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Original Research Article

## Seroprevalence and factors associated with toxoplasmosis among pregnant women attending antenatal care clinic at a tertiary teaching hospital in Uganda: a cross-sectional study

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### ABSTRACT

**Background:** Toxoplasmosis is an infection caused by a parasite called *Toxoplasma gondii* and is progressively increasing in pregnancy mainly with age and if left undiagnosed and untreated, can result into several adverse pregnancy outcomes. In Uganda, there is a paucity of information about toxoplasmosis in pregnancy. This study aimed to determine the seroprevalence and factors associated with toxoplasmosis among pregnant women attending antenatal care clinic at a Tertiary hospital in Uganda.

**Methods:** This was a cross-sectional study carried out from July 2022 to October 2022. 165 pregnant women were consecutively enrolled. Data gathered for analysis were collected using a pre-tested structured questionnaire. Blood samples were taken and test of *Toxoplasma gondii* specific IgG and IGM was done using EUROIMMUN ELISA test kits as instructed by manufacturer. Data was analyzed using STATA version 14.2. A bivariate and multivariate analysis were used to show the association between the dependent and independent variables considering  $p \leq 0.05$  at 95% confidence interval.

**Results:** The overall seroprevalence of toxoplasmosis in pregnancy was 16.4%. Age >35 years old (aOR: 8.36; 95% CI: 1.809-38.71;  $p=0.007$ ), having contact with cats (aOR: 3.55; 95% CI: 1.258-10.01;  $p=0.017$ ), drinking untreated water (aOR=4.08; 95% CI: 1.237-13.46;  $p=0.021$ ) and HIV status (aOR=8.91; 95% CI: 1.419-56.03;  $p=0.020$ ) were independently associated with toxoplasmosis infection.

**Conclusions:** The overall seroprevalence of toxoplasmosis in pregnancy was low as compared to the global seroprevalence. There is a need to educate pregnant women about the transmission routes and preventive measures of toxoplasmosis at antenatal care.

**Keywords:** Seroprevalence, Toxoplasmosis, Pregnant women, Antenatal clinic

### INTRODUCTION

Globally, it has been estimated that about 30% of people are infected with *Toxoplasma gondii* in general population.<sup>1</sup> However, in pregnancy, the global seroprevalence was seen to be 34.8%.<sup>2</sup> Seroprevalence of

toxoplasmosis has been shown to be higher in pregnancy women than in general population probably due to reduced cellular immunity during pregnancy.<sup>3</sup> This varies widely between countries and within the same country or even among different communities of the same region.<sup>4</sup> Over the past three to four decades, the seroprevalence of

toxoplasmosis has decreased in some areas of Eastern and Central Europe.<sup>5</sup>

Several epidemiological studies on toxoplasma infection in pregnancy have been done in different countries. In Africa, recent study demonstrated that the seroprevalence of toxoplasmosis in pregnancy ranges from 5.87% in Zambia to 88.60% in Ethiopia with an average of 51.01%.<sup>6</sup> Previous study has shown seroprevalence of toxoplasmosis in pregnancy as high as 92.5% in Ghana.<sup>5</sup> In 2020, seroprevalence of toxoplasmosis in pregnancy from a study done in Northwest Ethiopia was 67.8%.<sup>7</sup> In Cameroon, seroprevalence of toxoplasma infection in pregnant women was 35.4%.<sup>8</sup> In neighboring countries, overall seroprevalence of toxoplasmosis in pregnancy have been found to be 80.3% in Democratic Republic of Congo, 30.9% and 44.5% in North-western and Northern Tanzania respectively, 32% in Kenya and 12.2% in Rwanda.<sup>9-13</sup>

Studies showed association between Sociodemographic factors and toxoplasmosis in pregnancy, these include; increased maternal age, low income, low level of education and rural residents.<sup>14,15</sup> In many studies, Toxoplasmosis in pregnancy have been associated with behavioral factors such as eating undercooked meat, contact with cats, drinking unpasteurized milk, eating low vegetables and poor sanitation.<sup>14,16</sup> Concerning medical and obstetrical factors, studies showed a significant association between toxoplasmosis in pregnancy with high parity, history of abortions, history of stillbirths, history of blood transfusion and in women with HIV infection.<sup>4,17-20</sup>

