


# Seroprevalence of Anti-*Toxoplasma gondii* Antibodies and Associated Factors Among Pregnant Women Attending Antenatal Care at Debre Markos Referral Hospital, Northwest Ethiopia

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## ABSTRACT

**BACKGROUND:** *Toxoplasma gondii* is an obligate intracellular protozoan parasite that causes a zoonotic disease called toxoplasmosis. If the infection acquired during pregnancy is not detected and treated early, the parasite can be transmitted transplacentally to the fetus, resulting in congenital toxoplasmosis, which likely leads to serious consequences in the fetus. Toxoplasmosis constitutes a major public health problem particularly in low- and middle-income countries including Ethiopia. This study aimed to determine the seroprevalence and to assess the potential risk factors of toxoplasmosis among pregnant women attending antenatal care at Debre Markos Referral Hospital, Northwest Ethiopia.

**METHODS:** In this cross-sectional study, data on the sociodemographic and potential obstetric and behavioral risk factors were gathered through pretested structured questionnaires, and 3 mL of venous blood was also drawn from each of randomly selected 233 study subjects. The serum samples were separated from the blood samples and tested for anti-*Toxoplasma* antibody using Toxo-latex slide agglutination test. Logistic regression analysis was used to examine the association between risk factors considered and *T. gondii* infection.

**RESULTS:** The overall prevalence of *T. gondii* infection was 67.8%, indicating a high prevalence of toxoplasmosis in the study area. In multivariate analysis, keeping domestic cat at home was found to be the only explanatory variable of toxoplasmosis (adjusted odds ratio = 2.449, 95% confidence interval = 1.183–5.070,  $P = .016$ ). All sociodemographic variables and most of the potential obstetric and behavioral risk factors were not statistically significant explanatory variables of *T. gondii* infection.

**CONCLUSION:** The prevalence of toxoplasmosis is high in the study area. Thus, pregnant women should be aware of the potential risk of the disease when keeping cats at home and management of their litter. Inclusion of serologic screening for *T. gondii* infection at antenatal care is warranted for prevention of congenital toxoplasmosis.

**KEYWORDS:** Debre Markos, Ethiopia, pregnant women, seroprevalence, *Toxoplasma gondii*

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## Introduction

*Toxoplasma gondii*, a member of the Apicomplexa, is an etiologic agent of a zoonotic food-borne disease called toxoplasmosis. Toxoplasmosis is a significant public health concern worldwide, particularly in developing countries with warmer climates, where the public health infrastructure is less well established and the highly resistant oocyst contaminates the environment longer.<sup>1</sup> In general, *T. gondii* infection usually causes no illness or a mild clinical sign in immunocompetent individuals, but the outcomes of the infection may become severe and can occasionally be fatal in immunocompromised people, such as patients with human immunodeficiency virus (HIV)/AIDS or pregnant women.<sup>2,3</sup>

*Toxoplasma* infection is acquired through direct or indirect contact with cat feces. A member of the cat family, *Felidae* is the only definitive host and sheds environmentally resistant oocyst form of the pathogen in its feces.<sup>4</sup> Although a wide

range of hosts including human being act as intermediate host, in humans *T. gondii* infection is usually acquired by consumption of raw or undercooked meat. Infection can also be acquired by ingesting unwashed vegetables and fruit, drinking water containing oocysts excreted in the feces of infected cats, or contact with cat litter or soil.<sup>5,6</sup> In rare cases, transmission by blood transfusion or organ transplantation has also been reported.<sup>4</sup>

*Toxoplasma* infection acquired for the first time during pregnancy, if left undiagnosed and untreated, can be transmitted transplacentally to the fetus and results in congenital toxoplasmosis.<sup>7</sup> The risk of congenital transmission varies with the trimester during which maternal infection was acquired. The risk increases with trimesters of pregnancy; it is about 25% in the first, 54% in the second, and 65% in the third trimester of pregnancy.<sup>8,9</sup> Congenital toxoplasmosis can result in spontaneous abortion, still-birth, fetal death, neonatal death, or diverse congenital defects.<sup>10</sup> It has been well documented



that early identification and treatment of acutely infected pregnant women reduces rates of vertical transmission and disease severity in the affected fetus.<sup>11</sup>

Globally, it is estimated that about one-third of the population is infected with *T gondii*.<sup>12</sup> The infection is present in every country, and seropositivity rates range from less than 10% to more than 90%, with high seroprevalence (>50%) occurring in countries where raw meat is commonly eaten and in tropical regions of Latin America or sub-Saharan Africa (SSA) where cats are numerous and the climate is favorable for survival of oocysts.<sup>13,14</sup>

