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Seroprevalence of Toxoplasma Gondii IgG and IgM Antibodies and Risk Factors Among Pregnant Women in Pankshin Central, Plateau State, Nigeria

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Abstract **Original Research Article**

Background: Toxoplasmosis is a zoonotic disease caused by an obligate intracellular parasite called Toxoplasma gondii. Severe complications have been found associated with the disease in pregnancy such as stillbirth, seizures, microcephaly etc. yet, little or no attention has been given to the disease.

Aim: The aim of this research is to determine the seroprevalence of toxoplasmosis in pregnant women and the risk factors associated with the disease in Pankshin.

Methods: Blood samples were collected from 182 pregnant women who consented at the Pankshin General Hospital. Detection of Toxoplasma gondii IgM and IgG antibodies was carried out using commercially prepared Human Tox-IgG/IgM Cassettes (Innovation Biotech Beijing Co Ltd, China) according to prescribed protocol. Structured questionnaires specifically design for this study was used to obtain socio-demographic data and the associated risk factors.

Results: A total number of one hundred and eighty-two (182) pregnant women were examined, out of which thirty-four (34) were sero-positive for toxoplasma gondii antibodies while one hundred and forty-eight (148) were sero-negative. Of the 34 subjects (18.6%) that were sero-positive to T. gondii antibodies, 15(8.2%) were sero-positive to IgM, 14(7.7%) were sero-positive to IgG and 5(2.7%) were sero-positive to both IgM + IgG. Seropositivity to T. gondii increased with low educational level and increased trimester. Subjects who were more at risk of toxoplasmosis were those who keep cats, taste meat while cooking and eat dog meat.

Conclusion: Since toxoplasmosis is prevalent among population under study, government attention should be drawn to providing specific diagnostic resources in hospital laboratories and create awareness to the general public about the disease.

Keywords: Toxoplasmosis, Toxoplasma Gondii, Seroprevalence, Pregnant Women, Risk Factors, Pankshin, Zoonotic Disease, IgM, IgG, Cat Ownership, Undercooked Meat, Public Health Awareness.

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1.0 INTRODUCTION

Toxoplasma gondii causes the disease toxoplasmosis. It is reported that one third of the world's population is infected with the parasite (Akubuilo *et al;* 2020). It is an obligate intracellular protozoan distributed globally in humans and other warm-blooded animals and it belongs to the feline (cat) family which is the definitive host (Ocheme *et al;* 2023). Humans become infected by *T. gondii* through the ingestion of undercooked meat containing tissue cysts (tachyzoites and bradyzoites) or by the accidental ingestion of sporulated *oocysts* in food, water and soil contaminated with cat faeces (Lass *et al*., 2009 and Robert, 2012). When sporulated *oocyts* are ingested by an intermediate host such as humans, the released *sporozoites* form permanent cysts in various organs and tissues, but most commonly in the brain (Torgerson,& Mastroiacovo 2013). Infection is usually asymptomatic or associated with flu-like symptoms in more than 80% of immunocompetent individuals. But when primary infection occurs during pregnancy, *T. gondii* can cross the placenta and may be vertically transmitted to

the foetus resulting in congenital toxoplasmosis. This may cause disorders such as; abortion, stillbirth, microcephaly, hydrocephalus, chorioretinitis and even death (Fanigliulo *et al;* 2020). Similarly, toxoplasmosis can also be transmitted via organ transplantation and blood transfusion from an infected donor (Hamida *et al;* 2023).

In the environment, the *oocysts* can resist freezing and moderately high temperatures to remain viable for years in soil and water (Jones & Dubey, 2010). The rate of congenital transmission and the degree of severity of toxoplasmosis in foetuses vary depending largely on the stage of gestation and the time of infection. It is estimated that 6.5 million children under the age of five (5) years are affected by birth defects globally every year and out of this number, 3.3 million of them die while 3.2 million that survive may be disabled for life (WHO 2025) . Despite the severe congenital defects associated with toxoplasmosis, the disease has been greatly neglected in Nigeria. It is on this premise that this study is aimed at determining the prevalence of toxoplasmosis and the risk factors associated with *T. gondii* infection in antenatal clinic patients at Pankshin General Hospital, Plateau State.

2.0 MATERIALS AND METHODS

2.1. Study Area/Population

This study was carried out at the General Hospital, Pankshin Local Government Area of Plateau State involving pregnant women who registered for antenatal clinic during the time of study. This is because, the healthcare centre is strategically located, has quality facilities as well as quality services and is relatively cheap as a result, most pregnant women attend antenatal clinic in the centre.

