RESEARCH ARTICLE

Seroprevalence, risk factors and impact of *Toxoplasma gondii* infection on haematological parameters in the Ashanti region of Ghana: a cross-sectional study [version 1; peer review: 2 approved with reservations]

Samuel Kekeli Agordzo \(^{1}\), Kingsley Badu \(^{1,2}\), Mathew Glover Addo \(^{1}\), Christian Kwasi Owusu \(^{2}\), Abdul-Hakim Mutala \(^{1}\), Austine Tweneboah \(^{1}\), Dawood Ackom Abbas \(^{1}\), Nana Kwame Ayisi-Boateng \(^{3,4}\)

\(^{1}\)Theoretical and Applied Biology, Kwame Nkrumah University of Science and Technology, Kumasi, AK 192, Ghana
\(^{2}\)Kumasi Centre for Collaborative Research for Tropical Medicine, Kwame Nkrumah University of Science and Technology, Kumasi, Ghana
\(^{3}\)School of Medicine and Dentistry, Kwame Nkrumah University of Science and Technology, Kumasi, Ghana
\(^{4}\)The University Hospital, Kwame Nkrumah University of Science and Technology, Kumasi, Ghana

Abstract

**Background:** *Toxoplasma gondii* is an obligate, intracellular, apicomplexan parasite that causes toxoplasmosis. Although the global prevalence of toxoplasmosis has been estimated to be approximately 30%, there is limited seroprevalence data in Ghana, with a dearth of information on the impact of *T. gondii* on haematological parameters in exposed persons.

**Methods:** Questionnaires were administered to 300 consenting individuals to obtain demographic information and assessment of their risk of exposure to *T. gondii*. Using anti-*T. gondii* IgG/IgM combo test kits, seropositivity to *T. gondii* parasite-specific IgG and/or IgM was determined. A haematological analyser was used to measure haematological parameters.

**Results:** The participants included 58 males and 242 females, and ranged in age from 6 months to 84 years, with a median age of 27 years. There was an overall seroprevalence of 50.3% (n=151), with 49.7% (n=149) of the study participants seropositive for IgG and 1% (n=3) testing positive for IgM. Furthermore, the observed seroprevalence among pregnant women was 56.4% (n=62). With regards to the different communities in which the hospitals were located, a seroprevalence of 55.6% was observed in the rural community, 50.6% in the peri-urban community and 47.1% in the urban community. The study identified cat ownership, contact with cat litter [RR (95% CI: 1.76 (1.23-2.53), 1.66 (1.03-2.67), 1.25(1.00-1.57)) and age (p<0.001) as risk factors for infection. Analyses of haematological data also revealed significant differences between the red blood cell counts (p=0.038) and mean corpuscular volumes (p=0.0007) of seropositive and seronegative study participants.

**Conclusions:** About half of the study population, including a significant number of women of reproductive age carried antibodies against *T. gondii*, raising questions about the risk of congenital toxoplasmosis, as well as...
raising questions about the risk of congenital toxoplasmosis, as well as possible links to anaemia. We, therefore, recommend that screening for *Toxoplasma gondii* be included in the routine screening of pregnant women seeking antenatal care.

**Keywords**
Toxoplasma gondii, Haematology, Seroprevalence, IgG, Exposure, Risk Factors

**Corresponding author:** Kingsley Badu (kingsbadu@gmail.com)

**Author roles:** Agordzo SK: Formal Analysis, Investigation, Methodology, Writing – Original Draft Preparation; Badu K: Conceptualization, Funding Acquisition, Investigation, Methodology, Project Administration, Supervision, Validation, Writing – Original Draft Preparation, Writing – Review & Editing; Addo MG: Supervision, Validation, Writing – Review & Editing; Owusu CK: Data Curation, Formal Analysis, Writing – Review & Editing; Mutala AH: Data Curation, Investigation, Methodology; Tweneboah A: Data Curation, Investigation, Methodology, Validation; Abbas DA: Data Curation, Methodology, Resources; Ayisi-Boateng NK: Conceptualization, Data Curation, Investigation, Writing – Review & Editing

**Competing interests:** No competing interests were disclosed.

**Grant information:** AHM and SKA received graduate assistantship from the KNUST College of Science. KB received funds for laboratory supplies from the Africa Research Excellence Fund (AREF) Research Development Fellowship [MRF-157-0007-F-BADU]. Kingsley Badu is an Affiliate of the African Academy of Sciences (2017-2022).