According to the available published data, the seroprevalence of *T gondii* in Ethiopia among pregnant women varies considerably, the highest being reported from towns such as Gondar,<sup>15</sup> Jimma,<sup>16</sup> and Debre Tabor,<sup>17</sup> whereas there was a relatively low seroprevalence from Bahir Dar.<sup>18</sup> The major risk factors that have been identified so far for *T gondii* infection in Ethiopia include consumption of raw vegetable, presence of cat and its contact at home, HIV infection status, history of eating raw meat, and engagement in farming activities that involve contact with soil.<sup>16,19</sup>

Pregnancy is a unique immune condition often characterized as a highly risky state for the mother and fetus alike. Infection as a cause of maternal death is especially prominent in SSA compared with other regions of the world.<sup>20</sup> *Toxoplasma gondii* is common among childbearing age of women and pregnant women in Ethiopia.<sup>21</sup> In the present study area, like other parts of Ethiopia, practice of eating raw or undercooked meat and vegetables, keeping domestic cats at home, and presence of stray cats are common. Thus, for the success of designing and mounting prevention strategies in each local context, identifying the major risk factors associated with *T gondii* infection is important.

Besides, there is no recently published report that reveals the prevalence and associated risk factors of *T gondii* infection among pregnant mothers in the study area. Furthermore, pregnant women in the study area are not screened for *T gondii* infection during their prenatal care visit. We, therefore, conducted this study to determine the seroprevalence and to identify the major explanatory factors associated with *T gondii* infection among pregnant women attending antenatal care (ANC) at Debre Markos Referral Hospital, Ethiopia.

## Materials and Methods

### *Study design, period, site, and population*

A cross-sectional study was conducted from November 2015 to March 2016 among pregnant women who attended ANC at Debre Markos Referral Hospital, Amhara Region, Northwest Ethiopia.

Debre Markos is a town located 300 km northwest of the capital city, Addis Ababa, and 256 km from Bahir Dar, the capital city of the Amhara Regional State. The town had a total population of 107 129, of which 55 707 were women (52%) and

51 422 were men (48%). Debre Markos Referral Hospital provides service to an estimated 2 397 876 persons living in the town and to its surrounding populations. Apart from other services, the hospital provides ANC and delivery services for pregnant women.

All pregnant women attending ANC of Debre Markos Referral Hospital were considered as a source, whereas pregnant women who were willing to participate and consented to provide blood samples during the study period were considered as a study population. Pregnant women who declined to participate or refused to provide blood samples were excluded from this study.

### *Sample size determination and sampling technique*

Single population proportion formula<sup>22</sup> was used to determine the sample size of this study by considering 95% confidence interval (CI); critical value at 5% level ( $Z=1.96$ ); margin of error ( $d=5\%$ ); 18.5% seroprevalence of *T gondii* among pregnant women from previous study at Felege Hiwot Referral Hospital, Bahir Dar town, Northwest Ethiopia<sup>18</sup>; and 10% for expected nonresponse rate:

$$n = \frac{Z^2 P (1 - P)}{d^2}$$

Therefore, a total of 255 subjects were invited to participate in this study. Simple random sampling method was used to select the study participants among ANC attendees until the required sample size was obtained.

### *Method of data collection and processing*

*Questionnaire survey.* After explaining the objectives of the study and obtaining consent, data were gathered using a pre-tested questionnaire. Trained nurses working at ANC were in charge of distributing and collecting the filled questionnaires from the study subjects.

*Blood sample collection and processing.* Following the completion and handing over of questionnaires to nurses, 3 mL of venous blood was drawn and collected in EDTA-free tubes from each of the study participant by a trained laboratory technologist following standard operational procedures. Codes were written on the clinic cards of all enrolled mothers to avoid repeated inclusions during their subsequent visits. The blood specimens were allowed to clot at room temperature and then serum samples were separated by centrifugation at 3000 rpm for 5 minutes and collected in labeled tubes and transported with ice box from Debre Markos Referral Hospital to Biomedical and Microbiology laboratory, Biology Department, Bahir Dar University, for storage at  $-20^\circ\text{C}$  until use. In general, standardized procedures were strictly followed during blood sample collection, storage, and analysis.

