0067).

The study locations were Bingham University Teaching Hospital (BUTH), Faith Alive Foundation (FAF) and some selected highly patronised Primary Health Centres (PHCs) all situated in Jos North. The selected PHCs are located at Kabong, Dogon Agogo and Township. Jos is in the North Central geopolitical zone of Nigeria lying 9.1667°N, 9.7500°E with an area of 26,899 square kilometres with an estimated population of 3, 206,531 and of these 1, 607,533 are females based on the 2006 census.

The study population comprised of all consenting pregnant women who attended their ante-natal care (ANC) as well as those pregnant women who registered for their first visits and at these health facilities. Pregnant staff working in these health facilities were also included in this study. Sick pregnant women on admission in the health facilities and those who declined to participate were excluded.

#### Sample collection and laboratory analysis

Approximately 2mL of venous blood was collected aseptically from the participants using plain vacutainer tubes and needles. All samples were transported with ice box from the different health facilities to Faith Alive Foundation. Serum was obtained by centrifugation at 14000 rpm for 10 minutes and was stored at -80°C prior to analysis. Serum samples were tested for anti-T. gondii IgG and IgM antibodies respectively using ELISA test kits (Nantong Voyage Medical Co., Ltd, Jiangsu, China), according to the manufacturer's instruction. The cut-off point was calculated following the procedures according to the manufacturer's recommendations. A negative reaction was judged as indicative of the absence of significant Toxoplasma antibodies. A positive Toxoplasma IgG/IgM reaction was interpreted as an indication of either a past/recent infection respectively. A total of 158 samples were collected and analysed.

A structured questionnaire was used to abstract information on socio-demographic characteristics, and risk factors associated with infection by *T. gondii*. Confidentiality was maintained by interviewing each study participant privately with their identification number on the questionnaires. Information was provided in English or interpreted in the local dialect by a volunteer independent of the study team.

#### Data analysis

Data was recorded and coded in excel spread sheet before analysis using Statistical Package for Social Sciences (SPSS) version 24. Descriptive statistics was used to summarize the data. Relationship between categorical variables were determined using Chi-squared test. p-value  $\leq$  0.05 was considered significant.

#### Results

# Seroprevalence of *T. gondü* specific IgG and IgM antibodies

A total of 158 pregnant women between ages 15 to 50 years (mean age was 26.41 years) were tested for anti-*T. gondii* IgG and IgM antibodies. Out of the total samples, 42 (26.6%) were positive for anti-*Toxoplasma* IgG, while 9 (5.7%) were IgM positive and 7 (4.4%) were both IgG and IgM positive (**Table 1**).

## Prevalence of anti-*Toxoplasma gondii* antibodies based on demographic factors

Majority (44.8%) of the seropositive pregnant women were from Dogon Agogo, Jos-North centre. In this study, 33.9% of the seropositive pregnant women belonged to 15 - 30 years age-group. Single pregnant women recorded the highest positive case (75.0%). Most (40.0%) of the women were educated with primary school holders recording the highest seropositive case. Test of association between location of the health facilities investigated, age (years), marital status, education and seropositivity for T. gondii infection, showed no statistical significance (p > 0.05). In terms of occupation, 5 (38.5%) of the seropositive cases were found majorly among students, followed by traders and the least among civil servants. However, there was statistical significance between occupations of participants and seropositivity for T. gondii infection (p = 0.035) (Table 2). This study demonstrates that seropositive cases were found majorly among students.

# Prevalence of anti-*Toxoplasma gondii* antibodies with respect to risk factors

The prevalence of *T. gondii* infection among pregnant women in their first and second trimesters were 31.0% and 30.1% respectively. Among the respondents with positive results, 42.9% reported that they have had miscarriage twice. Ten (33.3%)

seropositive participants were rural residents. Most seropositive women [23 (41.0%)] responded a no to tasting of meat while cooking. Forty-three (38.1%) women reported that they preferred consuming their meat cooked till soft. In the current study, 44 (34.1%) seropositive pregnant women had the habit of eating raw or undercooked vegetables. Seropositive pregnant women who did not own cats accounted for 32.6%. Participants who did not own a dog/pig or other type of animals recorded the highest seropositive cases were 45 (36.3%). This study demonstrates that the difference between trimester (gestational period), miscarriage, residence, HIV status, whether they taste their meat when cooking, form in which they often eat meat, whether they eat raw or undercooked vegetable, owning a cat, and owning a dog/pig or other animals and seropositivity for T. gondii infection was not statistically significance (p > 0.05).