2.2. Study Design

This research is a hospital based, cross-sectional study conducted for the period of three (3) months (October to December, 2018) involving pregnant women who registered for antenatal clinic at Pankshin General Hospital.

2.3. Ethical Approval

Study permission was obtained from Plateau State Specialist Hospital and Plateau State Hospitals Management Board, Jos with Reg. Nos: NHREC/05/01/2010b and HMB/ADM/423/T/32 respectively.

2.4. Inclusion/Exclusion Criteria

All pregnant women who registered for antenatal clinic during the period of study and consented were included while those that did not give their consent were excluded from the study.

2.5 Sample Size

The sample size was determined using the formula describe by (Charan et al., 2021). Thus,

$$n = \frac{t^2 \times p (1-p)}{m^2}$$

Where: n= required sample size, t= confidence level at 95% = 1.96

p= established local prevalence 12.1%) Sowemimo et al., 2018)

, m= margin of error at 5%= 0.05

Hence, N= 163.44 ~ 163

Therefore, the sample size was one hundred and sixty-three. However, one hundred and eighty-two (182) was used as the sample size since it is with adequate sample size that one can make generalizations

2.6. Sample/Data Collection

About 5ml of blood was aseptically collected from vein puncture of each subject who gave their consent. Blood samples collected were allowed to clot and centrifuged at 4000 rpm for 5 minutes to obtain the serum after which, Pasteur pipettes were used (one for each blood sample) to transfer serum from plain tubes into cryovial tubes. Structured questionnaire specifically designed for this study was used to obtain demographic data and risk factors from patients. Cryovial tubes containing serum for each antenatal day were shipped in a shipment box stocked with ice packs from Pankshin to Jos (Plateau State Human Virology Research Centre) where they were stored at -25^{o}C temperature.

2.7. Detection of Toxoplasma Gondii Antibody

The detection of *Toxoplasma gondii* IgM and IgG antibodies was done using a commercially prepared Human Tox-IgG/IgM Cassettes (Innovation Biotech, Beijing Co Ltd, China). This device uses a highly specific antibody-antigen reaction and chromatographic immunoassay technology for the qualitative detection of *Toxoplasma gondii* IgG and IgM antibodies in human blood. The test kit contains recombinant *Toxoplasma gondii* IgM antigen which is pre-coated to the membrane in the test region (T1), recombinant *Toxoplasma gondii* IgG which is pre-coated to the membrane in the test region (T2) and the corresponding antibodies antibodies in the quality control region (C). When two to three drops of blood sample were added into well (S) of cassette, it reaches with pre-colloidal gold particles in the test. The mixture migrates chromatographically along the length of the test cassette and interacts with the immobilized anti-human IgG and IgM. If positive, colloidal gold will bind with anti-*Toxoplasma gondii* antibodies in the process of chromatography, the mixture will bind with the antigen which was pre-fixed on the membrane, a red line appears in the region T (T1 and T2). If negative, no red line will appear in the test regions (T1 and T2).

All sera samples that were already labelled were removed from -25^{o}C temperature and kept at 2-8^{o}C for five (5) hours to thaw.

2.8. Test Procedure

- All sera samples that were already labeled was removed from -25^{o}C temperature and kept at 2-8^{o}C for five (5) hours to thaw.

- The test devices (cassettes) were removed and kept at room temperature (18ºC-30ºC).
- The cassettes were removed from the foil pouch and placed on a cleaned and leveled surface.
- Using a marker, cassettes were labeled 1,2,3,4,…….182
- A disposable specimen dropper was used to transfer Two (2) drops of specimen from cryovial tubes into the sample well labeled (S) on the Cassette.
- Results were read after 15 minutes which was indicated by the appearance of a red line as the case may be.

2.8. Interpretation of results

Negative: Only the control line (C) is visible on the cassette.
IgM Positive: The control line (C) and IgM line (T1) are visible on the device.
IgG Positive: The control line (C) and IgG line (T2) are visible on the cassette.
IgM and IgG Positive: The control line (C), IgM (T1) and IgG line (T2) are visible on the device.

2.9. Data Analysis

The data collected from the questionnaire and the laboratory tests were entered into the Statistical Package for Social Sciences Programme (SPSS, Version 23). The data was subjected to chi square test for association.

