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Seroprevalence of *Toxoplasma gondii* infection and associated risk factors among pregnant women attending antenatal clinic at the Bamenda Regional Hospital, Cameroon

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Abstract:

Background: *Toxoplasma gondii* is a ubiquitous, coccidian parasite that causes toxoplasmosis. This infection, if acquired during pregnancy may result in severe damage. It affects a third of the world's population. In many developing countries, its prevalence is unknown, and data concerning its seroprevalence among pregnant women is scarce in our study area. The objective of this study is to determine the seroprevalence of *T. gondii* infection and the associated risk factors among pregnant women attending the antenatal clinic (ANC) at the Bamenda Regional Hospital in Cameroon. The results obtained will be useful in giving an estimate of the prevalence among pregnant women thus informing policy on preventive measures.
Methodology: This is a descriptive cross-sectional study of pregnant women recruited between January and April 2018 using systematic random sampling technique. Socio-demographic data of participants and predisposing factors to toxoplasmosis were collected using a pretested structured questionnaire administered to them. Five milliliters of blood were collected and the serum screened for IgG and IgM antibodies against *T. gondii* using the cassette and buffer immunochromatographic method. The positive IgG cases were tested further by ELISA technique. Data were analyzed using SPSS version 20. Associations between variables were tested by Chi square and p value < 0.05 was considered statistically significant.
Results: Of 127 women tested, 44 were seropositive for IgG *T. gondii* infection (34.6%) by cassette and buffer method and only 1 with both IgG and IgM antibodies (0.8%) were found among them with Elisa test. Pet ownership and handling of their litters were risk factors significantly associated with toxoplasmosis (p=0.013 and 0.006 respectively). Although the frequencies of consumption of raw dried meat and farming among the subjects were high, their associations with toxoplasmosis were not statistically significant.
Conclusion: The overall seroprevalence of *T. gondii* antibodies among the pregnant women is still high compared with previous finding in the same area (34.6 % for IgG and 0.8% for IgG and IgM). Pet ownership and handling of their litters were risk factors significantly associated with toxoplasmosis in this study. Screening of pregnant women during ANC and treatment of positive cases, are necessary to prevent congenital infections in the newborn. Health education on how to minimize exposure to the risk factors should be given.

Keywords: Risk factors, toxoplasmosis, congenital transmission, serological diagnosis.

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Séroprévalence de l'infection à *Toxoplasma gondii* et des facteurs de risque associés chez les femmes enceintes en visites prénatales à l'Hôpital Régional de Bamenda au Cameroun

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Résumé:

Contexte: *Toxoplasma gondii* est un parasite coccidien omniprésent qui cause la toxoplasmose. Cette infection, si elle est contractée pendant la grossesse, peut entraîner de graves dommages. Elle affecte un tiers de la population mondiale. Dans de nombreux pays en développement, sa prévalence est inconnue, et les données concernant sa séroprévalence chez les femmes enceintes sont rares dans notre zone d'étude. L'objectif de cette étude est de déterminer la séroprévalence de l'infection à *T. gondii* et les facteurs de risque associés chez les femmes enceintes fréquentant la clinique prénatale de l'hôpital régional de Bamenda au Cameroun. Ceci dans l'optique de donner des estimations sur sa prévalence chez ces femmes enceintes et de générer des stratégies à prendre pour des mesures préventives.
Méthodologie: Il s'agissait d'une étude descriptive transversale effectuée de janvier à avril 2018 à l'aide d'une technique d'échantillonnage systématique. Les données sociodémographiques et certains comportements qui prédisposaient les participants à l'infection ont été recueillis à l'aide d'un questionnaire préétabli. Cinq millilitres d'échantillon sanguin ont été prélevés et le sérum a été dépisté pour détecter les anticorps IgG et IgM contre *T. gondi* en utilisant la méthode cassette et tampon. Les cas positifs d'IgG ont été soumis à la technique ELISA. Les données ont été analysées à l'aide du logiciel SPSS version 20. Les résultats étaient jugés statistiquement significatifs lorsque la valeur p était inférieure à 0,05
Résultats: Des 127 femmes testées, 44 avaient des anticorps IgG pour l'infection à *T. gondii* à 34,6 % et seulement 1 avait des anticorps IgG et IgM à 0,8. Les facteurs de risque associés à l'infection étaient la possession et la manipulation des fèces des animaux de compagnie (p=0,013 et 0,006 respectivement). Les proportions concernant la consommation de viande crue séchée et l'agriculture été élevées, mais les associations n'étaient pas statistiquement significatives.
Conclusion: La séroprévalence globale des anticorps *T. gondii* chez les femmes enceintes reste élevées dans la région (soit 34,6 % pour l'IgG et 0,8% pour IgG et IgM). La possession et la manipulation des animaux de compagnie étaient les facteurs de risque importants associés à l'infection. Le dépistage précoce et systématique de l'infection devrait être envisagé pendant la consultation prénatale, et le traitement pour tout cas positif. Une éducation sanitaire sur les façons de réduire au minimum l'exposition aux facteurs de risque devrait être fournie.