**Copyright:** © 2019 Agordzo SK et al. This is an open access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

**How to cite this article:** Agordzo SK, Badu K, Addo MG et al. Seroprevalence, risk factors and impact of *Toxoplasma gondii* infection on haematological parameters in the Ashanti region of Ghana: a cross-sectional study [version 1; peer review: 2 approved with reservations] AAS Open Research 2019, 2:166 https://doi.org/10.12688/aasopenres.13022.1

**First published:** 26 Nov 2019, 2:166 https://doi.org/10.12688/aasopenres.13022.1
Introduction

Toxoplasma gondii — the causative organism for toxoplasmosis — is an obligate, intracellular, apicomplexan parasite with a wide geographic distribution and the ability to infect virtually any cell type across a broad host range, including humans, companion animals, livestock and wildlife. About a third of the world’s population is infected with T. gondii but the parasite does not usually cause clinically significant disease. Until recently, latent infections in humans were assumed to be asymptomatic; however, results from animal studies, personality and behavioural profiles, as well as psychomotor performance tests have led to a reconsideration of this assumption. Certain individuals, including foetuses, neonates, and the immune-compromised are at high risk for life-threatening complications from toxoplasmosis. Congenital transmission of T. gondii carries the risk of miscarriage or stillbirth, and children born with toxoplasmosis are likely to suffer from severe symptoms, such as hydrocephalus, calcifications of the brain or retinochoroiditis later in life if not treated.

People are typically infected by either accidentally ingesting infectious oocysts or eating undercooked meat containing tissue cysts (bradyzoites). Toxoplasma oocysts can be found in soil or water contaminated with cat faeces, making the consumption of raw vegetables and water from unsafe drinking sources important risk factors for infection. The consumption of raw or undercooked meat is also a risk factor as livestock and game often harbour bradyzoites. The parasite can also be transmitted in utero to a developing foetus if a woman is infected for the first time while pregnant. Solid-organ transplantation such as the heart, liver and kidney transplants are another means by which T. gondii infection can occur, although this is very rare.

During acute primary infection with T. gondii, anti-T. gondii immunoglobulin (Ig) M is initially produced. However, IgM titres decline over the next few months, becoming undetectable within a year. The immune system also produces anti-T. gondii IgG a few weeks after the initial infection. IgG antibody levels usually peak within one or two months after the infection but are still detectable throughout the lifetime of the infected individual. The seroprevalence of antibodies against T. gondii has been reported across the globe, and ranges from 51% to 72% in several countries in Latin America and the Caribbean, including Argentina, Brazil, Cuba, Jamaica, and Venezuela. In Scandinavia, the seroprevalence of antibodies specific to T. gondii is reported to vary between 11% and 28%, while in Southeast Asia, China and Korea, seropositivity to T. gondii exposure has been estimated to range from 4% to 39% in women of reproductive age. Furthermore, both active and latent infections of T. gondii have been reported in many African countries, particularly in individuals suffering from HIV/AIDS. For example, 88.2% of HIV-positive individuals seeking healthcare at Arba Minch Hospital in Ethiopia were also seropositive for T. gondii infection, and a study conducted in Burkina Faso among pregnant women revealed an overall seroprevalence of 31.4%.

In Ghana, Sefa-Boakye et al. and Ayi et al. reported seroprevalence rates of 83.6% and 92.5% among pregnant women in Kumasi and Accra, respectively. In the Central region of Ghana, Abu et al. reported an overall seroprevalence of 85% in a population-based study that investigated risk factors for T. gondii infection. A follow-up study later revealed that 2.6% of the study participants showed signs of ocular toxoplasmosis. In the Greater Accra region, Kwosie et al. analysed placental tissue samples from IgG-seropositive women post-delivery and estimated the risk of congenital transmission of the infection to be 39.8% based on the presence of T. gondii DNA in the placental samples.

In this study, we screened 300 individuals from three hospitals in the Ashanti region of Ghana and estimated the seroprevalence of T. gondii infection. Our primary objective was to investigate the seroprevalence, associated risk factors and haematological consequences of T. gondii infection in the Ashanti region of Ghana. Specifically, we determined 1) the seroprevalence of T. gondii-specific IgG and IgM; 2) the risk factors associated with T. gondii infection; and 3) the haematological consequences of T. gondii exposure in the Ashanti region of Ghana.

Methods

Ethical consideration

Prior to the commencement of the study, approval was obtained from all three hospitals. Ethical approval for the study was also given by the Committee on Human Research Publications and Ethics at the Kwame Nkrumah University of Science and Technology and Komfo Anokye Teaching Hospital in Kumasi, Ghana (reference number: CHRPE/AP/018/18). Written informed consent was obtained from all the study participants.

Study design

A cross-sectional study design was employed. Study participants were recruited during hospital visits that occurred between 30th January and 28th February 2018. Questionnaires were administered to obtain data on demographics and risk factors.