Seropositivity with respect to whether participants had regular contact with soil was 15 (40.5%). There was a statistically significant difference between regular contact with soil and seropositivity for *T.* gondii infection (p = 0.028). In relation to source of drinking water, seropositive cases were found highest among pipe-borne water consumers [16 (40.0%)]. There was a very statistical significance between source of drinking water and seropositivity for *T.* gondii infection (p = 0.002) (**Table 3**).

Prevalence (%)
35 (22.2)
7 (4.4)
42 (26.6)
114 (72.2)
2 (1.3)
9 (5.7)

Table 1. Prevalence of Anti-Toxoplasma gondii antibodies.

Parameter		IgG			IgM		
	No. of sample	No. positive (%)	χ <sup>2</sup>	<i>p</i> -value	No. positive (%)	χ <sup>2</sup>	<i>p</i> -value
Location							
KJN	29	4 (13.8)	6.333	0.176	3 (10.3)	4.012	0.404
DAJN	29	12 (41.4)			1 (3.4)		
TJN	32	8 (25.0)			0 (0.0)		
FAF	48	14 (29.2)			4 (8.3)		
BHUTH	20	4 (20.0)			1 (5.0)		
Total	158	42 (26.6)			9 (5.7)		
Age (years)							
15 - 30	112	30 (26.8)	0.737	0.692	8 (7.1)	1.517	0.468
31-40	44	12 (27.3)			1 (2.3)		
41 - 50	2	0 (0.0)			0 (0.0)		
Marital status	1						
Single	4	2 (50.0) 1.864	1.864	0.396	1 (25.0)	2.949	0.229
Married	152	40 (26.3)	_		8 (5.3)		
Widow	2	0 (0.0)			0 (0.0)		
Education							
Primary	30	10 (33.3)	4.062	0.255	2 (6.7)	3.929	0.269
Secondary	83	24 (28.9)	_		7 (8.4)		
Tertiary	38	8 (21.1)	_		0 (0.0)		
None	7	0 (0.0)			0 (0.0)		
Occupation							
Student	13	5 (38.5)	1.983	0.576	0 (0.0)	8.590	0.035*
Civil servant	25	6 (24.0)			0 (0.0)		
Trader	67	15 (22.4)			8 (11.9)		
Housewife	53	16 (30.2)			1 (1.9)		

Table 2. Prevalence based on demographic factors.

 $\chi^2$  = Chi square \* = Significant association exists

KJN = Primary Health Care Kabong Jos-North; DAJN = Primary Health Care Dogon-Agogo Jos-North; TJN = Primary Health Care Township Jos-North; FAF = Faith Alive Foundation; BHUTH = Bingham University Teaching Hospital.