Mots clés: Facteurs de risques, toxoplasmose, transmission congénitale, diagnostic sérologique.

Introduction:

Toxoplasma gondii, the causative agent of toxoplasmosis*,* infects warm-blooded animals including humans. Seroprevalence studies show that the organism is found in almost every country with different climates and social conditions (1). Globally, about one third of the world's human population is estimated to carry *T. gondii* parasite (2). Bradyzoites, tachyzoites and the sporozoites in oocysts are the three infectious stages of the coccidium, parasite to all hosts member of the cat family as its definitive host and has a wild range of intermediate hosts including humans (3). Humans usually acquire *T. gondii* infection through consumption of raw or undercooked meat, of improperly washed vegetables and fruits as well as drinking of water which contains oocysts. Moreover, transplacentally tachyzoites infect the fetus in pregnant women (4,5).

Toxoplasmosis is normally asymptomatic in healthy individuals but can cause maternal-fetal transmission in women who acquire primary *Toxoplasma* infection during pregnancy (6,7). The risk of vertical transmission and associated problems are determined by the gestational age at which the primary infection is acquired. Transmission to the fetus increases from the first trimester (10 to 24%) to the third trimester (60 to 90%), but the potential of congenital defect is more severe with earlier infections (8,9). Most pregnant women with acute acquired infection do not experience obvious symptoms or signs. A minority may experience malaise, low-grade fever, and lymphadenopathy (10). Severe clinical signs in the infected infant commonly observed in offspring of women whose infection was acquired early in gestation (6,11). A minority may experience malaise, low-grade fever, and lymphadenopathy (5). In the immunocompetent subjects, *Toxoplasma* infection is often asymptomatic, and frequently results in the chronic persistence of cysts within host tissues, that probably lie dormant for life. In contrast, in immunocompromised subjects, it is always life threatening.

Diagnosis of toxoplasmosis in humans is performed using different techniques. Acute and latent *T. gondii* infections during pregnancy are mostly diagnosed by serological tests including detection of anti-*T. gondii* specific IgM and IgG antibodies (12). Latex agglutination (LA) testing, enzyme-linked immunosorbent assay (ELISA), and/or indirect fluorescent antibody testing (IFAT) are some of the tests that used to detect the antibodies (13). In our context, most studies on prevalence of *T. gondii* infections have relied on antibody detection, although this is associated with false positives. A more reliable way of

diagnosing active infection is the use of molecular techniques such as PCR using blood, brain biopsy, and liver biopsy materials (14). However, this is very expensive and not easily available in most developing countries making its use in routine diagnosis not feasible. It is for this reason that diagnosis of toxoplasmosis in most health facilities in these countries still largely rely on serology, hence the use of this method in the current study