Study site

The study was conducted at three hospitals; namely, Kumasi South Hospital (KSH), Agona Government Hospital (AGH) and Kuntanase Government Hospital (KGH), all in the Ashanti region of Ghana. KSH is located in Atonsu, a community within the Asokwa Sub-metropolitan Assembly of the Kumasi Metropolis which has a population size of 1,730,249. Recruitment and sample collection were conducted from 31st January to 8th of February 2018 at KSH. At KGH, recruitment and data collection were carried out from 15th–16th and 27th–28th February 2018. KGH is situated in Kuntanase, the capital of the Bosumtwi district of the Ashanti Region with a population of 93,910 and located at about 28km from the capital city, Kumasi. AGH is located at about 37 km from Kumasi and is situated in the Sekyere South District, which has a population of 94,009. Recruitment and selection of participants took place from 19th–21st February 2018 at the Agona Government Hospital.

Participant selection

To be included in the study, individuals had to be at least six months of age and had to have been referred to the laboratories of one of the study hospitals (KSH, KGH and AGH).
Pregnant women were preferentially recruited into the study due to our interest in congenital toxoplasmosis. The selection of study subjects was based on their willingness to participate after being briefed about the study, and confirmed by signing or thumb-printing an informed consent form. In the case of minors, consent was obtained from parents or legal guardians. Critically ill patients (individuals in need of urgent medical attention) and children below 6 months were excluded from the study. We provided a written questionnaire to each of the participants as they waited for their turn at the laboratory of the various hospitals. The questionnaires were administered in the form of an interview to all participants by the investigators.

Variables
Data obtained from the questionnaires on factors that predispose respondents to infection (cat ownership, contact with cat litter, eating raw meat, consumption of raw or undercooked vegetables and sources of drinking water) were analysed. As part of the demographic data, age, sex, location (rural, urban or peri-urban), education and employment were also analysed. These variables were treated as predictor variables and compared with the anti-T. gondii serological results (endpoint variables).

Sample size determination
A minimum sample size of 207 was determined using the binomial model. This was calculated with a 95% CI and precision level of 5%: \( N = \left( \frac{Z^2 \times (P) \times (1-P)}{d^2} \right) \) using overall seroprevalence of 83.4% observed by Sefah-Boakye et al.\(^1\) in the Ashanti region. In this equation, \( N \) is the sample size, \( Z \) is the critical value of the binomial distribution at the 5% level (1.96), \( p \) is the overall seroprevalence (0.84), \( q = 1 – p \), and \( d \) (error) is the precision level (5%).

Quantitative variables
Participants who tested positive for parasite-specific IgG, IgM or both were classified as seropositive, while those with negative tests were described as seronegative. We compared the median haematological parameter values of the seropositive and seronegative categories to investigate the effects of infection on the different haematological indices.

Haematological analyses
Using sterile needles and syringes, qualified phlebotomists obtained 3 mL of venous blood from each participant. The blood samples were dispensed into labelled EDTA tubes to prevent clotting and were later used for haematological analyses. Complete blood counts were run on the blood using the Sysmex XP-300 Automated Haematology Analyzer. The analyser provided cell count data on red blood cells (RBC), white blood cells (WBC), lymphocytes, neutrophils and platelets. Other parameters measured included haemoglobin (Hb), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean platelet volume (MPV), plateletcrit and red blood cell distribution width (RDW).

Serological analyses
The blood samples were tested for the presence of anti-T. gondii IgG/IgM using the commercially available OnSite Toxo IgG/IgM rapid combo test manufactured by CTK Biotech, USA. This test kit runs on whole blood or serum. Following the manufacturer’s protocol, a 10 µL sample of blood was dropped into the sample/buffer well on the kit. Two drops of buffer were then added and allowed to stand for 10–15 min. Results were recorded as positive if both the test control line and M and/or G line(s) developed. Test results were seropositive for IgG if the G line developed in addition to the control (C) while blood samples were seropositive for IgM if the M line appeared in addition to the control. Seropositive results to IgG and IgM were obtained when both the M and G lines developed in addition to the control (C) band.

Data analysis
Data obtained from serological examination and questionnaires were keyed into Microsoft Excel (2016). Seroprevalence was calculated by expressing the number of seropositive individuals as a percentage of the total number tested. The chi-square and Mann-Whitney U tests were carried out in GraphPad Prism 6 (GraphPad Software, Inc., San Diego, CA, USA) to investigate the association between demographics and seropositivity, as well as to compare the differences in the median haematological values of seropositive and seronegative individuals respectively. STATA 14 was also used to determine the relative risk of seropositivity for each risk factor. Analysed results were considered statistically significant if p ≤ 0.05.

Results
In total, 300 participants were recruited for the study. Out of this number, 80.6% (n=242) were female, of which 45.5% (n=110) were pregnant. The median age for all participants was 27 years.

Seroprevalence of T. gondii
50.3% (n=151) of the 300 participants were seropositive for anti-T. gondii antibodies. Among the seropositive population, 98% (n=148) and 2% (n=3) of the participants were seropositive for IgG and IgM respectively, with 0.3% (n=1) of the participants seropositive for IgM but seronegative for IgG (IgM only), 49.3% (n=148) seropositive for IgG but seronegative for IgM (IgG only) and 1.3% (n=2) of the 151 seropositive participants seropositive for both IgG and IgM (Table 1).