*Serological detection of anti-Toxoplasma antibodies.* Toxo-latex is a slide agglutination test for the qualitative and semiquantitative detection of anti-*Toxoplasma* antibodies. The latex particles coated with soluble *T gondii* antigen are agglutinated when mixed with serum samples containing anti-*Toxoplasma* antibodies. The presence or absence of anti-*Toxoplasma* antibodies is detected by macroscopic examination of visible agglutination on the slide. The presence of agglutination indicates an antibody concentration equal to or greater than 4 IU/mL. The sensitivity and specificity of Toxo-latex slide agglutination test are 96.1% and 89.6%, respectively. The assay was conducted according to the manufacturer's instruction (SPINREACT, S.A./S.A.U Ctra. Santa Coloma, 7 E-17176 SANT ESTEVE DE BAS [GI] Spain).

*Data processing.* The data were checked for accuracy and completeness, and then entered and analyzed using Statistical Package for Social Sciences (SPSS) version 20. Descriptive analysis was used to describe the sociodemographic and clinical characteristics of study participants and to determine the prevalence of *T gondii* infection.  $\chi^2$  and univariate logistic regression analyses were also done to find the association between sociodemographic factors, clinical characteristics, and risk factors with prevalence of toxoplasmosis. All the variables with  $P < .25$  in univariate analysis were included in the subsequent multivariate logistic regression model for the objective of identifying the major explanatory variables.<sup>23</sup> Odds ratio (OR) at 95% CI was used as a measure of strength of association between risk factors considered and *T gondii* infection. In all cases, value of  $P < .05$  was considered as statistically significant.

#### *Ethical consideration*

An ethical clearance was granted by the Ethical Clearance Committee of the Postgraduate, Research and Community Service Office of College of Science, Bahir Dar University, and then a letter of support was written to Debre Markos Referral Hospital for possible cooperation. The purpose of the study was explained to the study participants, and then written informed consent was obtained from all voluntary subjects prior to their involvement in the study. In addition, the pregnant women were also told that they have the right to refuse to participate or withdraw from the study at any time. Blood samples and relevant data were collected using codes. To protect the confidentiality of the laboratory results and the information gathered, names of the study subjects and their card numbers were not recorded. Furthermore, privacy of the individuals was maintained by letting study subjects to independently respond to the questionnaire. Those who were found to be positive for *T gondii* were communicated back to ANC of Debre Markos Referral Hospital for monitoring and further management.

## Results

### *Sociodemographic and clinical features of the study participants*

Of the total 255 women who consented to participate in this study, 22 changed their mind not to be included. Thus, a total of 233 pregnant women were considered both for questionnaire survey and for anti-*Toxoplasma* antibodies seroprevalence analyses. The age of subjects ranged from 18 years (youngest) to 42 years (oldest), with the mean age of 26.11 years and standard deviation of  $\pm 4.56$  years. The majority (98 [42.1%]) of the study participants belonged to the category of 21 to 25 years of age, followed by 84 (36.1%) belonging to the age group of 26 to 30 years. Of the total study subjects, 207 (88.8%) were urban dwellers, 87 (37.3%) attained college/university certificate, and 100 (42.9%) were occupationally housewives.

Most (45.9%) of the women were in their third trimester, whereas 92 (39.5%) were in the second trimester, and the remaining 34 (14.6%) were in the first trimester at the time of this survey. Regarding gravidity, primigravida accounted for the most (100 [42.9%]), followed by those who were secundigravida (87 [37.3%]) and multigravida (46 [19.7%]). Pregnancy-related outcomes, for example, history of abortion, were reported by 37 (15.9%) subjects. Of the total study participants, 26 (11.2%) were HIV-positive (Table 1).

### *History of previous exposure of subjects to various risk factors of T gondii*

Among the study participants, 11 (4.7%) had a history of blood transfusion, 65 (27.9%) kept domestic cats in their homes, and 56 (24%) had a history of cleaning cat feces. More than half (127 [54.5%]) and nearly half (113 [48.5%]) of the pregnant women had a habit of eating unwashed/raw vegetables and raw/undercooked meat, respectively. Most (209 [89.7%]) of the study participants used tap water as a source of drinking water. Only 40 (17.2%) of the study participants reported that they had contact with soil, and most (94%) had hand washing practice with soap and water (Table 2).