There is uneven distribution of *T. gondii* prevalence among pregnancy and childbearing age from different parts of the world. In Europe, the prevalence of Toxoplasmosis varies between 20-50% in the South and between 50-70% in the West (15). Seroprevalence of 10.3% and 69% have been reported from Japan (16) and North-eastern Brazil (17). In Africa, data on *T. gondii* infection during pregnancy is scanty and its burden in pregnant women is an under-estimated public health concern. In these countries, higher (up to 92.5%) seroprevalence has been reported. However, the prevalence of infection varies widely between countries (from 10 to 80%) and often within a given country or between different communities in the same region (14). Pregnant women are not routinely investigated for *T. gondii* during pregnancy, and follow-up hardly exists (18,19). In Cameroon, Mbouo-Bandjoun in particular, the seroprevalence of *T. gondii* infection is 45.5% (20) which represents a real public health problem as there are limited researches that have been conducted about risk factors associated with such high rate.

At the Bamenda Regional Hospital (BRH) measurement of *T. gondii* immunoglobulins is part of the routine tests prescribed by consulting gynaecologists, however due to ignorance of the importance of this test and sometimes financial constraints, pregnant women often fail to have this test performed during the entire period of their pregnancies. Also, the diagnosis of *T. gondii* infection is solely based on a qualitative test, and only women with positive IgM are treated while those with positive are usually not tested further to confirm their diagnosis, and are therefore not treated. In addition, information is scarce on the seroprevalence of *T. gondii* infection in pregnant women particularly in the study area. It is for these reasons that the present study was undertaken, with the aim of determining the seroprevalence of *T. gondii* infection, and identifying associated risk factors among pregnant women attending the antenatal clinic (ANC) of the BRH, Cameroon.

Materials and method:

Study design and setting

This was a descriptive cross-sectional study involving pregnant women attending the antenatal clinic (ANC) at the Bamenda Regional Hospital (BRH) located in the capital city of North West Region of Cameroon. The ANC unit is part of the Obstetric and Gynaecologic department, one of the 21 departments that make up the second degree referral hospital. Its services include: reception/registration room, laboratory, two examination rooms and general conference hall where antenatal consultation and education of pregnant women take place. ANCs take place from Monday to Friday every week. This study was carried out for a period of four months (1st January to 31st April 2018).

Sample size and sampling technique

All participants, attendees of the ANC service, who met the inclusion criteria, were recruited over the study period, using a systematic random sampling method. After identification, the potential study participants were taken through the informed consent process whereby the study objectives, risks, benefits and study procedures were explained. A total of 127 pregnant women were enrolled among 243 who attended the ANC clinic during the study period.

Data collection

A pre-tested questionnaire was used to collect information by face-to-face interview on socio-demographic and economic status as well as epidemiological risk factors of *T. gondii* infection. Data was collected on the following variables: age, ethnic group, marital status, level of education, occupational status, age of pregnancy, area of residence, history of miscarriage and still births, owning of pets or domestic animals, types of pets, handling of pets litter, farm or gardening work, history of children with eye or head malformations, habit of consumption of undercooked meat, raw dried meat commonly known as 'Kilichi', raw vegetables, drinking water sources, and history of recent blood transfusion. The level of knowledge regarding toxoplasmosis and sources of information on *T. gondii* infection were also evaluated.

Sample collection and processing

Approximately 5 ml of venous blood was collected aseptically from each participant into a plain dry. The blood sample was allowed to clot and centrifuged at 3,000 rpm for 5 minutes, and the serum was separated into another plain dry. The sera were tested for anti-*T. gondii* IgM and IgG antibody using the cassette and buffer technique (an immunochromatographic test also named Rapid Diagnostic Test (RDT)). For the samples with a positive IgG but negative IgM, the quantitative testing was then done using

Enzyme Linked Immuno-Sorbant Assay (ELISA) test kit at the biochemistry lab, strictly following the manufacturer's instruction.