The communities within which the hospitals are located were classified as urban (Kumasi South Hospital), rural (Agona Government Hospital) and peri-urban (Kuntanase Government Hospital) based on their distance from Kumasi, the capital city. (Our classification is based on the population of the communities, where urban is highly populated city area, followed by peri-urban then, the rural area with the lowest population). The Seroprevalence rates of 47.1%, 50.6% and 55.6% were therefore observed from the urban, rural and peri-urban areas, respectively (Table 2).

A total of 110 pregnant women were recruited into the study, including 37 in their first trimester, 41 in the second trimester.
parts of the world, there is limited data on the epidemiology of the infection in Ghana, and a majority of the few studies available are restricted to Accra, the capital city. Studies investigating the epidemiology of the infection typically do so by determining seroprevalence rates based on the detection of T. gondii-specific antibodies. T. gondii-specific IgG is associated with previous exposure to the parasite and is used as a marker for latent infection while IgM is used as a marker of acute infection

In this study, an overall seroprevalence of 50.3% was observed, which is similar to a study from Accra that reported a seroprevalence rate of 49.7% \(^\text{[12]}\), in contrast to another Accra-based study that reported 92.5% seroprevalence \(^\text{[12]}\). The study also found a seroprevalence rate of 1.0% for parasite-specific IgM, suggesting active infections in these study participants. Other studies have previously reported fairly low seroprevalence of IgM, including 6.0% in the Central region of Ghana and 2.5% in Southwestern Ethiopia \(^\text{[15,18]}\). However, anti-T. gondii IgM seroprevalence rates as high as 29.7%, 39.1% and, even, 76.1% have been reported in Accra and Kumasi \(^\text{[11,12,17]}\). Variations in the seroprevalence of anti-T. gondii antibodies from different studies may be due to methodological differences, including the choice of commercial ELISA kits.

As the three hospitals were located in different settlement types (urban, peri-urban and rural), we assumed that the patient population at each hospital primarily comprised individuals from the surrounding communities. We, therefore, compared seroprevalence rates among participants from the urban, peri-urban and rural communities. In many instances, rural inhabitants regularly come into contact with potentially oocyst-contaminated soil as part of their normal farming activities. However, we found no significant association between seropositivity and settlement type, which agrees with findings from Munoz-Zanzi e.t. \(^\text{[19]}\), who also found no obvious differences in seroprevalence between rural villages and urban slums. In contrast, Kawashima et al. \(^\text{[20]}\) showed that seropositivity was significantly higher in rural communities compared to urban communities.

Acute infection by T. gondii in pregnancy can be detrimental to the foetus, resulting in stillbirth, chorioretinitis, hydrocephalus or even death \(^\text{[21]}\). The ability of almost all subclasses of IgG — with the notable exception of IgG2 — to cross the placenta provides some form of protection to the foetus \(^\text{[22]}\). The period of pregnancy in which the expectant mother becomes infected greatly influences the incidence and severity of the disease. Infection within the first trimester of pregnancy carries an approximately 6% risk of transmission to the foetus, while in the second and third trimesters, the risk of congenital transmission is about 33-47% and 60-81%, respectively \(^\text{[21]}\). Contrary to the gestational risk of transmission, the risk of developing clinical symptoms of the disease (congenital toxoplasmosis) is highest when maternal infection occurs within the first trimester and gradually reduces through the second and third trimesters \(^\text{[21]}\). Our study estimated an overall anti-T. gondii seroprevalence of 56.4% among pregnant women. This observation is lower than the 83.6% seroprevalence of T. gondii antibodies

### Table 1. Seroprevalence of anti-T gondii immunoglobulins.

| Tests          | Positive (n %) | Negative (n %) |
|---------------|----------------|----------------|
| IgG and/or IgM| 151 (50.3)     | 149 (49.7)     |
| IgM only      | 1 (0.3)        | 299 (99.7)     |
| IgG only      | 148 (49.3)     | 152 (50.7)     |

### Table 2. Seroprevalence of Toxoplasma gondii based on the type of community.

| Setting     | Total tested | Seropositive (n %) | \( \chi^2 \) | P-value |
|------------|--------------|--------------------|--------------|---------|
| Urban      | 140          | 66 (47.1)          | 1.457        | 0.482   |
| Peri-Urban | 81           | 45 (55.6)          |              |         |
| Rural      | 79           | 40 (50.6)          |              |         |

and 32 in the third trimester. The women ranged in age from 16 to 45 years, with a mean age of 28.6 years. Out of the 110 pregnant women, sixty-two were seropositive, indicating a general seroprevalence of 54.6%. Seropositivity for T. gondii was observed in 18 (48.6%) of the pregnant women in their first trimester, 22 (53.7%) in their second trimester and another 22 (68.8%) in their third trimester (Table 3).