In pregnancy, chronic toxoplasmosis has been found to be associated with intrauterine growth retardation, post term pregnancies, slow development of postnatal motor skills, psychiatric disorders and Eclampsia, whereas acute toxoplasmosis in pregnancy, if not diagnosed and treated, can cause miscarriage and congenital *Toxoplasma gondii* infection that causes fetal hydrocephalus, intrauterine fetal death, blindness, retinochoroiditis, neurological damage and intracranial calcification.<sup>15,21,22</sup>

Unpublished data from maternity registry at KIU-TH, in a period of 3 months from January 2020 to April 2020, a total of 1069 pregnant women were admitted, among them 99 mothers (9.5%) had abortions, 33 mothers (3.1%) had stillbirths, 49 (4.6%) had low birth weight, for which we don't know the cause and yet toxoplasmosis may be among the causes. In areas where the seroprevalence of toxoplasmosis in pregnancy is high such as France and Austria, routine screening has resulted in diminished congenital toxoplasmosis.<sup>23</sup> Routine screening of toxoplasmosis in pregnant women who attend antenatal care is not being practiced in Uganda and at KIU-TH.

## METHODS

A cross sectional study was conducted in which 165 pregnant women were consecutively recruited from

antenatal care clinic of Kampala international university teaching hospital (KIUTH) during the months of July to October 2022. KIUTH is a private not for profit teaching hospital for Kampala international University located in Southern-western Uganda, Bushenyi district, Ishaka Municipality approximately 370 kilometers from the center of Kampala capital. KIU-TH is composed of different departments including obstetrics and gynecology department offering outpatients and inpatients. It has a 700-bed capacity for the inpatients. The obstetrics and gynecology department has 85 bed capacity. The hospital approximately conducts 1521 total deliveries per year including 695 cesarean sections per year. Antenatal care clinic of this hospital is run from Monday to Friday and receives 5 to 10 pregnant women per day. The hospital has a modern laboratory able to carry out toxoplasmosis tests. The study participants were recruited consecutively. All women with eligible criteria were counselled about the study, consented and pretested questionnaire written in both English and Runyankole (local language) was used to collect data. The sample size calculated using Kish Leslie formula (1965).

$$n = \frac{z^2 p(1-p)}{d^2}$$

Where the prevalence rate was 12.2% at 95% confidence interval from a study done in Rwanda.<sup>13</sup> With estimated error of 5% the sample size was 165 participants who were consecutively recruited until we reached the desired number. Pregnant women who were critically/mentally ill were not included in this study. Information on data collection sheet was compiled in Microsoft excel version 16 and imported into STATA software version 14.2. The overall seroprevalence of Toxoplasmosis in pregnancy was calculated by the total number of pregnant women with positive anti- *Toxoplasma gondii* antibodies divided by the total number of pregnant women tested. This was expressed in frequency and percentage. Results presented on pie-chart. To determine factors associated with toxoplasmosis in pregnancy, a bivariate and multivariate logistic regression were used. Unadjusted odds ratios with their corresponding 95% CI and p value were reported and all factors with p-value  $\leq 0.05$  were moved and analyzed at multivariate level to control confounding variables. The factors in the final multivariate analysis were reported together with their adjusted odds ratios, 95% confidence intervals and with their p values. A variable were considered statistically significant if it had a  $p \leq 0.05$ . The results for this objective was presented in table form.

### *Toxoplasma gondii* testing

Venous blood sampling (2 ml taken from the forearm using anticoagulant free vacutainer) was done for all mothers who were recruited from antenatal care clinic of KIUTH. Serum was obtained by centrifugation for 10 min and stored at -20 celsius degrees. Test of *Toxoplasma gondii* specific IgG and IGM was done using EUROIMMUN ELISA test kits as instructed by manufacturer. Results

were obtained quantitatively by getting the ratio of extinction value of calibrator positive and negative control sera with cut points of <0.8 for negative and ≥0.8 for positive results for both *Toxoplasma gondii* specific IgG and IgM ELISA. Seropositivity was reported if samples were positive for *T. gondii* specific IgM or IgG positive or both. Tests were conducted within 24 hours after blood sampling.