Rapid immunochromatographic test for toxoplasmosis

The sera were tested by the rapid ICT (cassette and buffer) test. During the period of collection, the test components were allowed to get to room temperature every morning. When the sera were ready to test, a test cassette was removed from the pouch, placed on a clean surface and labeled with the corresponding patient's code. One drop (about 30-45µL) of the specimen was placed in the sample well using a plastic dropper. One drop of specimen diluent was then added to the specimen and the timer was set. After 15mins the cassette was examined for result. The ***c***assette and buffer ICT test contains a build in control (C) line which develops after adding the specimen and sample diluent. If the 'C' line does not develop, the whole procedure is reviewed and the test repeated with a new cassette. The result was negative or non-reactive if only the 'C' line was present with the absence of any burgundy line in both the test lines (M and G) indicating that no anti-*T. gondii* Ig was detected in the specimen. The result was considered positive or reactive for; (i) anti-*T. gondii* IgM if only the M line develops in addition to the 'C' line; (ii) anti-*T. gondii* IgG if only the 'G' line develops in addition to the 'C' line; and (iii) anti-*T. gondii* IgG and IgM if both the G and M lines develop, in addition to the 'C' line.

ELISA test for toxoplasmosis

Serum samples positive for IgG but negative for IgM in the rapid ICT were further tested using the ELISA Toxo (DRG kit, Germany) test. For this second testing, all specimens were refrigerated at -10°C. On test days, both the specimens and test components were allowed to thaw to room temperature. The different steps of the ELISA technique were then performed. Each patient specimen was diluted with the sample diluents (1 in 100) into new tubes and allowed to stand for 15 mins before the start of the assay. The required number of microliter strips or wells were labeled and inserted into the holder, with the first six corresponding to the blank, negative control, standards 1-3, and the positive control.

Briefly, 100µL each of the negative control, standards and positive control was dispensed in the corresponding well, while making sure the blank is always empty and dry. 100µL of each specimen was then dispensed into the corresponding wells as labeled, making sure to use each disposable tip only once. The wells were covered with the foil paper provided in the kit and allowed to incubate for 1 hour at 37ºC. The content of the wells was then briskly shaken out and rinsed 5 times with the diluted wash solution (300µL per well). The wells were stroked against an absorbent paper to make sure all the residual droplets and air bubbles were removed, as the precision of the result is directly affected by the quality of the washing. 100µL of the enzyme conjugate was then dispensed into each well except the blank and incubated at room temperature, making sure not to expose to direct sunlight. The content of the wells was briskly shaken out, the washing repeated as above, and the wells stroked against an absorbent paper. 100µL of substrate solution was then added to all the wells and incubated for 15mins at room temperature in the dark. 100µL of the stop solution was then added to each well and any blue color that developed during the incubation turned yellow and highly positive samples at times got dark precipitates. The wells were then inserted into the ELISA reader which has been pre-programmed and a graph of absorbance value (mean) of the negative control, standards and their respective concentrations. This was then used to calibrate the absorbance of each of the test specimens and gave corresponding values.

The normal value range for ELISA was established by the BRH laboratory, based on its patient's population in the geographical area; negative when <45 IU/mL, cut-off value at 50 IU/mL, grey zone (equivocal) between 45-55 IU/mL and positive when >55 IU/mL. Patients with equivocal results were called 2 weeks later (according to diagnostic principles of *T. gondii*) and blood samples were recollected for confirmation of test results, either the negative (memory immunity) or positive serology (active infection).

Data management and analysis

All data from the questionnaire and laboratory record were analyzed using SPSS version 20.0 software package. Descriptive statistic was performed to describe demographic profile of the study participants. The univariate analysis involved frequency distributions for categorical variables, and descriptive statistics for continuous and discrete variables was done to give an understanding of the characteristics of the sample, as well as description of the response variables (*T. gondii* infection). Bivariate analysis was used to investigate association between the response variable (*T. gondii* infection) and sociodemographic and other variables of interest (risk factors). The χ^2 test was used to test association between 2 categorical variables and p value <0.05 was considered as statistically significant.