3.0. RESULTS

3.1. Prevalence of Toxoplasmosis in the Study Population

A total number of one hundred and eighty-two (182) pregnant women were examined, out of which thirty-four (34) were sero-positive for *toxoplasma gondii* antibodies while one hundred and forty-eight (148) were sero-negative. Of the 34 subjects (18.6%) that were sero-positive to *T. gondii* antibodies, 15(8.2%) were sero-positive to IgM, 14(7.7%) were sero-positive to IgG and 5(2.7%) were sero-positive to IgM + IgG (figure1). This means that, most of the population under study had recent infection (IgM), others had previous infection (IgG) and some had both recent and previous infections (IgM/IgG)

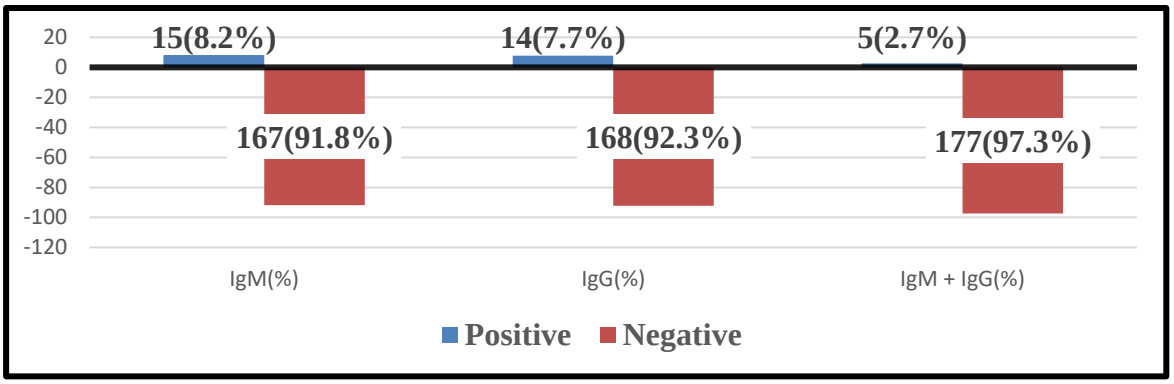

Figure 1: Distribution of *Toxoplasma gondii* antibodies among Antenatal Clinic Patients (n =182) in Pankshin General Hospital, Plateau State, Nigeria.

Table 1: Demographic distribution of Toxoplasma gondii antibodies among the study

	No. examined	No. (%) Positive (IgM)	No. (%) Positive (IgG)	No. (%) Positive IgM + IgG
Distribution of *Toxoplasma gondii* antibodies among the study subjects in relation to age				
12 – 21	19	1(5.3)	0(0.0)	0(0.0)
22 – 31	119	12(10.1)	12(10.1)	4(3.4)
32 – 41	34	1(2.9)	2(5.9)	1(2.9)
≥ 42	4	1(25.0)	0(0.0)	0(0.0)
p-value		0.34	0.41	0.86
Distribution in relation to Marital Status				
Single	14	0(0.0)	0(0.0)	0(0.0)
Married	160	14(8.8)	14(8.8)	5(3.1)
Widowed	2	1(50.0)	0(0.0)	0(0.0)

P-value		0.06	0.47	0.77
Distribution in relation to occupation				
Farming	20	2(7.2)	1(5.0)	0(0.0)
Business	69	5(10.0)	3(4.3)	1(1.4)
Civil servant	22	3(13.6)	4(18.2)	2(9.1)
Housewife	35	2(5.7)	3(8.6)	1(2.9)
Students	28	2(7.1)	3(10.7)	1(3.4)
Artisants	2	1(50.0)	0(0.0)	0(0.0)
p-value		0.33	0.41	0(0.0)
Distribution in relation to Location				
Rural	60	4(6.7)	3(5.0)	0(0.0)
Semi-urban	22	0(0.0)	3(13.6)	0(0.0)
Urban	94	11(11.7)	8(8.5)	5(5.3)
p-value		0.17	0.42	0.11
Distribution in relation to trimester				
First	16	2(12.5)	0(0.0)	0(0.0)
Second	58	2(3.4)	3(5.3)	1(1.7)
Third	102	11(10.8)	11(10.8)	4(3.9)
p-value		0.23	0.21	0.56

Table 2: Distribution of Toxoplasma Gondii Antibodies in Relation to Risk Factors

