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Prevalence and Associated Risk Factors of *Toxoplasma gondii* Antibodies among Pregnant Women Attending Maiduguri Teaching Hospital, Nigeria

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Toxoplasma gondii (*T. gondii*) is one of the few known zoonotic parasites that have the ability to induce miscarriage and congenital transmission. This study sought to determine and update the prevalence of anti-toxoplasma IgM and IgG antibodies in pregnant women and their associated risks at the University of Maiduguri Teaching Hospital, Maiduguri, Nigeria. Blood samples from 360 pregnant women aged 19-42 years were analyzed for anti-toxoplasma IgM, IgG antibodies and IgG avidity using Euroimmun® ELISA kits. Structured questionnaires were used to obtain participants' sociodemographic data. Out of the 360 pregnant women studied, 32 (8.9%) and 144 (40.0%) were seropositive for *T. gondii* specific IgM and IgG antibodies, respectively. Of those with IgM seropositivity, 26 (7.2%) had primary infection (i.e., IgM+IgG-low IgG avidity) while 6 (1.7%) had reactivated infection (IgG+IgM+high IgG avidity). The IgM seropositivity was statistically significant among women who kept cat pets than those that don't (75 vs. 25%, p = 0.025). The IgG seropositivity rate was significantly higher among pregnant women from urban than those from rural residential areas (48.4 vs. 19.2%, p = 0.001). Likewise employed women were more likely to contract primary toxoplasmosis than the unemployed (82.2 vs. 31.6%, p = 0.0025). Serological evidence of primary toxoplasmosis was significantly high among pregnant women studied while a significant proportion of other women were at risk of contracting primary toxoplasmosis. Screening for toxoplasmosis during antenatal care should be encouraged in order to detect infected women so that appropriate clinical modalities can be instituted.

Key words: Toxoplasmosis, serology, antenatal screening, IgG avidity

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INTRODUCTION

Toxoplasma gondii (*T. gondii*) is an obligate intracellular coccidian parasite that has a complex life cycle alternating between asexual reproduction taking place in several tissues of vertebrates (intermediate hosts) and sexual reproduction taking place in epithelial linings of the digestive tract of cats (definitive host) (Skariah *et al*., 2010). Cats become contaminated when they ingest animal flesh or food encysted with *T. gondii* and rarely by ingesting oocysts directly from feces of other infected cats (Skariah *et al*., 2010).

Humans become infected either congenitally (mother-to-fetus *in-utero* transmission), through food or water contaminated with cat feces or by eating under-cooked meat of infected animals (Skariah *et al*., 2010). In fact, toxoplasmosis was once a leading infectious cause of food-borne death after salmonellosis and listeriosis in the USA (Jones *et al*., 2001). Rarely can it be transmitted through organ transplantation and transfusion of infected blood (Skariah *et al*., 2010; Jones *et al*., 2001). Infected cats are usually asymptomatic and begin to shed unsporulated oocysts which are noninfectious (up to 10^6/day) in their feces 7-14 days after exposure (Dubey *et al*., 2009). Most cats shed oocysts only once in their lifetime (Cook *et al*., 2000). Within days to weeks, the oocytes sporulate and become infectious. Oocysts survive best in warm and humid conditions and thus can remain infectious for many months (Dubey and Jones, 2008). Oocysts also withstand exposure to freezing for up to 18 months, especially if they are covered and out of direct sunlight as such they can be transmitted on fomites (Dubey and Jones, 2008).

After ingestion by susceptible human host, oocysts release sporozoites, which change into tachyzoites. These tachyzoites are present during acute infection and are capable of invading cells and replicate (Dubey and Jones, 2008). They are disseminated widely and circulate from 3-10 days in immunocompetent hosts before changing into bradyzoites and forming cysts in tissues (Dubey and Jones, 2008). These cysts remain present during latent infection. Once infected, humans are believed to remain infected for life. Unless immuno suppression occurs and the organism reactivates, human hosts usually remain asymptomatic (Dubey and Jones, 2008).

Infections due to *T. gondii* are considered a worldwide zoonosis of great public health importance. Epidemiological studies suggest that prevalence of *T. gondii* infection in pregnant women varies greatly among different countries; in Europe it varies from 9-63% (Masini *et al*., 2008). In Korea, the seroprevalence of toxoplasmosis was reported as low as 3.7% (Han *et al*., 2008) while the prevalence is as high as 41.6-66.9% in other Asian countries such as India and Jordan (Borkakoty *et al*., 2007). Seroprevalence varies considerably with high seroprevalence (over 50%) occurring in countries where raw meat is commonly eaten and in tropical regions of Latin America or Sub-Saharan Africa where cats are numerous and the climate is favourable for oocysts survival (Borkakoty *et al*., 2007).