Ethical considerations

Ethical clearance (N°2017/0051H/UBa/IRB) was obtained from the Institutional Review Board at the Faculty of Health Science of the University of Bamenda. The administrative authorizations were obtained from the Delegation of Public Health of the North West Region and the General Supervisor of the Bamenda Regional Hospital. Moreover, written informed consent was obtained from all study participants prior to interview and blood collection. Confidentiality of the collected information and laboratory test results was maintained, and used solely for research purposes and neither for stigmatization nor to generate profit. The results of the study were made available for all participants as they desire with the attending physician for further management of the cases.

Results:

A total of 127 pregnant women age 14 to 50 years (27.4 ± 6.21 years) out of 243 who attended BRH ANC clinic were included and tested for anti-*T. gondii* IgG and IgM antibody. Of this, 43 (33.9%) were IgG seropositive and 1 (0.8%) was positive for both IgG and IgM. Table 1 shows the socio-demographic characteristics and obstetric history of the study participants with regard to seroprevalence of *T. gondii* infection. The age group 24 to 34 years had the highest seroprevalence (48.0%). Majority of the participants (91.3%) were Grassfield ethnic group and the least represented ethnic groups were the Beti and others (such as Sawa and Bakossi) with 1.6% each. No statistically significant association between *T. gondii* infection and the various ethnic groups was observed.

Among the 127 women who took part in this study, the grassfield was the ethnic group more represented and more infected with 90.9% of positive cases (40/44). The majority (59.0%) were married and represented 61% (27/44) of positive cases. There was no significant association between *T. gondii* infection and marital status of the participants. There was also no significant association between age of pregnancy, area of residence, history of miscarriage and stillbirth and *T. gondii* infection ($p>0.05$). With respect to gestational age, most of the participants were in their first trimester (50.4%) among which 54.5% (24/44) were positive for *T. gondii* infection. Regarding the area of residence, 79.5% were living in urban areas among which 75 % (33/44) were positive to *T. gondii* infection. Women with history of miscarriage represented 38.6% and 40.9% of positive cases, while those with a history of still birth (11.8%) represented 11.4% of positive cases.

Table 2 depicts the seroprevalence of study participants and factors associated with *T. gondii* infection. Among the total respondents, 32 (37.0%) own a pet and 21 (16.5%) handled pet litters. There was a significant association between *T. gondii* infection and owing a pet ($p=0.013$), and handling pet litters ($p=0.006$). There was no significant association between *T. gondii* infection and farming or gardening, and source of drinking water ($p>0.05$). However, among the 127 participants who practiced farming or gardening, 64 (61.36%) were positive for *T. gondii* infection. With respect to sources of drinking water, 96/127 had tap water, and among these, 68.2% were positive (30/44) for *T. gondii* infection. Ninety nine of 127 pregnant women responded that they consume raw fruits/vegetables, of which 81.8% were positive for *T. gondii* infection; 9 of 127 responded that they consume undercooked meat, of which 11.4% were positive to *T. gondii* infection; while 52.3% of those who consume raw dried meat were postive for *T. gondii* infection. There was no significant association between *T. gondii* infection and habit of consuming raw fruits/vegetables, undercooked meat, and raw dried meat. The pregnant women who had had blood transfusion in the year before their pregnancy represented 81.8% of positive cases but there was no significant association between *T. gondii* infection and history of blood transfusion.