To identify risk factors for T. gondii infection, the risk-ratio and chi-square tests were conducted to investigate the association between a few predisposing factors and seropositivity. Cat ownership (p=0.002) and contact with cat litter (p=0.003) were strongly associated with seropositivity for T. gondii. Handling of raw meat was also identified as a risk factor for exposure (p=0.05) (Table 3).

Certain demographic factors were also investigated to ascertain their association with seropositivity. The results revealed that the location, sex, education and employment status of participants were not predisposing factors to seropositivity. Age was, however, significantly associated with seropositivity (p-value = 0.000001306) (Table 4).

### Table 3. Effect of T. gondii infection on haematological parameters

T. gondii is capable of infecting virtually any nucleated cell, including erythroblasts. The effect of T. gondii on certain blood parameters was therefore investigated. A comparison between seropositive and seronegative individuals demonstrated significant differences in red blood cell counts (p=0.0380) and mean corpuscular volume (p=0.0007) (Table 5).

### Discussion

*Toxoplasma gondii* is an intracellular parasite with a wide geographical distribution and an extremely broad host range. Despite the high prevalence of *T. gondii* infections in many...
Table 3. Seroprevalence of anti-Toxoplasma gondii immunoglobulins in pregnant women.

| Gestation period   | Total tested | Positive (n %) | Negative (n%) | \( \chi^2 \) | RR (95% CI) | P-value |
|--------------------|--------------|----------------|---------------|-------------|------------|---------|
| First trimester    | 37           | 18 (48.6)      | 19 (51.4)     | 2.53        | 1.27 (0.94-1.72) | 0.1119 |
| Second trimester   | 41           | 22 (53.7)      | 19 (46.3)     |             |            |         |
| Third trimester    | 32           | 22 (68.8)      | 10 (31.3)     |             |            |         |

Table 4. Association between demographic factors and seropositivity of Toxoplasma gondii.

| Demographics | Total tested | Positive (n %) | Negative (n%) | \( \chi^2 \) | RR (95% CI) | P-value |
|--------------|--------------|----------------|---------------|-------------|------------|---------|
| Location     |              |                |               |             |            |         |
| Rural        | 81           | 45 (55.6)      | 36 (44.4)     | 1.07        | 1.12 (0.90-1.38) | 0.3     |
| Urban        | 140          | 66 (47.1)      | 74 (52.9)     |             |            |         |
| Peri-Urban   | 79           | 40 (50.6)      | 39 (49.4)     |             |            |         |
| Sex          |              |                |               |             |            |         |
| Male         | 58           | 23 (39.7)      | 35 (60.3)     | 3.28        | 0.65 (0.40-1.04) | 0.07    |
| Female       | 242          | 127 (52.5)     | 115 (47.5)    |             |            |         |
| Education    |              |                |               |             |            |         |
| None         | 40           | 23 (57.5)      | 17 (42.5)     | 0.95        | 0.96 (0.88-1.05) | 0.33    |
| Basic        | 56           | 27 (48.2)      | 29 (51.8)     |             |            |         |
| JHS          | 84           | 51 (60.7)      | 33 (39.3)     |             |            |         |
| SHS          | 86           | 38 (44.2)      | 48 (55.8)     |             |            |         |
| Tertiary     | 30           | 10 (33.3)      | 20 (66.7)     |             |            |         |
| Adult education | 4    | 2 (50.0)       | 2 (50.0)      |             |            |         |
| Employment   |              |                |               |             |            |         |
| Unemployed   | 145          | 75 (51.7)      | 70 (48.3)     | 2.69        | 1.24 (0.96-1.6) | 0.1     |
| Employed     | 152          | 75 (49.3)      | 77 (50.7)     |             |            |         |
| Retired      | 3            | 2 (66.7)       | 1 (33.3)      |             |            |         |
| Age          |              |                |               |             |            |         |
| 0–18         | 67           | 19 (25.4)      | 56 (73.6)     | 16.66       | 1.29 (1.14-1.46) | <0.001 |
| 19–44        | 175          | 92 (52.7)      | 73 (47.3)     |             |            |         |
| 45->         | 58           | 40 (67.0)      | 20 (33.5)     |             |            |         |

Table 5. Effect of Toxoplasma gondii infection on haematological parameters.

| Haematological parameters | Seropositive | Seronegative | P-value |
|---------------------------|--------------|--------------|---------|
|                           | Median (IQR) | Median (IQR) |         |
| White blood cells (10^3)/µL | 3.00 (2.10-4.30) | 3.35 (2.20-5.00) | 0.2886 |
| Lymphocytes (10^3)/µL     | 1.70 (1.20-2.70) | 1.90 (1.00-2.70) | 0.7428 |
| Neutrophils (10^3)/µL     | 0.80 (0.60-1.20) | 0.80 (0.60-1.30) | 0.3436 |
| Red blood cell (10^3)/µL  | 4.25 (3.78-4.64) | 4.40 (4.00-4.89) | 0.0380* |
| Haemoglobin (g/dl)        | 11.50 (10.25-12.40) | 11.45 (10.40-12.98) | 0.6012 |
| Mean corpuscular volume (/FL) | 99.40 (93.20-104.30) | 96.4 (88.65-101.20) | 0.0007*** |
reported by Sefah-Boakye et al., in the same region (Ashanti)\textsuperscript{11}. Furthermore, we observed a trend of increasing seroprevalence with gestational period, with similar observations made in other studies from Kumasi, Accra and Southwestern Ethiopia\textsuperscript{11,18,23}.