In pregnancy, the most common mechanisms of acquiring infection are through consuming raw or very undercooked meats or contaminated water, or exposure to soil (farming or gardening without gloves) or cat feces (Elmore *et al*., 2010). It has been shown that over 90% of women who contract *T. gondii* infection remain asymptomatic and spontaneously recover (Kravetz and Federman, 2005), only a small proportion will develop clinical signs of the disease (Di Carlo *et al*., 2008). The clinical presentation in pregnant women is not more severe than in non-pregnant women and most often occurs as an influenza-like illness (low-grade fever, malaise, cervical lymphadenopathy) with an incubation period of 5-18 days following exposure (Paquet *et al*., 2013).

Toxoplasma gondii infection can be detected by the help of serological testing, amniocentesis or by the presence of abnormal ultrasound findings. Serologic testing is often the first step in diagnosis through the use anti-toxoplasma specific IgG and IgM antibodies. However, there exist some diagnostic challenge associated with differentiating between a primary and a chronic infection; and results of IgG and IgM testing can often be difficult to interpret. For this reason, it is important to consult an expert clinical parasitologist when confirming diagnosis (Paquet *et al*., 2013).

Serodiagnosis of toxoplasmosis is usually achieved by detecting IgG and IgM against *T. gondii*. In addition, IgG avidity test is an important additional test routinely performed in most developed societies. Low IgG avidity suggest acute infection while high IgG avidity confirms chronic or reactivated infection (Liesenfeld *et al*., 1997). The presence of IgM antibodies alone cannot be considered reliable for making a diagnosis of acute toxoplasmosis infection. The IgM antibody titres rise from 5 days to weeks following acute infection, reaching a maximum after 1-2 months and decline more rapidly than IgG (Liesenfeld *et al*., 1997). Although, IgM antibodies can decrease to low or undetectable levels, in many cases they may persist for years following the acute infection. The IgG antibodies appear later than IgM and are usually detectable within 1-2 weeks after the infection, with the peak reached within 12 weeks to 6 months after acute infection. They will be detectable for years after acquired infection and are usually present throughout life (Liesenfeld *et al*., 1997).

Knowing when infection occurred during pregnancy is important in evaluating the risk of congenital transmission, initiating antibiotic therapy and ensuring appropriate prenatal counseling. The IgG avidity test has been prescribed for such utility. The IgG avidity measures the strength of IgG binding to the organism. Avidity, in most cases but not all, shifts from low to high after about 5 months. If the avidity is high, this suggests infection occurred at least 5 months before testing (Montoya, 2002). Transmission to fetus occurs predominantly in women who acquire primary infection during pregnancy.

In view of these, this study sought to determine and update the prevalence of anti-toxoplasma IgM and IgG antibodies of pregnant women and their associated risks at University of Maiduguri Teaching Hospital, Maiduguri,

Nigeria with the hope to detect acutely infected women and those at risk for congenital transmission so that appropriate clinical intervention can be instituted.

MATERIALS AND METHODS

Study area: This cross-sectional study was carried out in the University of Maiduguri Teaching Hospital, Maiduguri, Borno state, Nigeria. The study was approved by the Ethical Research Committee, University of Maiduguri Teaching Hospital, Nigeria. Maiduguri, the capital city of Borno state, Nigeria, located in northeastern Nigeria shares borders with neighboring countries, such as Niger Republic, Chad and Cameroon. Within Nigeria, Maiduguri shares borders with other states, such as Adamawa, Yobe and Gombe and has Sahel savannah vegetation. The annual average temperature of Maiduguri ranges from 19.1-34.7°C and average annual precipitation is 562 mm. In Nigeria, there has been no consensus of opinion regarding conducting *T. gondii* screening or other "TORCH" panel tests for pregnant women during their antenatal visits.

Sample size calculation: The sample size was determined from using the prevalence rate of the study conducted by Deji-Agboola *et al*. (2011) in Nigeria with overall seroprevalence of 40.2%, therefore, the minimum sample size at 95% confidence level was 379. However, only 360 women voluntarily consented to participate in the study.