Antibodies	**No. examined**	**No. Yes (%)**	**No. No (%)**	**χ2**	**df**	**p-value**
Distribution of antibodies in relation to ownership of cats						
IgM	15	12(80.0)	3(20.0)	0.002	1	0.96
IgG	14	10(71.4)	4(28.6)	0.616	1	0.43
IgM + IgG	5	4(80.0)	1(20.0)	0.001	1	0.98
Distribution of antibodies in relation to dog meat consumption						
IgM	15	12(80.0)	3(20.0)	1.149	1	0.28
IgG	14	9(64.3)	5(35.7)	0.077	1	0.78
IgM + IgG	5	5(100.0)	0(0.0)	2.465	1	0.12
Distribution of T. gondii antibodies in relation to tasting of meat while cooking						
IgM	15	8(53.3)	7(46.7)	0.032	1	0.86
IgG	14	9(64.3)	5(35.7)	1.052	1	0.31
IgM + IgG	5	4(80.0)	1(20.0)	1.716	1	0.19
Distribution of T. gondii antibodies in relation to pork consumption						
IgM	15	6(40.0)	9(60.0)	0.255	1	0.61
IgG	14	6(42.9)	8(57.1)	0.520	1	0.47
IgM + IgG	5	2(40.0)	3(60.0)	0.080	1	0.78

DISCUSSION

This result indicated a lower prevalence (18.6%) compared to those obtained from other parts of the world. Among which are, India, France, Southern Ethiopia and Port Harcourt, Nigeria with 22.4%, 37.0%, 23.9% and 65.6% respectively (Singh *et al.*, 2014; Tourdjman *et al*., 2015; Sowemimo *et al*., 2018 & Jula *et al.*, 2018). Variation in prevalence as seen above could be attributed to differences in climatic conditions such as high temperature, relative humidity etc., which favours the viability of Oocysts in an environment awaiting favourable conditions to infect other intermediate host. Pankshin L.G.A. especially the Northern part where this research took place has an extremely low temperature with high relative humidity (Avuwa *et al.*, 2022). For this reason, the viability of the Oocysts may not be adequately supported. Similarly, sample size may also be responsible for the variation observed in prevalence in that, a larger sample size is likely to produce a higher prevalence as well as a significant association compared to a smaller sample size. In this research, a smaller sample size (182) was used compared to those that have been reported (Singh *et a*l., 2014; Tourdjman *et al*., 2015; and Jula *et al.*, 2018) as a result, no significant association was established among studied variables. However, it is also possible to obtain a higher prevalence with a lower sample size especially when there is high infectivity (Oboro *et al.*, 2016; Sowemimo *et al*., 2018).

Countries that have high sanitation standards and hygienic practices are likely to reduce transmission of the disease unlike their counterparts who do not see sanitation and personal hygiene as important. Eating habits may also be responsible for the variation in prevalence in this research. The consumption of raw and undercooked meat is a common practice in Asian countries such India which indicated high prevalence (Singh *et al.*, 2014). However, low prevalence was obtained in this study likely because such practice is not common in this part of the world. Socio-economic and literacy status of studied subjects is likely to be responsible for the variation indicated in this study. Countries that are socio-economically stable with high literacy level are less likely to be exposed to *Toxoplasma gondii* infection compared to their counterparts with low socio-economic stability and literacy level. Similarly, in this study, most subjects infected by toxoplasmosis had low literacy level.

Prevalence in terms of individual antibodies indicated that, 8.2% (15) were positive for IgM, 7.7% (14) were positive for IgG and 2.7% (5) were positive for IgM+IgG. This implies that, as at the time of this research, the studied population was still being exposed to *T. gondii* which is evident in the 8.2% IgM obtained in this research. Similarly, the 7.7% IgG showed that before the time of this research, the study subjects were since exposed to *Toxoplasma gondii* and it is likely that the disease has been with the population without knowing as reported in Pakistan (Sadiqui et al., 2019).

5.2: Distribution of *Toxoplasma Gondii* Antibodies in Relation to Demographic Factors

It was revealed in this study, that out of 34 positive cases obtained, the age group most affected was between 22-31years (23.5%) followed by 32-41 years (11.8%). This is in line with the study reported in Sudan (Wahaj *et al*., 2018) suggestive of the child bearing age and sometimes, the depressed nature of the immune system in pregnancy. However, seropositivity to *T. gondii* has been reported among subjects with higher ages which also showed the opportunistic nature of the parasite as described by (Wilking *et al*., 2016; Abdul *et al.*, 2016),

Our research showed that based on marital status, subjects that were married constituted the highest number of those that were seropositive to *T. gondii*. This may be attributed to the fact that, most of these women are engaged in meaningful activities such as farming that will bring food to their table as a result; they become more exposed to the parasite. However, statistically, no significant difference was established between seropositivity to *T. gondii* and marital status. P>0.05. Most subjects that were seropositive to *T. gondii* were into business especially perishable goods. This is because, Pankshin is strategically located such that, it links Mangu LGA to Langtang North and some parts of Bauchi State which makes it a commercial centre. In addition, the two higher institutions of learning; Federal College of Education and State College of Health and Technology, Pankshin, all sited in Pankshin town is a contributory factor to the high involvement of the study subjects into business. This is similar to the study reported from Southern Ethiopia (Sadiqui et al., 2019).