In disease control, prevention and elimination, identification of the risk factors for infection are key. Horizontal transmission of \textit{T. gondii} may occur via contact with cat litter, consumption of raw meat, eating of raw or undercooked vegetables, organ transplant and blood transfusion. We identified cat ownership, contact with cat litter and handling of raw meat as important risk factors for infection among our study population. Since \textit{T. gondii} undergoes sexual reproduction in the gastrointestinal tract of domestic felines, individuals who owned cats or had regular contact with cat litter had 76\% and 66\% greater risk of \textit{T. gondii} infection respectively compared to individuals who neither owned cats nor handled cat litter. Furthermore, since \textit{T. gondii} is capable of encysting in the tissues of infected animals, people who regularly handle raw meat are also at greater risk of infection\textsuperscript{24}. We also investigated the association between certain demographic parameters and seropositivity. Consistent with findings from other studies in Ghana and Iran, seropositivity increased with age\textsuperscript{11,15,25,27}. This is due to the fact that infection has strongly been associated with contact with the soil where the oocyst prevails for years. The longer an individual survives, the greater the likelihood of coming into contact with contaminated soil hence the increase in seroprevalence with age.

\textit{T. gondii} is capable of invading any nucleated cell, including immature red blood cells\textsuperscript{26}. As a mechanism of survival, intra-cellular parasites need to exit and re-infect other cells. The mechanism by which \textit{T. gondii} exits infected cells is still not clearly understood. Some \textit{in vitro} studies have demonstrated that \textit{T. gondii} exits the cell by exerting tension on the host cell membrane, often causing the cell to rupture\textsuperscript{27}. Other studies have suggested that the parasite exits the host cell by disrupting the cytoskeleton in a manner similar to \textit{Plasmodium}, resulting in cell lysis\textsuperscript{28}. We found significant differences in the RBC counts and mean corpuscular volumes of \textit{T. gondii} seropositive and seronegative individuals. While the exact molecular processes driving these differences are still unclear, there is increasing evidence that latent toxoplasmosis may cause chronic low-grade inflammation\textsuperscript{29}, which may, in turn, lead to anaemia\textsuperscript{30}.

\section*{Conclusion}

The seroprevalence of \textit{T. gondii} infection was high among our study population, including among pregnant participants. In addition, cat ownership, contact with cat litter and age were identified as major risk factors for infection. Furthermore, additional research is needed to fully clarify the links between latent toxoplasmosis and anaemia. In conclusion, we recommend that testing for infection by the parasite be included in routine screening of pregnant women seeking antenatal care.

\section*{Data availability}

\subsection*{Underlying data}

Harvard Dataverse: Replication Data for: Seroprevalence, Risk Factors and Impact of Toxoplasma gondii Infection on Haematological Parameters in the Ashanti Region of Ghana: A Cross-Sectional Study, https://doi.org/10.7910/DVN/HXJONT\textsuperscript{31}.

This project contains the following underlying data:
- Data includes, demography of study participants, risk factors, IgG response and measurement of haematological parameters
- Data dictionary

\subsection*{Extended data}

Harvard Dataverse: Replication Data for: Seroprevalence, Risk Factors and Impact of Toxoplasma gondii Infection on Haematological Parameters in the Ashanti Region of Ghana: A Cross-Sectional Study, https://doi.org/10.7910/DVN/HXJONT\textsuperscript{31}.

This project contains the following extended data:
- Questionnaire used in the present study.

Data are available under the terms of the Creative Commons Zero “No rights reserved” data waiver (CC0 1.0 Public domain dedication).

\section*{Acknowledgements}

We acknowledge the Kuntanase, Agona and Kumasi South Hospitals for allowing us the space in their laboratories. We are also grateful to all participants for their voluntary participation in this study. The KNUST Graduate Assistantship Program has been of immense support to Samuel Kekeli Agordzo and Abdul-Hakim Mutala.