Participants and settings: This study was conducted on 360 blood samples of pregnant women attending antenatal clinics of the UMTH who consented to participate in the study and excluded those who declined to participate in the study or refused to consent. The Median age of the women was 30 years with range of 18-40 years.

Data collection: Structured Questionnaire interview was used to collect demographic data, such as age, place of residence, gestational age, educational status, occupation, marital status, whether they keep cat pets or not.

Sample collection and preparation: Samples were collected between January and April, 2014. Five milliliter of blood was collected aseptically into plain vacutainer tubes. The tubes were then appropriately labelled with patients' laboratory number. Sera from these blood samples were separated by allowing the blood to clot at room temperature and centrifuged at 3000 rpm for 10 min. The sera were then separated using clean Pasteur pipettes, transferred into serum containers and stored at -20°C for 10 days. After sample collection, the 360 sera were transported to University of Abuja Teaching Hospital, Gwagwalada, Abuja (about 850 km from Maiduguri) in cold chain. Upon arrival they were immediately (<24 h) set for laboratory analysis.

Laboratory analysis: Serum samples were analyzed by enzyme-linked immunosorbent assay (ELISA) using anti-toxoplasma IgM, IgG and IgG avidity kits (Euroimmun®, Leubeck, Germany) with product numbers; EI2410-9601G, EI 2410-9601M and EI 2410-9601-1G, respectively.

Analytical procedure: All samples and reagents were brought to room temperature. The test was performed according to manufacturer's instructions. The Optical Density (OD) was read using a BioRad® ELISA microplate reader at 450 nm wavelength.

Ethical clearance and informed consent: This study was conducted in accordance with the Declaration of Helsinki and the protocol was approved by the human research ethical committees of University of Maiduguri Teaching Hospital, Maiduguri, Borno state, Nigeria. All the subjects gave their written informed consent for inclusion before they participated in the study. All data were analyzed anonymously throughout the study.

Statistical analysis: The data obtained from the questionnaire and the results of the laboratory analysis were entered into Microsoft excel and analyzed using SPSS (statistical package for social sciences Version 20, California Inc., USA). Results obtained were reduced to percentages. The Pearson Chi square at 95% confidence interval and 0.05 level of significance was used to determine the relationships between sociodemographic and antibodies prevalence.

RESULTS

Table 1 shows findings of the serological test conducted on subjects. For purpose of diagnostic convenience, it was classified into 4 humoral immune responses. The first category were those immuned against *T. gondii* infection [IgG (+) and IgM (-)], this comprised of 144 (40.0%) women. In the second category, 6 (1.7%) women had reactivated or chronic *T. gondii* infection [IgG (+) IgM (+) and high IgG acidity indices]. In the third category, 26 (7.2%) pregnant woman had primary *T. gondii* infection [IgG (-) IgM (+) and low IgG avidiy indices]. However, 190 (52.7%) of the remaining women were susceptible to *T. gondii* infection [IgG (-) IgM (-)].

Table 1: Summary of anti-toxoplasma specific IgM and IgG antibodies distribution among pregnant women studied

	Tested		
Antibodies	No.	%	Inference
IgG (+) IgM (-)	144	40.0	Previous exposure
IgG (+) IgM (+) and high IgG avidity indices	6	1.7	Reactivated or chronic infection
IgG (-) IgM (+) and low IgG avidity indices	26	7.2	Primary infection
IgG (-) IgM (-)	190	52.7	Susceptible

Table 2: Age distribution of anti-toxoplasma IgG and IgM among pregnant women

	Subjects tests		IgG seropositivity		IgG seronegativity		IgM seropositivity	
Age (years)	No.	%	No.	%	No.	%	No.	%
≤20	60	16.7	46	76.7	14	23.3	2	0.6
21-25	104	28.9	56	53.8	48	46.1	19	5.3
26-30	92	25.6	32	34.8	60	65.2	7	1.9
≥31	104	28.9	10	9.6	94	90.4	4	1.2
Total	360	100.0	144	40.0	216	60.0	32	8.9

Table 3: Anti-toxoplasma IgG and IgM seropositivity across education level of pregnant women