			IgG			IgM	
Parameter	No. of sample	No. positive (%)	$\chi^2$	<i>p</i> -value	No. positive (%)	$\chi^2$	<i>p</i> -value
Trimester							
First	29	9 (31.0)	1.339	.339 0.512	0 (0.0)	3.157	0.206
Second	53	16 (30.2)			5 (9.4)		
Third	76	17 (22.4)			4 (5.3)		
Total	158	42 (26.6)			9 (5.7)		
Miscarriage							
Once	39	7 (17.9)	1.983	1.983 0.576	2 (5.1)	1.408	0.704
Twice	7	2 (28.6)			1 (14.3)		
> Twice	7	2 (28.6)			0 (0.0)		
Never	105	31 (29.5)			6 (5.7)		
Residence							1
Urban	128	32 (25.0)	0.865	0.352	9 (7.0)	2.237	0.135
Rural	30	10 (33.3)	1		0 (0.0)	1	1
HIV status							
Positive	5	0 (0.0)	2.652	0.266	0 (0.0)	0.442	0.802
Negative	151	42 (27.8)			9 (6.0)		
No idea	2	0 (0.0)			0 (0.0)		
Taste meat duri	ng cooking	?					
Yes	102	23 (22.5)	2.399	0.121	5 (4.9)	0.338	0.561
No	56	19 (33.9)			4 (7.1)		
Form in which r Mildly cooked	33	4 (12.1)	4.503	0.105	0 (0.0)	3.634	0.162
Cooked till soft	113	34 (30.1)			9 (8.0)		
Cooked but tough	10	2 (20.0)			0 (0.0)		
Do you eat raw	or underco	oked vegetal	bles?				
Yes	129	35 (27.1)	0.109	0.742	9 (7.0)	2.145	0.143
No	29	7 (24.1)			0 (0.0)		
Do you own a ca							
Yes	23	7 (30.4)	0.205	0.651	0 (0.0)	1.625	0.202
No	135	35 (25.9)			9 (6.7)		
Do you own a do	0.0		-				
Yes	34	5 (14.7)	3.131	0.077	1 (2.9)	0.612	0.434
No	124	37 (29.8)			8 (6.5)		
Do you have reg			_				
Yes	37	15 (40.5)	4.823	0.028*	0 (0.0)	2.918	0.088
No	121	27 (22.3)			9 (7.4)		
What is your so			1				
Well	19	5 (26.3)	0.693	0.875	1 (5.3)	14.592	0.002**
Pipe-borne water	40	9 (22.5)			7 (17.5)		
Bore-hole Packaged water	46 53	14 (30.4) 14 (26.4)			0 (0.0) 1 (1.9)		

 Table 3. Prevalence based on risk factors.

 $\chi^2$  = Chi square; \* = Significant association exists;

\*\* = Very Significant association exists

#### Discussion

Toxoplasmosis has been recorded in pregnant women all over the world. The goal of this study was to assess the seroprevalence of *T. gondii* infection and related risk factors among pregnant women in Jos, Jos-North metropolis. *T. gondii* infection was shown to be prevalent in 32.3% of the 158 pregnant women in the study. Out of the total seropositive cases, 42 (26.7%) had *Toxoplasma* IgG antibodies, 9 (5.7%) had *Toxoplasma* IgM antibodies, and 7 (4.4%) had both *T. gondii*-specific IgG and IgM antibodies.

The total seroprevalence (32.3%) in this investigation indicates a decrease in T. gondii infection. Increased literacy levels, along with more hygienic hygiene practices, may help to explain why the reduction has occurred. The percentages reported in Lagos, Nigeria, Tanzania, Sokoto, Nigeria, and Sri Lanka were 32.8%, 30.8%, 27.7%, and 29.9% respectively [13-16]. This figure (32.3%), on the other hand, is lower than the previously reported seroprevalence of 79.3% and 81.8% positivity from an Ethiopian study of disparity pregnant women [17]. This in seropositivity rates could be related to the diagnostic procedures used, which have varying sensitivities and specificities, as well as local relative risk factors linked with geographic locations. The results of this study, when comparing IgG and IgM positive, are consistent with those of other investigations conducted in Northern Tanzania, Sri Lanka, and Lagos (Nigeria). When compared to IgG positivity, the investigations found few or no cases of IgM positivity [13, 16-18].

Serological identification of T. gondiispecific IgM indicates recent or current/acute infection, whereas the presence of T. gondii-specific IgG indicates past or latent infection [19]. Furthermore, the presence of both IgG and IgM antibodies in circulation implies that the infection was recently acquired because IgM antibodies rapidly decline following recently acquired illnesses [20]. IgM antibodies normally develop first in patients with acute T. gondii infection, according to research, and antibody levels become negative in a few months. This could possibly be due to a problem with calculating the exposure time. Due to sustained IgM positivity in chronic stages of T. gondii infection, the detection of IgM antibodies may not necessarily establish acute infection [21]. Acute T. gondii infection can also indicate an increased chance of transfer from mother to child [22]. According to a previous study, the probability of congenital infection from acute *T. gondii* infection during pregnancy is about 50% in the absence of treatment [23]. Early detection of infections in pregnant women is critical for initiating interventions to limit the risk of transmission and potential consequences for the new-born. As a result, screening for *Toxoplasma* infection during prenatal care should be included as part of the antenatal investigation.