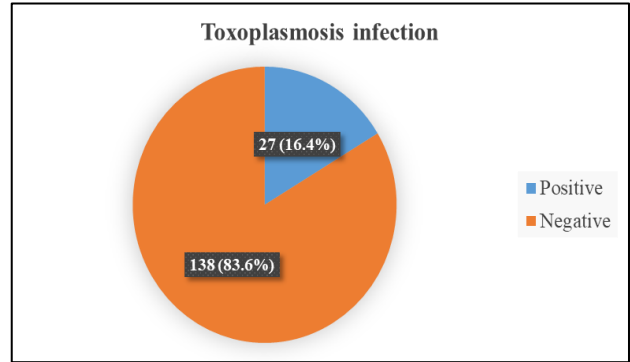
**RESULTS**

With a response rate of 100%, the study enrolled 165 pregnant mothers where majority of them ranged between 26 to 35 years of age 84 (51.5%), from rural area 113 (68.5%), had attended secondary school (43.6%), married 158 (95.8%) and multigravida 127(77.0%) (Table 1).

Of 165 pregnant women recruited in this study, 27 women had toxoplasmosis infection translating to the overall prevalence of 16.4%. Majority of participants didn't have toxoplasmosis infection 138 (83.6%) (Figure 1).

In this study, maternal age, contact with cats, drinking untreated water and HIV status remained independently associated with toxoplasmosis infection at multivariate level. Women who were >35 years old (aOR: 8.36; 95% CI: 1.809-38.71; p=0.007) and those who reported to have

had contact with cats (aOR: 3.55; 95% CI: 1.258-10.01; p=0.017) were respectively 8 and 3 times more likely to have toxoplasmosis infection. Women who drink untreated water (aOR=4.08; 95% CI: 1.237-13.46; p=0.021) and those who are seropositive for HIV (aOR=8.91; 95% CI: 1.419-56.03; p=0.020) were respectively 4 and 9 times more likely to have toxoplasmosis infection (Table 2).



**Figure 1: Seroprevalence of toxoplasmosis among pregnancy women attended antenatal care clinic at KIU-TH.**

**Table 1: Characteristics of study participants.**

Variables	Category	N	Percentage (%)
Age (In weeks)	18-25	63	38.2
	26-35	84	51.5
	>35	17	10.3
Educational level	No formal education	3	1.8
	Primary	68	41.2
	Secondary	72	43.6
	Tertiary	22	13.3
Residence	Rural	113	68.5
	Urban	52	31.5
Marital status	Married	158	95.8
	Single	7	4.2
Gravidity	Primigravida	38	23.0
	Multigravida	127	27.0

**Table 2: Factors associated with toxoplasmosis among pregnant women attended antenatal care clinic at KIU-TH.**

Variables	Toxoplasmosis infection		COR (95%CI)	P value	AOR (95%CI)	P value
	Negative, (n=138) (%)	Positive, (n=27) (%)				
<b>Age group (In years)</b>						
18-25	56 (40.6)	7 (25.9)	Ref		Ref	
26-35	72 (52.2)	13 (48.2)	1.44 (0.540-3.860)	0.463	1.35 (0.434-4.246)	0.599
>35	10 (7.2)	7 (25.9)	5.60 (1.612-19.45)	0.007*	8.36 (1.809-38.71)	0.007*
<b>Gravidity</b>						
Primigravida	33 (23.9)	5 (18.5)	Ref		Ref	
Multigravida	105 (76.1)	22 (81.5)	1.38 (0.485-3.939)	0.544		
<b>Education</b>						
Tertiary	21 (15.2)	1 (3.7)	Ref		Ref	

Continued.