### *Seroprevalence of T gondii infection and its association with sociodemographic characteristics*

Of the total 233 serum samples examined, 67.8% were found to be positive for *T gondii*-specific antibodies. The association between toxoplasmosis and sociodemographic characteristics of the study subjects is presented in Table 1. A slightly higher rate of *T gondii* infection was observed among women in the age group of 31 to 35 (80%) years than those under the age categories of 36 to 40 (75%) and 21 to 25 (72.4%) years. However, the difference in the distribution was not statistically significant across the age groups ( $P = .49$ ). Regarding residence, higher seroprevalence of toxoplasmosis was found among

**Table 1.** Association between toxoplasmosis and sociodemographic characteristics and obstetric, clinical variables of the study subjects.

VARIABLES	NO. EXAMINED	SEROPREVALENCE		$\chi^2$ (DF)	P VALUE
		POSITIVE	NEGATIVE		
Age interval, y					
15-20	24 (10.3%)	15 (62.5%)	9 (37.5%)		
21-25	98 (42.1%)	71 (72.4%)	27 (27.6%)		
26-30	84 (36.1%)	52 (61.9%)	32 (38.1%)	4.41(5)	.49
31-35	15 (6.4%)	12 (80%)	3 (20%)		
36-40	8 (3.4%)	6 (75%)	2 (25%)		
41-45	4 (1.7%)	2 (50%)	2 (50%)		
Residence					
Urban	207 (88.8%)	143 (69.1%)	64 (30.9%)	1.37(1)	.24
Rural	26 (11.2%)	15 (57.7%)	11 (42.3%)		
Educational status					
Illiterate	30 (12.9%)	20 (66.7%)	10 (33.3%)		
Primary education	39 (16.7%)	29 (74.4%)	10 (25.6%)	1.08(3)	.78
Secondary education	77 (33%)	50 (64.9%)	27 (35.1%)		
College/above	87 (37.3%)	59 (67.8%)	28 (32.2%)		
Occupational status					
Farmer	17 (7.3%)	11 (64.7%)	6 (35.3%)		
Housewives	100 (42.9%)	65 (65%)	35 (35%)	0.89(3)	.83
Business women	57 (24.5%)	40 (70.2%)	17 (29.8%)		
Government employed	59 (25.3%)	42 (71.2%)	17 (28.8%)		
Trimester of pregnancy					
First trimester	34 (14.6%)	27 (79.4%)	7 (20.6%)		
Second trimester	92 (39.5%)	59 (64.1%)	33 (35.9%)	2.68(2)	.26
Third trimester	107 (45.9%)	72 (67.3%)	35 (32.7%)		
Gravidity					
Primigravida	100 (42.9%)	61 (61%)	39 (39%)		
Secundigravida	87 (37.3%)	65 (74.7%)	22 (25.3%)	4.08(2)	.12
Multigravida	46 (19.7%)	32 (69.6%)	14 (30.4%)		
History of abortion					
Yes	37 (16%)	26 (70.3%)	11 (29.7%)		
No	194 (84%)	131 (67.5%)	63 (32.5%)	0.12(1)	.73
HIV status					
Positive	26 (11.2%)	15 (57.7%)	11 (42.3%)		
Negative	207 (88.8%)	143 (69.1%)	64 (30.9%)	1.37(1)	.24

HIV, human immunodeficiency virus.

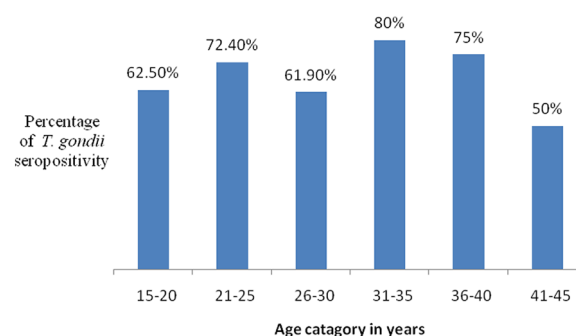
**Table 2.** Association between seroprevalence of *Toxoplasma gondii* infection and its potential risk factors.