\section*{References}

1. Dubey JP, Jones JL: \textit{Toxoplasma gondii} infection in humans and animals in the United States. Int J Parasitol. 2006; 36(11): 1257–78.\textsuperscript{32} PubMed Abstract | Publisher Full Text
2. Montoya JG, Liesenfeld O: \textit{Toxoplasmosis}. Lancet. 2004; 363(9425): 1965–76.\textsuperscript{33} PubMed Abstract | Publisher Full Text
3. Flegr J: Effects of \textit{Toxoplasma} on human behavior. Schizophr Bull. 2007; 33(3): 757–69.\textsuperscript{34} PubMed Abstract | Publisher Full Text | Free Full Text
4. Montoya JG: Laboratory diagnosis of \textit{Toxoplasma gondii} infection and toxoplasmosis. J Infect Dis. 2002; 185(Suppl 1): S73–82.\textsuperscript{35} PubMed Abstract | Publisher Full Text
5. Robbins JR, Zeldovich VB, Poukchanski A, et al.: Tissue barriers of the human placenta to infection with \textit{Toxoplasma gondii}. Infect Immun. 2012; 80(1): 418–28.\textsuperscript{36} PubMed Abstract | Publisher Full Text | Free Full Text
6. Galvan-Ramirez Mde L, Troyo R, Roman S, et al.: A systematic review and meta-analysis of \textit{Toxoplasma gondii} infection among the Mexican population. Parasit Vectors. 2012; 5(1): 271.\textsuperscript{37} PubMed Abstract | Publisher Full Text | Free Full Text

Page 7 of 12
7. Galván-Ramírez ML, Covarrubias X, Rodríguez R, et al.: Toxoplasma gondii antibodies in Mexican blood donors. Transfusion 2005; 45(2): 281–2. 
PubMed Abstract | Publisher Full Text

8. Muralikrishna P, Sunil B, Savita RK, et al.: Toxoplasmosis-The Public Health significance. International Journal of Science, Environment and Technology. 2017; 6(5): 2752–2758. 
Reference Source

9. Yohannes T, Debake S, Zemene E: Latent Toxoplasma gondii Infection and Associated Risk Factors among HIV-Infected Individuals at Arba Minch Hospital, South Ethiopia. AIDS Res Treat. 2014: 652941. 
PubMed Abstract | Publisher Full Text | Free Full Text

10. Bamba S, Cissé M, Sangaré I, et al.: Seroprevalence and risk factors of Toxoplasma gondii infection in pregnant women from Bobo Dioulasso, Burkina Faso. BMC Infect Dis. 2017; 17(1): 482. 
PubMed Abstract | Publisher Full Text | Free Full Text

11. Sethe-Baileye J, Grimpong EH, Dompere A, et al.: Seroprevalence of Toxoplasma gondii infection among Pregnant Women in the Ashanti Region of Ghana. Int J Microbiol Adv Immunol. 2016; 4(1): 70–74. 
Publisher Full Text

12. Ayi I, Edu SA, Apea-Kubi KA, et al.: Sero-epidemiology of toxoplasmosis amongst pregnant women in the greater accra region of gana. Ghana Med J. 2009; 43(3): 107–14. 
PubMed Abstract | Publisher Full Text | Free Full Text

13. Abu EK, Boampong JN, Ayi I, et al.: Infection risk factors associated with seropositivity for Toxoplasma gondii in a population-based study in the Central Region, Ghana. Epidemiol Infect. 2015; 143(9): 1904–12. 
PubMed Abstract | Publisher Full Text

14. Abu EK, Boampong JN, Amoaebeng JK, et al.: Epidemiology of Ocular Toxoplasmosis in Three Community Surveys in the Central Region of Ghana, West Africa. Ophthalmic Epidemiol. 2016; 23(1): 14–9. 
PubMed Abstract | Publisher Full Text

15. Kwofie KD, Ghanaah A, Osei JH, et al.: Indication of Risk of Mother-to-Child Toxoplasma gondii Transmission in the Greater Accra Region of Ghana. Matern Child Health J. 2016; 20(12): 2581–2588. 
PubMed Abstract | Publisher Full Text

16. Dinhru V, Gajurel K, Pomares C, et al.: Significance of a Positive Toxoplasma Immunoglobulin M Test Results in the United States. J Clin Microbiol. 2015; 53(11): 3601–5. 
PubMed Abstract | Publisher Full Text | Free Full Text

17. Ayeh-Kumi PF, Opoku AG, Kwaile-Nuako G, et al.: Sero-prevalence of toxoplasmosis among patients visiting the Korle-Bu Teaching Hospital, Accra, Ghana. Afr J Infect Dis. 2010; 1(3): 147–150. 
Reference Source

18. Zemene E, Yewhalaw D, Aberra S, et al.: Seroprevalence of Toxoplasma gondii and associated risk factors among pregnant women in Jimma town, Southwestern Ethiopia. BMC Infect Dis. 2012; 12(1): 337. 
PubMed Abstract | Publisher Full Text | Free Full Text