Variables	Toxoplasmosis infection		COR (95% CI)	P value	AOR (95% CI)	P value
	Negative, (n=138) (%)	Positive, (n=27) (%)				
Secondary	59 (42.7)	13 (48.2)	4.60 (0.570-37.56)	0.152		
Primary	56 (40.6)	12(44.4)	4.50 (0.550-36.77)	0.161		
No formal education	2 (1.5)	1 (3.7)	10.5 (0.459-239.7)	0.141		
<b>Residence</b>						
Urban	48 (34.8)	4 (14.8)	Ref		Ref	
Rural	90 (65.2)	23 (85.2)	3.06 (1.002-9.380)	0.049*	2.18 (0.551-8.681)	0.266
<b>Marital status</b>						
Single	5 (3.6)	2 (7.4)	Ref		Ref	
Married	133 (96.4)	25 (92.6)	0.46 (0.086-2.558)	0.382		
<b>Contact with cats</b>						
No	107 (77.5)	10 (37.0)	Ref		Ref	
Yes	31 (22.5)	17 (63.0)	5.86 (2.440-14.11)	<0.001*	3.55 (1.258-10.01)	0.017*
<b>Eat undercooked meat</b>						
No	113 (81.9)	19 (70.4)	Ref		Ref	
Yes	25 (18.1)	8 (29.6)	1.90 (0.748-4.836)	0.176		
<b>Drink untreated water</b>						
No	97 (70.3)	8 (29.6)	Ref		Ref	
Yes	41 (29.7)	19 (70.4)	5.61 (2.277-13.86)	<0.001*	4.08 (1.237-13.46)	0.021*
<b>HIV status</b>						
Negative	134 (97.1)	22 (81.5)	Ref		Ref	
Positive	4 (2.9)	5 (18.5)	7.6 (1.896-30.56)	0.004*	8.91 (1.419-56.03)	0.020*
<b>Drink unpasteurized milk</b>						
No	112 (81.2)	21 (77.8)	Ref		Ref	
Yes	26 (18.8)	6 (22.2)	1.23 (0.451-3.354)	0.685		
<b>Aware of toxoplasmosis</b>						
No	122 (88.4)	26 (96.3)	Ref		Ref	
Yes	16 (11.6)	1 (3.7)	0.29 (0.037-2.310)	0.244		

\*P≤0.05; cOR: crude odds ratio; aOR; adjusted odds ratio.

## DISCUSSION

In this study, the overall seroprevalence of toxoplasmosis in pregnant women was 16.4%. Our study findings were in agreement with 16% and 17.3% obtained in the studies done in Northern America and London respectively.<sup>24,25</sup> This similarity could be due to the fact that those countries have almost the same average annual temperature with Uganda. Temperature have been shown to affect survival and infectivity of *Toxoplasma gondii* oocytes.<sup>26</sup> Our findings are in close proximity with those found in a study done in Rwanda where the overall seroprevalence of toxoplasmosis in pregnancy was 12.2%.<sup>13</sup> This could be due to the fact that this region is in close proximity to Rwanda and they share the same environmental conditions.

This overall seroprevalence of the current study was lower than the global seroprevalence of toxoplasmosis in pregnancy which was found to be 34.8%.<sup>26</sup> It was also lower than the overall seroprevalence in Africa 51.01% as well as in most African countries, 92.5% in Ghana, 70.8% in Ethiopia, 41.1% in Angola, and 33.8% in Nigeria.<sup>1,4,27,28</sup> This is one of the lowest seroprevalence found among our neighboring countries, 80.3% in Democratic Republic of Congo, 32% in Kenya and from 30.9% to 44.5% in Tanzania.<sup>9-12</sup> It has been shown that hot weather increases

the survival time and infectivity of *Toxoplasma gondii* parasite because it favors sporulation of oocytes.<sup>26</sup> Thus, a relatively cold weather in this region might explain the lower seroprevalence of toxoplasmosis in pregnancy compared to the seroprevalence reported from the above studies. This difference could be due to the fact that those studies used a bigger sample size compared to the current study.