VARIABLES	NO. EXAMINED	SEROPREVALENCE OF <i>T GONDII</i> INFECTION		$\chi^2$ (DF)	P VALUE
		POSITIVE	NEGATIVE		
<b>Blood transfusion</b>					
Yes	11 (4.7%)	11 (100%)	0 (0%)		
No	222 (95.3%)	147 (66.2%)	75 (33.8%)	5.48(1)	.02
<b>Presence of domestic cat at home</b>					
Yes	65 (27.9%)	50 (76.9%)	15 (23.1%)		
No	168 (72.1%)	108 (64.3%)	60 (35.7%)	3.43(1)	.06
<b>Cleaning cat feces</b>					
Yes	56 (24%)	41 (73.2%)	15 (26.8%)		
No	177 (76%)	117 (66.1)	60 (33.9%)	0.99 (1)	.32
<b>History of eating raw/undercooked meat</b>					
Yes	113 (48.5%)	76 (67.3%)	37 (32.7%)	0.03(1)	.86
No	120 (51.5%)	82 (68.3%)	38 (31.7%)		
<b>History of eating raw/unwashed vegetable</b>					
Yes	127 (54.5%)	85 (66.9%)	42 (33.1%)	0.09(1)	.75
No	106 (45.5%)	73 (68.9%)	33 (31.1%)		
<b>Water source</b>					
River	13 (5.6%)	5 (38.5%)	8 (61.5%)		
Well	11 (4.7%)	6 (54.5%)	5 (45.5%)	6.16(2)	.05
Tap water	209 (89.7%)	147 (70.3%)	62 (29.7%)		
<b>Soil contact</b>					
Yes	40 (17.2%)	23 (57.5%)	17 (42.5%)		
No	193 (82.8%)	135 (69.9%)	58 (30.1%)	2.4(1)	.13
<b>Hand washing with soap</b>					
Yes	219 (94%)	149 (68%)	70 (32%)		
No	14 (6%)	9 (64.3%)	5 (35.7%)	0.85(1)	.77

urban residents compared with rural counterparts; nevertheless, this was not statistically significant ( $P = .24$ ) (Figure 1).

Relatively higher (74.4%) rate of *T gondii* infection was detected among subjects who attained primary level of education compared with participants who were illiterate and those who had attained secondary education and college and above. However, this was not statistically significant ( $P = .78$ ).

Occupationally, nearly two-thirds of farmers (64.7%), housewives (65%), business women (70.2%), and government employee (71.2%) were found to be positive for *T gondii*, and no statistically significant association was found between occupation status of the study subjects and prevalence of *T gondii* infection ( $P = .83$ ).

**Figure 1.** Percentage of *Toxoplasma gondii* seropositivity within each age categories of the study subjects.

Of the total 87 participants who had pregnancy for second time and of the total 34 subjects who were at the first trimester of gestational age, 75% and almost 80% subjects, respectively, were found to be positive for *T gondii* infection. Despite the observed disparity in the distributions of the infection among the categories of gravidity ( $P=.13$ ) and gestational age ( $P=.26$ ), no statistically significant associations were obtained with the disease.

Of the total 26 (11.2%) participants who tested HIV seropositive, most (57.7%) were found to be positive for toxoplasmosis; however, in  $\chi^2$  analysis no statistically significant association was detected between HIV status of the present participants and seroprevalence of toxoplasmosis ( $P=.24$ ).

The seroprevalence rate of *T gondii* infection among the study participants who had a previous history of abortion was slightly higher (70.3%) compared with participants who did not have a history of abortion (67.5%); however, there was no statistically significant association between history of abortion and prevalence of toxoplasmosis.

#### *Seroprevalence of toxoplasmosis and potential risk factors*

From a total of 11 pregnant women who had a history of blood transfusion, all (100%) subjects were detected positive for toxoplasmosis. There was statistically significant association between blood transfusion and prevalence of toxoplasmosis ( $P=.02$ ). Regarding practice of keeping domestic cat at home, relatively higher (50 [76.9%]) *T gondii* seropositivity was observed among subjects who had cat in their home compared with those who did not have this practice. However, the association was not statistically significant ( $P=.06$ ).

Moderately higher seroprevalence of *T gondii* was observed in subjects who were engaged in cleaning cats' feces at their residence compared with those who did not. However, no statistically significant association was observed between cleaning of cats' excrement and *T gondii* infection ( $P=.32$ ). Regarding history of eating raw/undercooked meat, no significant difference in the distribution of seropositivity of the infection was observed between subjects who had the history of eating raw/undercooked meat and those who did (67.3% vs 68.3%), and no statistically significant association was also observed between history of eating raw/undercooked meat and risk of the disease under study ( $P=0.86$ ). Similarly, almost similar rates of *T gondii* seropositivity were observed between subjects who had a habit of eating raw/undercooked vegetable and those who did not (66.9% vs 68.9%), and no significant association was also observed between the habit of consuming raw/unwashed vegetable and fruits and seroprevalence of *T gondii* infection ( $P=.75$ ).