	Subjects tests		IgG seropositivity		IgG seronegativity		IgM seropositivity	
Education level	No.	%	No.	%	No.	%	No.	%
None	10	4.9	18	100.0	0	0.0	1	0.3
Primary	82	26.4	10	12.2	72	87.8	16	4.4
Secondary	160	43.9	40	25.0	120	75.0	10	2.8
Tertiary	100	24.7	76	76.0	24	24.0	5	1.4
Total	360	100.0	144	40.0	216	60.0	32	8.9

Table 4: Distribution of anti-toxoplasma IgG and IgM seropositivity with places of residence of pregnant women

	Subjects tests		IgG seropositivity		IgG seronegativity		IgM seropositivity	
Place of residence	No.	%	No.	%	No.	%	No.	%
Urban	256	53.8	124	48.4	132	51.5	20	5.6
Rural	104	46.2	20	19.2	84	80.8	12	3.3
Total	360	100.0	144	40.0	216	60.0	32	8.9

Table 5: Distribution of anti-toxoplasma IgG positivity with occupation of pregnant women

	Subjects tests		IgG seropositivity		IgG seronegativity	
Occupation	No.	%	No.	%	No.	%
Unemployed	158	43.9	108	68.4	50	31.6
Employed	202	56.1	36	17.8	166	82.2
Total	360	100.0	144	40.0	216	60.0

Staging of these immunological responses was made possible by virtue of IgG avidity testing conducted on pregnant women with IgM seropositivity.

From Table 2, age group 21-25 had the highest IgG and IgM seropositive subjects, 56 (53.8%) and 19 (5.3%) respectively while IgG and IgM seropositivity was least among pregnant women greater than 31 years and those ≤20 years which are 10 (9.6%) and 2 (0.6%), respectively. There was no significant relationship between age distributions of anti-toxoplasma IgG and IgM seropositivity among pregnant women (p-value>0.05).

From Table 3, pregnant women with tertiary education had the most toxoplasma IgG seropositivity, 76 (76.0%), while only 10 (12.2%) of those with primary education was seropositive. However, pregnant women with primary education had the most toxoplasma IgM seropositivity, 16 (4.4%) while only 1 (0.3%) of those with no formal education was IgM seropositive. There was no significant relationship between education level and distributions of anti-toxoplasma IgG and IgM seronegativity among the pregnant women (p-value = 0.999 and 0.250, respectively).

From Table 4, anti-toxoplasma IgG and IgM seropositivity among pregnant women who reside in urban area, 124 (48.4%) and 20 (5.6%) were significantly higher than those who reside in rural areas, 20 (19.2%) and 12 (3.3%) respectively (p = 0.001 and 0.025).

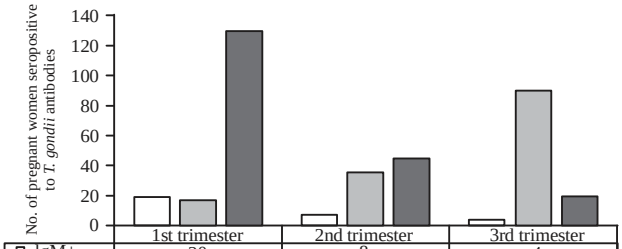
From Table 5, pregnant women who were employed were significantly IgG seronegative (susceptible) and at higher risk of contracting primary *T. gondii* infections (82.2% versus 31.6%, p = 0.001).

From Fig. 1, pregnant women at first trimester had the highest anti-toxoplasma IgM seropositivity and least IgG seropositivity, 20 (62.5%) and 18 (12.5%) respectively while those at third trimester had the least IgM seropositivity and highest IgG seropositivity 4 (12.5%) and 90 (63.9%), respectively. There was no significant relationship between anti-toxoplasma antibodies and gestational age (p-value = 0.065).

From Fig. 2, pregnant women with domesticated cat pets exhibited significantly higher anti-toxoplasma IgM and IgG antibodies than those without cat pets, [24 (75%), 8 (25%)] versus [112 (77.8%), 32 (22.2%)], respectively.

DISCUSSION

To the best of our knowledge, this is the most recent study from Nigeria to explore the prevalence of *T. gondii* antibodies among one of the most largely studied category of subject i.e. pregnant women. The last published study was that of Deji-Agboola *et al*. (2011).