Table 1: Socio-demographic and obstetric characteristics of study participants in relation to seroprevalence of *Toxoplasma gondii* infection

Variables	Seroprevalence		Total (%) n = 127	OR (95% CI)	*p value*
	Positive (%) n = 44	Negative (%) n = 83			
Age groups (years)					
14-24	14 (11,0)	30 (23,6)	44 (34.6)	0.766 (0.347-1.687)	0.507
24-34	21 (16.5)	40 (31.5)	61 (48.3)	1.026 (0.491-2.144)	0.945
34-44	6 (4.7)	11 (8.7)	17 (2.4)	0.890 (0.304-2.604)	0.831
44-50	3 (2.4)	2 (2.4)	5 (3,9)	2.963 (0.476-18.441)	0.244
Ethnic group					
Mbororo	1 (0.8)	3 (2.4)	4 (3.1)	0.020 (0.063-6.145)	0.683
Grassfield	40 (31.5)	76 (59.8)	116 (91.3)	0.921 (0.254-3.335)	0.900
Beti	1 (0.8)	1 (0.8)	2 (1.6)	1.907 (0.116-31.244)	0.651
Bayangi	1 (0.8)	2 (1.6)	3 (2.4)	0.942 (0.083-10.686)	0.961
Others	1 (0.8)	1 (0.8)	2 (1.6)	1.907 (0.116-31.244)	0.651
Marital status					
Single	13 (10.2)	25 (19.7)	38 (29.9)	0.973 (0.437-2.164)	0.946
Married	27 (21.2)	48 (37.8)	75 (59.0)	1.158 (0.549-2.44)	0.700
Divorced	1 (0.8)	0	1 (0.8)	5.894 (0.235-147.799	0.280
Cohabiting	3 (2.4)	10 (7.9)	13 (10.2)	0.562 (0.146-2.161)	0.401
Level of education					
FSLC	12 (9.4)	18 (14.2)	30 (23.6)	1.354 (0.582-3.150)	0.481
Ordinary	11 (8.7)	17 (13.4)	28 (22.0)	1.294 (0.544-3.076)	0.556
Advance	9 (7.1)	19 (15.0)	28 (22.0)	1.031 (0.423-2.513)	0.946
Tertiary	12 (9.4)	29 (22.8)	41 (32.3)	0.721 (0.32-1.612)	0.425
Employment status					
Unemployed	11 (8.6)	17 (13.4)	28 (22.0)	1.294 (0.544-3.076)	0.559
Public sector	6 (4.7)	16 (12.6)	22 (17.3)	0.671 (0.242-1.859)	0.443
Private sector	9 (7.1)	15 (11.8)	24 (18.9)	1.166 (0.464-2.929)	0.744
Self employed	11 (8.7)	23 (18.1)	34 (26.8)	0.869 (0.377-2.004)	0.743
Student	7 (5.5)	12 (8.8)	29 (14.3)	1.119 (0.406-3.0827)	0.827
Trimester of pregnancy					
1st trimester	24 (18.9)	40 (31.5)	64 (50.39)	1.290 (0.619-2.685)	0.496
2nd trimester	18 (14.2)	38 (29.9)	56 (44.1)	0.819 (0.391-1.718)	0.599
3rd trimester	2 (1.6)	5 (3.9)	7 (5.5)	0.761 (0.141-4.095)	0.750
Residence					
Urban	33 (26.0)	68 (53.5)	101 (79.5)	0.662 (0.274-1.599)	0.359
Rural	10 (7.9)	15 (11.8)	25 (19.7)	1.333 (0.542-3.279)	0.531
Unknown	1 (0.8)	0	1 (0.8)	0.215 (0.026-1.749)	0.150
History of miscarriage					
Yes	18 (14.2)	31 (24.4)	49 (38.6)	1.161 (0.549-2.452)	0.695
No	22 (17.3)	52 (41.0)	74 (58.4)		
History of still birth					
Yes	5 (3.9)	10 (7.9)	15 (11.8)	0.936 (0.299-2.931)	0.909
No	39 (30.7)	73 (57.5)	112 (88.2)		

OR: Odd ratio, CI: Confidence interval, n: Frequency, %: Percentages, FSLC: First School Living Certificate

Table 2: Seroprevalence rate and risk factors for *Toxoplasma gondii* infection among study participants