Significantly high (70.3%) *T gondii* infection was seen among the study subjects who used tap as a source of drinking water compared with those who used well and river water ( $P=0.03$ ). Among 40 pregnant women who had a previous

history of contact with soil, 23 (57.5%) were found positive for *T gondii*, whereas the rate of infection was 69.9% among those who did not have a previous history of contact with the soil. However, there was no significant association between a history of soil contact and seroprevalence of *T gondii* ( $P=.13$ ).

Comparatively similar rate of *T gondii* infection was detected among those who had the practice of washing their hands with soap and those who did not have this habit ( $P=.77$ ). The association between the seroprevalence of *T gondii* infection and its potential risk factors is presented in Table 2.

#### *Factors associated with prevalence of toxoplasmosis*

*Univariate analysis of the risk factors of T gondii infection in study participants.* In univariate analysis, no statistically significant associations were found between *Toxoplasma* infection and sociodemographic, behavioral, and obstetric variables (Table 3). However, statistically significant association was observed between the status of gravidity and *T gondii* infection. Almost 2 times increased risk of *T gondii* infection was observed among women who had second pregnancy (crude odds ratio=1.88, 95% CI=1.00-3.54,  $P=.04$ ).

*Multivariate logistic regression analysis of selected variables.* All sociodemographic, behavioral, and obstetric variables with a  $P<.25$  in univariate analysis were selected and entered for multivariate logistic regression model to identify the most important explanatory variables of the disease. After multivariate analysis, it was found that the presence of domesticated cats at home was a statistically significant explanatory variable of *T gondii* infection ( $P=.016$ ; adjusted odds ratio=2.449, 95% CI=1.183-5.070). The odds of being infected by *T gondii* in pregnant women who had domestic cats at their home was 2.45 times higher than pregnant women who did not have domestic cats in their homes (Table 3).

## Discussion

Toxoplasmosis is one of the most prevalent parasitic infectious diseases of medical and veterinary importance due to its implication in abortion and congenital disease in its intermediate hosts.<sup>24,25</sup> Infections with this protozoan have been reported from all around the world, and it has been estimated that 20% to 90% of adults have come into contact with the parasite during their lifetime.<sup>26</sup> There are few epidemiological data indicating a very high prevalence of *T gondii* infection in humans in Ethiopia.<sup>2</sup> This study was set to examine the prevalence of *T gondii* infection among pregnant women who attended ANC center of Debre Markos Referral Hospital, Ethiopia.

The seroprevalence of *T gondii* infection in humans can be graded as high (>50%), medium (30%- 50%), and low (<30%).<sup>27</sup> In this study, more than half (67.8%) of the study participants were tested positive for *T gondii*, which indicates a high prevalence of *T gondii* infection in the study area. In this cross-sectional study, we did not find any statistically

**Table 3.** Univariate and multivariate logistic regression analysis of variables potentially associated with *Toxoplasma gondii* infection among pregnant women in Debre Markos Referral Hospital, Northwest Ethiopia.

VARIABLES	SEROPREVALENCE OF <i>T GONDII</i>		COR (95% CI, P VALUE)	AOR (95% CI, P VALUE)
	POSITIVE (%)	NEGATIVE (%)		
<b>Residence</b>				
Urban	143 (69.1%)	64 (30.9%)	1.00	1.00
Rural	15 (42.3%)	11 (57.7%)	0.61 (0.266-1.402, .24)	0.69 (0.25-1.92, .48)
<b>Gestation period</b>				
First trimester	27 (79.4%)	7 (20.65)	1.00	1.00
Second trimester	59 (64.1%)	33 (35.9%)	0.46 (0.18-1.18, .10)	0.40 (0.15-1.09, .07)
Third trimester	72 (67.3%)	35 (32.7%)	0.53 (0.21-1.34, .18)	0.52 (0.19-1.40, .20)
<b>Gravidity</b>				
Primigravida	61 (61.0%)	39 (39.0%)	1.00	1.00
Secundigravida	65 (74.7%)	22 (25.3%)	1.88 (1.00-3.54, .04)*	1.91 (0.98-3.73, .05)
Multigravida	32 (69.6%)	14 (30.4%)	1.46 (0.69-3.08, .31)	1.73 (0.76-3.91, .18)
<b>HIV status</b>				
Positive	15 (57.7%)	11 (42.3%)	0.61 (0.26-1.40, .24)	0.43 (0.17-1.08, .07)
Negative	143 (69.1%)	64 (30.9%)	1.00	1.00
<b>Presence of cat at home</b>				
Yes	50 (76.9%)	15 (23.1%)	1.85 (0.95-3.57, .06)	2.45 (1.18-5.07, .01)*
No	108 (64.3%)	60 (35.7%)	1.00	1.00
<b>Water source</b>				
River water	5 (38.5%)	8 (61.5%)	0.24 (0.08-0.83, .02)	0.26 (0.06-1.08, .06)
Well water	6 (54.5%)	5 (45.5%)	0.50 (0.14-1.72, .27)	0.41 (0.10-1.60, .20)
Tap water	147 (70.3%)	62 (29.7%)	1.00	1.00
<b>Soil contact</b>				
Yes	23 (57.5%)	17 (42.5%)	1.72 (0.85-3.45, .12)	0.77 (0.34-1.76, .54)
No	135 (69.9%)	58 (30.1%)	1.00	1.00