However, the overall seroprevalence from this study is much higher than that was found in Zambia 5.87%.<sup>25</sup> This could have been attributed to the fact that the study in Zambia was done from pregnant women who attended national referral hospital in Lusaka capital city, those women might have been practicing good hygiene in contrast to our study that was done in rural setting.

In general, toxoplasmosis in pregnancy has been shown to vary widely between countries, within countries and among different community of the same country. The reason for this variation could be due to difference in climate conditions, geographical condition, socio-economic status, dietary and hygiene habits between different countries/communities.<sup>2,4</sup>

In this study, different factors were assessed for the toxoplasmosis among pregnant women. At multivariate analysis, four factors were shown to be significantly

associated with toxoplasmosis in pregnancy, those were: age, contact with cats, drinking untreated water and HIV status.

We found that pregnant women aged more than 35 years were significantly associated with toxoplasmosis ( $p=0.007$ ). This is in agreement with other studies done in Ethiopia, Brazil and Egypt.<sup>14,16,17</sup> This significant association between increased age and toxoplasmosis in pregnancy might be due to the fact that increase in maternal age increases the likelihood of long duration of exposure to *Toxoplasma gondii*. It has been shown that 1 year increase in maternal age is consistent with 7% increased risk of contracting *Toxoplasma gondii* infection.<sup>9</sup> In contrast, studies done in Cameroon and Tanzania showed no significant association between increased age and toxoplasmosis in pregnancy.<sup>8,11</sup> This could be attributed to different socio-demographic and behavioral characteristics. In fact, in the study done in Cameroon, the percentage of pregnant women above 35 years old was low.

Contact with cats was significantly associated with toxoplasmosis in this study ( $p=0.017$ ). Similar findings have been shown in studies conducted in Ethiopia and Indonesia.<sup>5,30</sup> This significant association found in the current study might be due to the fact that cats are known to be the definitive host of *Toxoplasma gondii* and they excrete the oocytes with feces; due to this, they are considered as the major source of *Toxoplasma gondii* infection to humans.<sup>31</sup> Studies conducted in Sudan and Tanzania showed contrasting results where contact with cats had no significant association with toxoplasmosis in pregnancy.<sup>11,27</sup>

Drinking untreated water was found to be significantly associated with toxoplasmosis in pregnancy ( $p=0.021$ ). Similar findings have been shown in other studies conducted in Rwanda and Tanzania.<sup>10,13</sup> This association might be due to the fact that untreated water may contain *Toxoplasma gondii* oocytes that can cause toxoplasmosis in pregnancy. It has been shown that *Toxoplasma gondii* oocytes can maintain its infectivity in water for about 2 years.<sup>32</sup>

In this study, HIV infection was also significantly associated with toxoplasmosis in pregnancy ( $p=0.020$ ). Similar findings were seen in the study conducted in Angola.<sup>4</sup> *Toxoplasma gondii* is one of the opportunistic parasite in HIV infected individuals. This association might be due to immuno-deficiency associated with some HIV positive patients especially those who are not compliance on anti-retroviral drugs. It could also be due to high prevalence of HIV in this region. In contrast, a study done in Brazil showed no significant association between HIV infection and toxoplasmosis in pregnancy probably due to lower prevalence of HIV infection found in Brazil as compared to Uganda.<sup>11,17</sup>

### **Study strength and limitations**

This is the first documented study on seroprevalence and factors associated with toxoplasmosis in pregnant women in Uganda and for that, the results will be used as a baseline for other studies in the country. It was an institutional based cross sectional study, so the findings may not generalize the entire population however it cover big catchment areas in western Uganda.

### **CONCLUSION**

This study demonstrated that the seroprevalence of toxoplasmosis among pregnant women who attended antenatal care clinic at KIUTH is low as compared to the global seroprevalence. Increased maternal age, contact with cats, drinking untreated water and HIV infection were the independent factors associated with toxoplasmosis in pregnancy. We recommend antenatal care clinics in Uganda to provide counselling and testing to pregnant women as well as education to avoid contact with cats and drinking untreated water in order to prevent toxoplasmosis during pregnancy.

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