	1st trimester	2nd trimester	3rd trimester
lgM+	20	8	4
lgG+	18	36	90
lgG-lgM-	130	48	20

Fig. 1: Anti-toxoplasma IgM seropositivity across gestational age of pregnant women

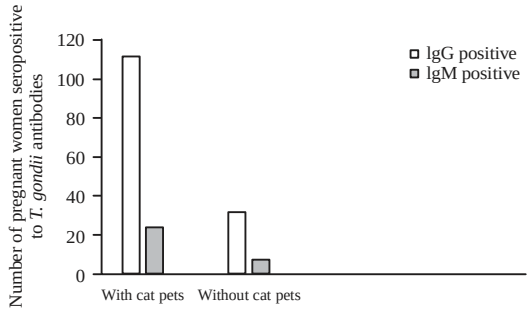


Fig. 2: Anti-toxoplasma IgM seropositivity across pregnant women and contact with domesticated cat pets

In this study, we reported an overall 48.9% seroprevalence of *T. gondii* infection in pregnant women from the northeastern city of Nigeria. Out of the 360 subjects, 6.7% had Primary infection of *T. gondii* acquired during pregnancy. This could be passed to their fetus and might lead to serious consequences such as miscarriage, neurological and visual disorders (Tenter *et al*., 2000). This seroprevalence is in agreement with a previous study by Singh and Pandit (2004), who reported a total seroprevalence of 48.3% in India. Our finding is relatively higher than previous Nigerian studies in Lagos and Zaria who recorded 40.4 and 29.9%, respectively (Ishaku *et al*., 2009; Deji-Agboola *et al*., 2011) and other European studies with seroprevalence of 10% reported in the United Kingdom (Nash *et al*., 2005) and Norway (Jenum *et al*., 1998). Conversely, higher seroprevalence of 63.5% was reported in Colombia (Castro *et al*., 2009) and 77.5% in Brazil (Porto *et al*., 2008). Higher prevalence rates were also reported in some neighboring African countries of Sudan (Elnahas *et al*., 2003) and Morocco (El Mansouri *et al*., 2007). The difference in prevalence rate between different countries can be explained by variation of geographical and climatic conditions between different areas where higher seroprevalence is associated with hotter and wetter areas, which are conditions favourable for sporulation of oocysts compared to less humid areas (Nijem and Al Amleh, 2009; Kistiah *et al*., 2011).

The high prevalence of *T. gondii* antibodies in Maiduguri town whose predominant residents Muslims are expected with the presence of relatively high population of cats in several households. This may be because Muslims find pleasure keeping cat pets in their homes. Our study showed significant relationship between *T. gondii* antibodies and presence of domestic cats, which is in consonance with studies from Taiwan (Lin *et al*., 2008), Ethiopia (Zemene *et al*., 2012) and France (Baril *et al*., 1999). They reported significant relationship between contacts with domesticated cats and prevalence of *T. gondii*. However, other studies showed no significant relationship (Pal *et al*., 2011; Mwambe *et al*., 2013; Ishaku *et al*., 2009).

The highest prevalence of *T. gondii* IgM and IgG antibodies was observed among women within the age group 21-25 years with 46 (76.7%) and 19 (5.3%), respectively, while IgG seronegativity was higher in older subjects (those ≥30 years). However, there was no significant difference in the seroprevalence of *T. gondii* infection observed between different age groups. This is in line with the data obtained from a study in Saudi Arabia (Al-Qurashi *et al*., 2001). This may be an indicator of high susceptibility of primary *T. gondii* infection among older subjects than younger individuals. This paradoxically shows that seroprevalence of IgG antibodies declined with age. The fewer IgG seropositivity observed from older subjects may be as a result of waned-off IgG subclass over several years of previous exposure (Casadevall, 2004).

The fact that all the unemployed women in our study were housewives might increase the IgG seroprevalence of toxoplasmosis since they are usually in direct contact with vegetables during food preparation. These vegetables are usually cultivated in gardens where cats and other infected animals' feces are often used as manure. *Toxoplasma gondii* cysts in food stuffs are not easily destroyed by washing. They become source of infection when these vegetables are consumed without appropriate washing. More so, they interact more with the pets, clean the house (including the feces of the pets) as they primarily are engaged in house chores. In theory, the unemployed are presumably immune against toxoplasmosis than the employed who exhibit relatively higher

IgG seronegativity, thus placing them to higher susceptibility to primary infection. However, in the absence of poor hand hygiene, these vegetables could be a main route for reinfection with different strains of *T. gondii* (Dabritz and Conrad, 2009).