AOR indicates adjusted odds ratio; CI, confidence interval; COR, crude odds ratio; HIV, human immunodeficiency virus.

\*Statistically significant at  $P < .05$ .

significant association of *T gondii* infection with most of the potential risk factors analyzed. However, the contribution of eating raw meat, ownership and contact with domestic cats, management of cats' litter, hand hygiene practices, eating unwashed fruits or vegetables, lack of awareness of pregnant women toward the source of infection, modes of transmission, and prevention methods for the observed higher prevalence of this disease in the study area cannot be ruled out.

The prevalence of toxoplasmosis in the current finding is almost similar to the prevalence of *T gondii* infection previously reported among pregnant women in Debre Tabor, Northwest Ethiopia (68.4%),<sup>17</sup> Cameroon (70%)<sup>28</sup> and (69.9%),<sup>29</sup> and Tehran, Iran (68.9%).<sup>30</sup> However, our finding is

lower than the prevalence of *T gondii* infection reported from Gondar, Northwest Ethiopia (88.6%),<sup>15</sup> Addis Ababa (85.4%),<sup>31</sup> Jimma town (83.6%),<sup>16</sup> and Kinshasa, Democratic Republic of Congo (80.3 %).<sup>32</sup>

On the contrary, the finding of this study is considerably higher than studies conducted in Bahir Dar, Ethiopia (18.5%),<sup>18</sup> Mozambique (18.7%),<sup>33</sup> Sudan (20.2%),<sup>34</sup> Burkina Faso (20.3%),<sup>35</sup> Kosovo (29.4%),<sup>26</sup> Jazan Province, Saudi Arabia (24.1%),<sup>36</sup> and Tanzania (41.7%).<sup>37</sup>

The discrepancy in the rates of *T gondii* infection across studies may be due to the differences in the sensitivity of diagnostic methods used, climatic condition, cultural difference in hygienic and feeding habits, and literacy statuses of the study

subjects. Variations in degree of contact, management and interaction of humans with definitive and reservoir hosts of *T gondii*, and lack of awareness about the disease and its transmission may also account for the difference across studies within the same or different populations.

In this study, the presence of domestic cats at home showed significant association with *T gondii* infection, and the odds of being infected with *T gondii* was 2.45 times higher among pregnant women who had domestic cats at their homes than those who did not have domestic cats in their homes. This is consistent with the studies reported from Gondar University Teaching Hospital,<sup>15</sup> Bahir Dar Felege Hiwot Referral Hospital, Northwest Ethiopia,<sup>18</sup> and Debre Tabor,<sup>17</sup> in which cats' presence at home was found to be a significant risk factor for contracting this illness. Contrary to these findings, multiple studies carried out elsewhere failed to implicate cats' presence at home as a predictor of risk of *T gondii* infection, even though cats are the primary sources of oocyst that causes human and livestock *T gondii* infection.<sup>31,32,34,37</sup> It should be noted that the likelihood of *T gondii* infection might not only be due to the presence of cats at home but also the way the cats' litter is handled.

Blood transfusion,<sup>38</sup> consumption of raw/unwashed vegetables,<sup>39</sup> and source of drinking water<sup>40</sup> have also been documented as risk factors of toxoplasmosis. However, Gelaye et al<sup>31</sup> reported absence of statistically significant association between blood transfusion, consumption of raw vegetables, and *T gondii* infection. This study also did not find statistically significant association between these potential risk factors and prevalence of toxoplasmosis. This may suggest that these risk factors play a limited role in this study for transmission of the parasite in study participants. Besides, the absence of a statistically significant relationship between the prevalence of *Toxoplasma* infection and eating raw/undercooked meat among investigated population does not confirm that this factor has no influence on the transmission of toxoplasmosis. This finding is also comparable with the report of Endris et al,<sup>15</sup> Agmas et al,<sup>17</sup> Gelaye et al,<sup>31</sup> Doudou et al,<sup>32</sup> and Shao et al.<sup>37</sup> This finding is inconsistent with the report of Awoke et al,<sup>18</sup> who reported that having a habit of eating raw/undercooked meat is found to be a major risk factor contributing to maternal *T gondii* infection in the study participants.