DAJN centre had the most seropositive patients, followed by FAF, based on the location of the health facilities. It could be that this is because both facilities see a lot of pregnant women from across the council area. Furthermore, no disparities between participants from the five health facilities investigated were found.

Seroprevalence was observed to be greater in age groups in the current investigation (15-30 years). Seropositivity did not appear to increase with age in this study. Previous researchers have found that seroprevalence of *T. gondii* infection increases with age, as seen in ages 35-38 years and >48 years, as reported by **Yohannes et al.** [24] and **Tegegne et al.** [25]. The observed disparity in infection rates could be attributed to the age classification of research participants in the current investigation. This could also be due to a lack of personal cleanliness. This emphasizes the importance of continuing to educate women of childbearing age about toxoplasmosis prevention. Seropositivity was not statistically significant as a function of age.

The few unmarried (single) subjects in this study, on the other hand, exhibited a higher *T. gondii* infection seroprevalence. In Rwanda, similar conclusions were reported [26]. This is most likely due to societal variables such as adventurous outdoor eating combined with the fact that they are not married, which may make them more susceptible to toxoplasmosis.

Seroprevalence was found to be considerably greater in persons with only a primary school education in this investigation. Seropositivity to *T. gondii* was observed to decrease with education, which is consistent with the findings of previous researchers who showed that lower levels of education were linked to an increased risk of toxoplasmosis [27]. Containers, knives, cutting boards, and other preparation surfaces contaminated with raw meat can spread Toxoplasma gondii.

People with lower levels of education may be less likely to wash cutting boards, knives, and other kitchen implements with soap after cutting raw meat. This could explain the high prevalence in this study's participants with only an elementary school education. However, just a few of the non-educated persons tested negative for *T. gondii* infection. In this investigation, seropositivity to *T. gondii* did not differ significantly by age, marital status, or educational level.

The participants' occupation was discovered to be a major factor contributing to Toxoplasma infection in this study. The acquisition of T. gondii through contaminated settings could be the cause of the high prevalence among students, followed by dealers. This helps to explain why most seropositive cases were discovered among students and dealers, respectively. This finding is consistent with studies that have identified occupation as a factor that impacts the chance of contracting T. gondii infection [28]. There was a substantial link between individuals' occupation and T. gondii infection seropositivity, confirming findings of Paul et al. [18].

The prevalence of positive *T. gondii* infection was quite high among pregnant women in their first trimester (30.2%) and second trimester 31.0%, but this was not statistically significant. This is the time after infection when the risk of foetal transfer is greatest if no treatment is given [29]. In addition, earlier research has revealed that *T. gondii* infections acquired by a mother during the second or third trimester had a higher (up to 68%) probability of infecting their unborn babies, with less severe repercussions [20]. As a result, almost all the seropositive pregnant women identified in this study are at risk of passing the virus on to their unborn children. This research is in line with what has been reported in Brazil and Sri Lanka [16, 30].

Seropositivity was found to be greater among pregnant women who had experienced two miscarriages [3 (42.9%)]. This indicates that toxoplasmosis may be a risk factor for miscarriage. In this investigation, there was no significant relationship between seropositivity for *T. gondii* infection and miscarriage. This is consistent with reports of *T. gondii* infection and abortion history in pregnant women in Jimma, Ethiopia [31].

It was discovered that pregnant women living in rural areas had a greater seroprevalence than those living in urban areas. Living in rural locations has been found to be a risk factor for *T*. *gondii* infection, implying that low socioeconomic level, difficulties obtaining health services, high exposure, and a lack of understanding about disease transmission contribute to a high prevalence [32]. Although not statistically significant, the location of residence was linked to a high seroprevalence of *T. gondii* infection. Similar findings have been reported by others [33].

The HIV status of those who tested positive for *T. gondii* infection was mostly HIV negative [51 (33.8%)]. The study participants' HIV status had no bearing on their seropositivity to *T. gondii* infection.