Variables	Seroprevalence		Total (%) (n = 127)	OR (95% CI)	*p* value
	Positive (%) (n = 44)	Negative (%) (n = 83)			
Owning pet					
Yes	17 (13.4)	15 (11.8)	32 (37.0)	2.854 (1.25-6.51)	0.013
No	27 (21.3)	68 (53.5)	85 (74.8)		
Contact with cat					
Yes	8 (6.3)	6 (4.4)	14 (10.7)	2.851 (0.921-8.829)	0.069
No	36 (28.1)	77 (60.6)	113 (88.8)		
Handling pet litter					
Yes	13 (10.2)	8 (6.3)	21 (16.5)	3.931 (1.48-10.42)	0.006
No	31 (24.4)	75 (59.1)	106 (83.5)		
Farming/gardening					
Yes	27 (21.3)	37 (29.1)	64 (50.4)	1.975 (0.937-4.161)	0.075
No	17 (12.4)	46 (36.2)	63 (48.6)		
Source of drinking water					
Tap	30 (21.9)	66 (52.0)	96 (73.9)	0.552 (0.241-1.264)	0.159
Spring	7 (5.5)	7 (5.5)	14 (11.0)	2.054 (0.671-6.289)	0.207
Well	5 (3.9)	6 (4.7)	11 (8.7)	1.645 (0.472-5.730)	0.434
Bottle	2 (1.6)	3 (2.4)	5 (3.9)	1.413 (0.227-8.776)	0.712
Tap and stream	0	1 (0.79)	1 (0.79)	0.618 (0.025-15.488)	0.769
Habit of eating raw fruits/vegetables					
Yes	36 (28.8)	63 (49.6)	119 (78.0)	1.429(0.571-3.572)	0.446
No	8 (5.9)	20 (15.7)	28 (21.9)		
Habit of eating undercooked meat					
Yes	5 (3.94)	4 (3.15)	9 (7.09)	2.532(0.644-9.961)	0.184
No	39 (30.7)	79 (62.2)	118 (90.7)		
Habit of eating raw dried meat					
Yes	23 (18.1)	30 (23.6)	53 (41.7)	1.935(0.921-4.063)	0.081
No	21 (16.5)	53 (41.7)	74 (58.3)		
History of blood transfusion					
Yes	6 (4.7)	8 (6.3)	14 (11.1)	1.480(0.479-4.574	0.495
No	38 (29.9)	75 (59.1)	113 (89.0)		

OR: Odd ratio, CI: Confidence interval, n: Frequency, %: Percentages

Discussion:

The present study reports the seroprevalence of *T. gondii* infection among pregnant women in the BRH to be 35.4% (34.6% for IgG and 0.8% for both IgG and IgM. This rate is similar to the those reported in Nigeria by Umar et *al.,* (21) and Bello *et al.,* (2), but lower than rates previously reported by Guemgne *et al*., (20), Nguefack *et al.,* (21) and Njunda *et al.,* (22) in Cameroon and Yohanes *et al*., (23) in Ethiopia. In the study by Njunda *et al.,* (22), the rate was 70% in pregnant women, however, the study looked at a metropolitan population in a third degree referral health facility (Douala General Hospital) whereas in the present study, it was conducted in a second degree referral unit (BRH). Also Douala is a town with a more precarious living condition which could also explain the higher prevalence reported in that study. In the study by Yohanes *et al*., (23) on 232 pregnant women, the overall seroprevalence of *T. gondii* infection was 79.3% with 175 (75.43%) positive for IgG, 9 (3.9%) for IgM, 2 of which were positive for both IgG and IgM. Another study carried out by Wams *et al.,* (24) in Njinikom-Cameroun reported a prevalence of 54.5%, which is also higher than the rate in our study.