This study showed a highly significant relationship between seroprevalence in pregnant women from urban compared to those at rural residential areas. This can be due to high income of urban settlers and their different eating habits especially consuming ready-made chickens and junky food prepared from restaurants with poor hygienic conditions, which may have been the major source for *T. gondii* transmission (Sroka *et al*., 2010). The absence of a significant relationship between the prevalence of antibodies to *T. gondii* among pregnant women in Maiduguri based on their level of education does not rule out that this factor has no effect on the transmission of *T. gondii* infection.

The use of IgG avidity testing was of great diagnostic importance in differentiating the period of infection whether it was more than 5 months or less. In patients where IgM test were not discriminatory, this is useful in order to classify primary toxoplasmosis and assess risk of congenital transmission. The high IgG avidity results could help in calculating the timing of the infection whether it was predated to conception or postdated if the patient can undergo this test within 5 months of gestational, although there is paucity of data with regards to the use avidity testing on IgM positive samples in laboratories of developing nations to rule out acute infection. Our study was first of its kind from Nigeria to show that low IgG avidity is obtainable in IgM positive women.

Previous data showed that risk of congenital transmission increases with gestational age, with the highest rates (60-81%) in the third trimester compared with 6% in the first trimester (Foulon *et al*., 1999; Dunn *et al*., 1999). On the other hand, disease severity, decreases with gestational age, with first trimester infection resulting in miscarriage (Chen *et al*., 2005). From our study, pregnant women at first trimester had the highest anti-toxoplasma IgM seronegativity and least IgG seropositivity, 20 (5.6%) and 18 (5.0%), while those at third trimester had the least IgM seropositivity and highest IgG seropositivity 4 (1.1%) and 90 (25%), respectively. However, there was no significant difference between anti-toxoplasma antibodies and gestational age. This finding is in line with a previous study by Aqeely *et al*. (2014).

It is worthy to note that the overall risk of congenital infection from primary toxoplasmosis during pregnancy has been shown to range from 20-50% in the absence of appropriate treatment (McLeod *et al*., 2009). *Toxoplasma gondii* infected neonates have been shown to be at substantial risk of developing long-term sequelae when no treatment is given to them (Brown *et al*., 2009).

The results of this study showed a significantly high seroprevalence. This justifies the need to include laboratory testing for *T. gondii* infection during antenatal investigations and to educate women about risk factors that could predispose them to contracting the parasite in order to prevent maternal and subsequent congenital infections. Considering the evidence of information from our study, we strongly recommend that healthcare facilities should follow-up women with IgM seropositive results, appropriately treat them and monitor their immune responses over time, more so, the neonates should be thoroughly examined after delivery in order to detect any sequelae maternal toxoplasmosis that might have been provoked *in-utero*.

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REFERENCES

Al-Qurashi, A.R., A.M. Ghandour, O.E. Obeid, A. Al-Mulhim and S.M. Makki, 2001. Seroepidemiological study of *Toxoplasma gondii* infection in the human population in the Eastern Region. Saudi Med. J., 22: 13-18.

Aqeely, H., E.K. El-Gayar, D.P. Khan, A. Najmi and A. Alvi *et al*., 2014. Seroepidemiology of *Toxoplasma gondii* amongst pregnant women in Jazan province, Saudi Arabia. J. Trop. Med. 10.1155/2014/913950

Baril, L., T. Ancelle, V. Goulet, P. Thulliez, V. Tirard-Fleury and B. Carme, 1999. Risk factors for Toxoplasma infection in pregnancy: A case-control study in France. Scand. J. Infect. Dis., 31: 305-309.

Borkakoty, B.J., A.K. Borthakur and M. Gohain, 2007. Prevalence of *Toxoplasma gondii* infection amongst pregnant women in Assam India. Indian J. Med. Microbiol., 25: 431-432.

Brown, E.D., J.K. Chau, S. Atashband, B.D. Westerberg and F.K. Kozak, 2009. A systematic review of neonatal toxoplasmosis exposure and sensorineural hearing loss. Int. J. Pediatr. Otorhinolaryngol., 73: 707-711.

Casadevall, A., 2004. The methodology for determining the efficacy of antibody-mediated immunity. J. Immunol. Methods, 291: 1-10.

Castro, A.T., A. Gongora and M.E. Gonzalez, 2009. *Toxoplasma gondii* antibody seroprevalence in pregnant women from Villavicencio, Colombia. Orinoquia, 12: 91-100.