Even though; no significant difference statistically was established between seropositivity to *T. gondii* with occupation. Furthermore, in Saudi Arabia, a significant relationship statistically was established between occupation and seropositivity (Abdullah *et al*., 2017). This variation could be as a result of differences in sample size since higher sample size as well as high number of subjects being seropositive would likely produce significant difference statistically. It was also observed that subjects with lower educational level were at higher risk of contracting toxoplasmosis than their highly educated counterparts. The reason is that, with high educational level, an individual is most likely to be aware of some personal hygienic practices that can prevent infection by the parasite unlike the uneducated. This is in line with researches conducted in Borno State, Nigeria, and Pakistan (Oyinloye *et al*., 2014 and Abdul *et al.*, 2016) respectively. However, no significant association was established statistically between seropositivity to *Toxoplasma gondii* and educational level (P>0.05).

Most subjects seropositive to *T. gondii* antibodies were from the urban centre (Pankshin town) where they reside to do their businesses. While doing their businesses, they may consume raw vegetables and poorly prepared or roasted meat that are hawked or sold on the streets which may serve as sources of infection. Most of those seropositive to *T. gondii* antibodies were in their third

trimester indicating that infection was acquired after being pregnant which confers higher risk of congenital transmission than those acquired before pregnancy. This is in line with the research conducted in South-Western Ethiopia and Kano (Abdul *et al.*, 2016; Yusuf *et al*., 2016) respectively

5.3 Distribution of *Toxoplasma Gondii* Antibodies in Relation to Risk Factors

It was found that subjects that own cats were more infected by *Toxoplasma gondii* than their counterparts who did not own cats. This is an indication that those infected might have had contact with cat litters containing sporulated Oocyst responsible for their seropositivity. This is similar with other studies that were reported from India, South-Western Ethiopia and Kano (Singh *et al*., 2014; Abdul *et al.*, 2016; Yusuf *et al.*, 2016 and Jula *et al.*, 2018) respectively.

However, other researchers have reported increase in seropositivity to history of contact with cats from Pakistan and Sudan (Sadiqui et al., 2019 & Wahaj *et al*., 2018) respectively. Subjects that were mostly infected by *Toxoplasma gondii* eat dog meat which is a common Practice among people of Pankshin L.G.A; they eat it as a delicacy either cooked or roasted. This might be that during preparation, the meat is not adequately cooked or roasted as a result tissue cyst that would have been destroyed by heat, is ingested alongside with the meat. This is in line with the study reported from Pakistan (Sadiqui *et al*., 2019) . In this research, most subjects seropositive to *Toxoplasma gondii* taste meat while cooking. This means that, tissue cyst might be ingested by the subjects without knowing while tasting. However, no Significant difference was established statistically, which might be due to the low seropositivity, unlike the study reported from Port Harcourt, Nigeria which showed a significant association due to high seropositivity to *T. gondii* antibodies (Oboro *et al.*, 2016; Sowemimo *et al*., 2018) Similarly, most subjects who were seropositive to *T. gondii* do not consume pork as seen in this study. This may be attributed to the fact that; pork consumption is not a common practice in Pankshin as a result; their seropositivity to *T. gondii* may be due to other causes. However, other researchers reported significant relationship between seropositivity to *T. gondii* and pork consumption (Sadiqui *et al*., 2019).

5.4 CONCLUSION

In conclusion, this study highlights a relatively low prevalence of seropositivity for *Toxoplasma gondii* antibodies among pregnant women, with 18.6% testing positive. The presence of IgM, IgG, and both antibodies indicates varying stages of infection. While demographic factors, such as lower educational levels, increased trimester and perishable goods business were associated with higher seropositivity rates, no significant association was found. Additionally, risk factors such as cat ownership, tasting meat while cooking, and consuming dog meat were linked to a higher likelihood of infection, though none of these associations were statistically significant. These findings suggest the need for inclusion of routine screening for toxoplasmosis in pregnant women during antenatal, continued public health education on toxoplasmosis and further investigation into the risk factors contributing to its transmission.

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