*Toxoplasma gondii* transmission is influenced by a pregnant woman's cooking habits and preferences, particularly the type of meat she prefers. According to the findings, seroprevalence was higher in individuals who did not taste meat while cooking than in those who did. This could be owing to those who refused to try meat because it has been proven that food animals, such as poultry, beef, and mutton, are a major source of *T. gondii* infection [34]. Even though most Nigerian households taste meat for flavour during and before cooking, it was not recognized as a significant risk factor for *Toxoplasma* infection in this study. This finding contradicts prior findings in Sri Lanka [16].

Seropositivity was more commonly found among pregnant women who preferred their meat cooked till tender. This could be attributable to the sorts of meat eaten and the rate at which animals become infected. Furthermore, transmission could occur through the use of plates or containers to serve the meat, as well as exposure to household pets. This study found no evidence that eating meat frequently was a significant risk factor for *T. gondii* infection seropositivity.

Seroprevalence was found to be greater in pregnant women who ate raw or undercooked vegetables in the current investigation. There was no link seen between *T. gondii* seropositivity and raw or undercooked vegetable consumption. In contrast, studies in Central Ethiopia [35] and Lagos, Nigeria [13] found that there was association. This finding is consistent with the findings of a study conducted at Felege Hiwot Hospital [33], which found no link between eating raw/unwashed vegetables and fruits and *T. gondii* infection. The observed disparities could be related to differences in the study population's food habits and hygienic practices.

Cat ownership was not shown to be linked with *T. gondii* seropositivity in this investigation. Studies in Egypt [36] and Burkina Faso [37] found similar results. These findings contradict recent research [13, 38], which found that cats play a key role in T. gondii epidemiology, as the only known source of infective oocyst contamination in the environment. This discrepancy among studies could indicate that the risk of developing T. gondii infection is not solely due to the presence of cats in the home, but also due to exposure to faeces from a cat shedding oocyst while gardening. Epidemiological surveys make it difficult to assess the link between cats and toxoplasmosis since frequent exposure to cat faeces or a lack of preventative measures may increase the likelihood of toxoplasmosis transmission. Cats in Jos are not kept indoors and are permitted to roam freely, polluting the environment severely. Seropositivity in relation to owning a dog, pig, or other animal was not found to be significantly linked to T. gondii infection. Other research in Ethiopia have revealed similar results [17].

Regular soil exposure was revealed as a significant risk factor for Toxoplasma IgG seropositivity in the current study. The soil might be regarded as a potential source of human infection. Regular soil contact also means coming into direct contact with cat faeces-contaminated soil. Cat oocysts can live for several years if they are exposed to the right conditions [39]. This explains why soil exposure is a substantial risk factor for T. gondii IgG serology positivity. Furthermore, protective equipment like as gloves is rarely worn, resulting in infective oocysts in the soil being mistakenly consumed by humans during gardening. The findings of this investigation are consistent with Sri Lankan reports [16].

This study discovered a greater frequency among pipe-borne water users based on the sources of drinking water. The relationship between the source of drinking water and seropositivity for *T. gondii* infection was statistically significant. This is in line with reports that have been documented in many parts of the world as having an impact on *Toxoplasma* transmission [33].

In general, the lack of a statistically significant association between *Toxoplasma* infection seroprevalence and these putative factors does not imply that they have no impact on toxoplasmosis transmission. However, it is possible that such conditions play a little effect in the transmission of the parasite in the analysed subjects in the study area.

#### Conclusion

The overall seroprevalence of T. gondii infection among pregnant women in Jos, Jos-North Plateau State, was not high. Only the participants' occupation was found to be substantially linked with T. gondii seropositivity, out of all the sociodemographic characteristics studied. Regular contact with dirt and the source of drinking water were found to be the most important predictors of T. gondii infection among the study participants. They have also been shown to have an impact on Toxoplasma transmission in many places of the world. As a result, health extension workers and other public health professionals should emphasize regular serological testing during pregnancy, as well as health education to all pregnant women and women considering pregnancy, to prevent the condition.

#### **Conflict of interest statement**

We declare that we have no conflict of interest.

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