Chen, K.T., A. Eskild, M. Bresnahan, B. Stray-Pedersen, A. Sher and P.A. Jenum, 2005. Previous maternal infection with *Toxoplasma gondii* and the risk of fetal death. Am. J. Obstet. Gynecol., 193: 443-449.

Cook, A.J., R.E. Gilbert, W. Buffolano, J. Zufferey and E. Petersen *et al*., 2000. Sources of toxoplasma infection in pregnant women: European multicentre case-control study. Br. Med. J., 321: 142-147.

Dabritz, H.A. and P.A. Conrad, 2009. Cats and *Toxoplasma*: Implications for public health. Zoonoses Pub. Health, 57: 34-52.

Deji-Agboola, A.M., O.S. Busari, O.A. Osinupebi and A.O.J. Amoo, 2011. Seroprevalence of *Toxoplasma gondii* antibodies among pregnant women attending antenatal clinic of federal medical center, Lagos, Nigeria. Int. J. Biol. Med. Res., 2: 1135-1139.

Di Carlo, P., A. Romano, M.G. Schimmenti, A. Mazzola and L. Titone, 2008. Materno-fetal *Toxoplasma gondii* infection: Critical review of available diagnostic methods. Infez Med., 16: 28-32.

Dubey, J.P. and J.L. Jones, 2008. *Toxoplasma gondii* infection in humans and animals in the United States. Int. J. Parasitol., 38: 1257-1278.

Dubey, J.P., D.S. Lindsay and M.R. Lappin, 2009. Toxoplasmosis and other intestinal coccidial infections in cats and dogs. Vet. Clin. North. Am. Small Anim. Pract., 39: 1009-1034.

Dunn, D., M. Wallon, F. Peyron, E. Petersen, C. Peckham and R. Gilbert, 1999. Mother-to-child transmission of toxoplasmosis: Risk estimates for clinical counseling. Lancet, 353: 1829-1833.

El Mansouri, B.R.M., F. Sebti, F. Amarir, M. Laboudi, R. Bchitou, M. Hamad and M. Lyagoubi, 2007. Seroprevalence of toxoplasmosis in pregnant women in Rabat, Morocco. Bull. Soc. Pathol. Exot., 100: 289-290.

Elmore, S.A., J.L. Jones, P.A. Conrad, S. Patton, D.S. Lindsay and J.P. Dubey, 2010. *Toxoplasma gondii*: Epidemiology, feline clinical aspects and prevention. Trends Parasitol., 26: 190-196.

Elnahas, A., A.S. Gerais, M.I. Elbashir, E.S. Eldien and I. Adam, 2003. Toxoplasmosis in pregnant Sudanese women. Saudi Med. J., 24: 868-870.

Foulon, W., J.M. Pinon, B. Stray-Pedersen, A. Pollak and M. Lappalainen *et al*., 1999. Prenatal diagnosis of congenital toxoplasmosis: A multicenter evaluation of different diagnostic parameters. Am. J. Obstet. Gynecol., 181: 843-847.

Han, K., D.W. Shin, T.Y. Lee and Y.H. Lee, 2008. Seroprevalence of *Toxoplasma gondii* infection and risk factors associated with seropositivity of pregnant women in Korea. J. Parasitol., 94: 963-965.

Ishaku, B., I. Ajogi, J. Umoh, I. Lawal and A.J. Randawa, 2009. Seroprevalence and risk factors for *Toxoplasma gondii* infection among antenatal women in Zaria, Nigeria. Res. J. Med. Med. Sci., 4: 483-488.

Jenum, N.F., B. Stray-Pedersen, K.K. Melby, G. Kapperud, A. Whitelaw, A. Eskild and J. Eng, 1998. Incidence of *Toxoplasma gondii* infection in 35,940 pregnant women in Norway and pregnancy outcome for infected women. J. Clin. Microb., 36: 2900-2906.

Jones, J.L., D. Kruszon-Moran, M. Wilson, G. McQuillan, T. Navin and J.B. McAuley, 2001. *Toxoplasma gondii* infection in the United States: seroprevalence and risk factors. Am. J. Epidemiol., 154: 357-365.

Kistiah, K., A. Barragan, J. Winiecka-Krusnell, A. Karstaedt and J. Frean, 2011. Seroprevalence of *Toxoplasma gondii* infection in HIV-positive and HIV-negative subjects in Gauteng, South Africa. South Afr. J. Epidemiol. Infect., 26: 225-228.