The observed differences in the seroprevalence rate of anti-*T. gondii* infection may be due to differences in the two study populations and the sample size. In fact, the present study was confined to an urban population in Bamenda, as opposed to the rural population in the study by Wams *et al.,* (24). As such there is perceived higher level of awareness, and preventive measures about *T. gondii* in our study population. The variation in seroprevalence of *T. gondii* infection may be due to differences in geographical distribution of the parasite, socioeconomic, personal hygienic practices, and feeding habit of the study participants. In addition, differences in test methods may also account for the variation.

The presence of IgM antibodies during pregnancy indicates acute *T. gondii* infection with high risk of maternal-fetal transmission (25). A previous study has estimated that in the absence of treatment, the risk of congenital infection from acute *T. gondii* infection during pregnancy is about 50% (26). Early diagnosis of infections in pregnant mothers is of great importance so that measures that can reduce the risk of transmission and possible sequelae in the newborn are promptly initiated. Therefore, screening of pregnant women for *Toxoplasma* infection should be considered as a part of the routine investtigation during ANC follow up.

Our study did not show any statistically significant association between the trimester of pregnancy and *Toxoplasma gondii* infection, which agrees with studies from Egypt by Mandour *et al.,* (27) in 2017 and Yemen by Saif *et al.,* (28) in 2014 but disagrees with those of Khan *et al.,* (29), Alanyande *et al*., (30) and Bello *et al*., (2) who reported significant association. A significant association between pet ownership and *T. gondii* infection was found. Similarly, Khan *et al.,* (29) in 2011 and Yohanes *et al*., (23) in 2017 showed coherent associations. Contrarily, studies by Pal *et al*., (31) and Saif *et al*., (28) showed different results. There was no significant association between cat ownership and *T. gondii* infection, similar to what has been reported by Ebrahimzadeh *et al.,* (32), Shao *et al.,* (15), Murebwayire *et al*., (14), Jumaian (33), Mandour *et al*., (18), Makiani *et al*., (34), Njunda *et al*., (22), Wam *et al*., (24)] and Saif *et al*., (28). However, many studies among which Moura *et al*., (1), Dwinata *et al*., (3), Umar *et al.,* (25), Yohanes *et al*., (23), Agmas *et al*., (36), Nissapatorn *et al*., (37), Duan *et al*., (38) and Nguefack *et al.,* (21) demonstrated the contrary. This may be explained by the fact that the presence of cat in the house is not enough to cause zoonosis but rather handling of cats' litter is of more importance. Our study demonstrated a significant association between handling of pets' litter and *T. gondii* infection, a finding that agrees with that of Dwinata *et al*., (3) in 2016.

There was no significant association between farming or gardening and *T. gondii* infection, which is similar to studies by Moura *et al*., (1), Agmas *et al*., (36), Makiani *et al*., (34), Nissapatorn *et al*., (37), and Yohanes *et al*., (23) but contradicts the studies by Jumaian *et al.,* (33) in 2005 and Mandour *et al.,* (18) in 2017. The findings of these studies appeared different because contamination of soil occurs principally after defecation of cats, and contamination rate largely depend on cat density, with urban areas where cat density is low tending to have low soil contamination rate, and therefore low prevalence of *T. gondii* infection.

Conclusion:

This study reports seroprevalence of *T. gondii* infection in pregnancy to be 34.6%. The risk factors associated with *T. gondii* infection include pets' ownership and handling of their litters. There is need for routine screening of pregnant women for *T. gondii* infection during ANC and treatment of cases. Education on hygiene and awareness of risk exposures regarding *T. gondii* infection to minimize its effects among pregnant women and the general population are imperative.

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Authors' contributions:

NNF and FP were involved in the design of the study, drafted the protocol with input from other authors. NNF, DWP and KFHL monitored laboratory work and analyzed the data. GBM performed the laboratory analysis and collected the results. NNF and KFHL drafted and finalized the manuscript for publication. DWP and TWA edited the manuscript. All authors contributed to the writing of the paper and approved the final version.

Conflict of interest:

Authors declared no conflict of interest

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