The existence of a strong agro-pastoral activity especially in rural areas attributed to an increase in the spread of zoonotic diseases in rural residence. Several studies have demonstrated the coexistence between humans and animals as contributing factor raising the risk of *T gondii* infection.<sup>41</sup> On the contrary, better socioeconomic condition, improved access to potable water and sanitation facilities, improved awareness about good hygiene, education, and rare exposure to stray cats in urban setting are supposed to minimize the risk of toxoplasmosis. Contrary to these observations, recent studies by Aqeely et al<sup>36</sup> and Nasir et al<sup>42</sup> reported a highly significant association

between the seroprevalence of *T gondii* infection in pregnant women residing in urban settings compared with those women residing in rural areas. Consistent with these findings, this study also found a higher prevalence of toxoplasmosis in pregnant women from urban areas compared with those from rural areas, but no statistically significant association between residence and toxoplasmosis was obtained.

Olariu et al<sup>43</sup> in Timis County (Romania) reported an increase in *Toxoplasma* antibodies with age groups. In their study, the highest (80%) seropositivity for *T gondii* was detected among participants within the age range of 31 to 35 years. This finding is comparable with the study reported from Cameroon<sup>28</sup> and another study conducted in Ethiopia.<sup>18</sup> Conversely, Agmas et al<sup>17</sup> found significant association between age group and prevalence of toxoplasmosis and implicated age as possible risk factors of *T gondii* infection, suggesting that the higher age groups had long duration of exposure.

In our study, most of the pregnant women in the first, second, and third trimester of their gestation were infected with *T gondii*, but no significant association was found between seropositivity of anti-*T gondii* antibodies and gestational ages of the pregnant women, and this is comparable to a finding reported from greater Accra region of Ghana.<sup>44</sup> Contrary to this, 2 studies from Ethiopia by Endris et al<sup>15</sup> and Fenta<sup>45</sup> demonstrated statistically significant positive association between the seroprevalence of *T gondii* infection and the second and third trimesters of gestational age of pregnancy.

The seroprevalence of toxoplasmosis is high in immunocompromised patients such as HIV/AIDS, and transplant or cancer. In general, the seroprevalence of antibodies to *T gondii* among HIV-infected patients mirrors the rate of seropositivity in the general population.<sup>46</sup> In SS, an increase in toxoplasmosis prevalence has been documented in association with HIV/AIDS.<sup>47</sup> In this study, about 60% of HIV-positive pregnant women were found to be positive for toxoplasmosis, but this was not significantly associated with *T gondii* infection. This is in line with studies reported from Addis Ababa, Ethiopia,<sup>31</sup> and Tanzania,<sup>37</sup> which also reported similar finding.

In our study, marginally significant nearly 2-fold of *T gondii* infection was observed among secundigravida and multigravida pregnant women. This finding is supported by the reports of Awoke et al<sup>18</sup> and Yohanes et al,<sup>39</sup> who also found no significant of association of gravidity with seroprevalence of *T gondii* infection.

The Toxo-Latex agglutination test we used in our serodiagnosis could not differentiate between recent and past *T gondii* infections, and we did not follow up and determine the serostatus of infants born from *T gondii*-infected mothers.

In conclusion, the seroprevalence of *T gondii* infection observed among pregnant women in our study is high. In this study, the presence of cats at home was identified as the only explanatory variable of *T gondii* infection. However, none of the sociodemographic, obstetric, and behavioral variables were



significantly associated with the prevalence of toxoplasmosis. Still, health education interventions are effective at increasing the awareness of ANC attendees toward the source of infection, modes of transmission, and prevention methods of the disease to mitigate the risk of maternal toxoplasmosis and its subsequent transmission to their fetus. Besides, serological screening of *T. gondii* infection during ANC should be considered at Debre Markos Referral Hospital.


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### Author Contributions

SM and AM conceived the study idea and developed the study design, SM conducted the study, analyzed data, and wrote the manuscript. AM and EN checked analyses and interpretation of the data and critically reviewed the manuscript. All authors read and approved the final manuscript.

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### Data Availability

All data generated during this study are not publicly available due individual privacy concerns, however are available from the corresponding author on reasonable request.

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