Kravetz, J.D. and D.G. Federman, 2005. Prevention of toxoplasmosis in pregnancy: Knowledge of risk factors. Gynecol. Infect. Dis. Obstet., 13: 161-165.

Liesenfeld, O., C. Press, J.G. Montoya, R. Gill, J.L. Isaac-Renton, K. Hedman and J.S. Remington, 1997. False-positive results in immunoglobulin M (IgM) toxoplasma antibody tests and importance of confirmatory testing: The Platelia Toxo IgM test. J. Clin. Microbiol., 35: 174-178.

Lin, Y.L., Y.S. Liao, L.R. Liao, F.N. Chen, H.M. Kuo and S. He, 2008. Seroprevalence and sources of *Toxoplasma* infection among indigenous and immigrant pregnant women in Taiwan. Parasitol. Res., 103: 67-74.

Masini, L., L. Casarella, R.L. Grillo, M.P. Zannella and G.C. Oliva, 2008. Epidemiologic study on anti-*Toxoplasma gondii* antibodies prevalence in an obstetric population. Italian J. Gynaecol. Obstet., 20: 159-166.

McLeod, R., F. Kieffer, M. Sautter, T. Hosten and H. Pelloux, 2009. Why prevent, diagnose and treat congenital toxoplasmosis? Mem. Inst. Oswaldo Cruz., 104: 320-344.

Montoya, J.G., 2002. Laboratory diagnosis of *Toxoplasma gondii* infection and toxoplasmosis. J. Infect. Dis., 185: S73-S82.

Mwambe, B., S.E. Mshana, B.R. Kidenya, A.N. Massinde and H.D. Mazigo *et al*., 2013. Sero-prevalence and factors associated with *Toxoplasma gondii* infection among pregnant women attending antenatal care in Mwanza, Tanzania. Parasites Vectors, Vol. 6 10.1186/1756-3305-6-222

Nash, J.Q., S. Chissel, J. Jones, F. Warburton and Q.N. Verlander, 2005. Risk factors for toxoplasmosis in pregnant women in Kent, United Kingdom. Epidemiol. Infect., 133: 475-483.

Nijem, K.I. and S. Al Amleh, 2009. Seroprevalence and associated risk factors of toxoplasmosis in pregnant women in Hebron district, Palestine. East Mediterr. Health J., 15: 1278-1284.

Pal, S., N. Das and D. Pal, 2011. Sero-prevalence and risk factors of *Toxoplasma gondi* in pregnant women in kolkata, India. J. Renin-Angiotensin-Aldosterone Syst., 26: 27-33.

Paquet, C., Q.C. Trois-Rivieres and M.H. Yudin, 2013. Toxoplasmosis in pregnancy: Prevention, screening and treatment. J. Obstet. Gynaecol. Can., 35: 78-81.

Porto, A.M.F., M.M.R. de Amorim, I.C.N. Coelho and L.C. Santos, 2008. Serologic profile of toxoplasmosis in pregnant women attended at a teaching-hospital in Recife. Revista Assoc. Med. Bras., 54: 242-248.

Singh, S. and A.J. Pandit, 2004. Incidence and prevalence of toxoplasmosis in Indian pregnant women: A prospective study. Am. J. Reprod. Immunol., 52: 276-283.

Skariah, S., M.K. McIntyre and D.G. Mordue, 2010. *Toxoplasma gondii*: Determinants of tachyzoite to bradyzoite conversion. Parasitol. Res., 107: 253-260.

Sroka, J., A.W. Ojcik-Fatla, J. Szymanska, J. Dutkiewicz, V. Zajac and J. Zwolinski, 2010. The occurrence of *Toxoplasma gondii* infection in people and animals from rural environment of Lublin region-estimate of potential role of water as a source of infection. Ann. Agric. Environ. Med., 17: 125-132.

Tenter, A.M., A.R. Heckeroth and L.M. Weiss, 2000. *Toxoplasma gondii*: From animals to humans. Int. J. Parasitol., 30: 1217-1258.

Zemene, E., D. Yewhalaw, S. Abera, T. Belay, A. Samuel and A. Zeynudin, 2012. Seroprevalence of *Toxoplasma gondii* and associated risk factors among pregnant women in Jimma town, Southwestern Ethiopia. BMC Infect. Dis., Vol. 12. 10.1186/1471-2334-12-337