19. Munoz-Zanzani C, Campbell C, Berg S: Seroepidemiology of toxoplasmosis in rural and urban communities from Los Rios Region, Chile. Infect Ecol Epidemiol. 2016; 6(1): 30597. 
PubMed Abstract | Publisher Full Text | Free Full Text

20. Kawasaki T, Khin-Sane-Win, Kawabata M, et al.: Prevalence of antibodies to Toxoplasma gondii among urban and rural residents in the Philippines. Southeast Asian J Trop Med Public Health. 2000; 31(4): 742–6. 
PubMed Abstract

21. Dunn D, Wallon M, Peyron F, et al.: Mother-to-child transmission of toxoplasmosis: risk estimates for clinical counselling. Lancet. 1999; 353(9167): 1829–33. 
PubMed Abstract | Publisher Full Text

22. Palmeira P, Quinello, C, Silveira-Lessa AL, et al.: IgG placental transfer in healthy and pathological pregnancies. Clin Dev Immunol. 2012: 985646. 
PubMed Abstract | Publisher Full Text | Free Full Text

23. Ayi I, Sowah AO, Blay EA, et al.: Toxoplasma gondii infections among pregnant women, children and HIV-seropositive persons in Accra, Ghana. Trop Med Health. 2016; 44(1): 17. 
PubMed Abstract | Publisher Full Text | Free Full Text

24. Swai ES, Schoonman L: Seroprevalence of Toxoplasma gondii infection amongst residents of Tanga district in north-east Tanzania. Tanzan J Health Res. 2009; 11(4): 205–9. 
PubMed Abstract | Publisher Full Text

25. Jafari R, Sadaghian M, Safari M: Seroprevalence of Toxoplasma gondii infection and related risk factors in Tabriz City, Iran. 2006. J Res Health Sci. 2012; 12(2): 119–121. 
PubMed Abstract

26. Joiner KA, Dubremetz JF: Toxoplasma gondii: a protozoan for the nineties. Infect Immun. 1993; 61(4): 1169–72. 
PubMed Abstract | Free Full Text

27. Lund E, Lycke E, Sournander P: A cinematographic study of Toxoplasma gondii in cell cultures. Br J Exp Pathol. 1961; 42(4): 357–62. 
PubMed Abstract | Free Full Text

28. Lavine MD, Arrazabala G: Exit from host cells by the pathogenic parasite Toxoplasma gondii does not require motility. Eukaryot Cell. 2008; 7(1): 131–140. 
PubMed Abstract | Publisher Full Text | Free Full Text

29. Bay-Richter C, Petersen E, Liebenberg N, et al.: Latent toxoplasmosis aggravates anxiety- and depressive-like behaviour and suggest a role of gene-environment interactions in the behavioural response to the parasite. Behav Brain Res. 2019; 364: 133–139. 
PubMed Abstract | Publisher Full Text

30. Wang Z, Zhang DX, Zhao G: Infection-stimulated anemia results primarily from interferon gamma-dependent, signal transducer and activator of transcription 1-independent red cell loss. Chin Med J (Engl). 2015; 128(7): 948–55. 
PubMed Abstract | Publisher Full Text | Free Full Text

31. Badu K: “Replication Data for: Sero-prevalence, Risk Factors and Impact of Toxoplasma gondii Infections on Haematological Parameters in the Ashanti Region of Ghana: A Cross-Sectional Study”. Harvard Dataverse, V2, UNF: 6: m?epmGQTGFSAO5ayw0Qwet-=fileUNF. 2019. http://www.doi.org/10.7910/DVN/NOJONT
Open Peer Review

Current Peer Review Status: ? ?

Version 1

Reviewer Report 04 May 2020

https://doi.org/10.21956/aasopenres.14110.r27426

© 2020 Blay E. This is an open access peer review report distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Emmanuel Awusah Blay
Department of Pathology and Microbiology, University of Nebraska Medical Center, Omaha, NE, USA

The manuscript titled “Seroprevalence, risk factors and impact of Toxoplasma gondii infection on haematological parameters in the Ashanti region of Ghana: a cross sectional study” by Agordzo et al., presented evidence on the current serological prevalence of T. gondii and association with selected haematological parameters.

Though the seroprevalence of T. gondii has been widely published in different risk groups in Ghana, this research adds to the body of existing literature and knowledge and presents evidence on the possible association between T. gondii infections and haematological parameters. Additionally, this research provides additional evidence on possible regional variations in seroprevalence in Ghana, as most of the existing research has been conducted on the coastal regions of the country. Different geographical regions may possibly present additional evidence of variations in infections and associated risk patterns of T. gondii.

Comments:

1. In providing the data for the seroprevalence for the study in Table 1, the authors provided the general seroprevalence from the study. However, considering that the data was obtained from different populations i.e. Pregnant women, infants, critically ill persons and general hospital attendees, it will be more informative to show which of these groups showed higher prevalence in IgG only, IgM only and both IgG and IgM. Such details would provide details as to which groups are most susceptible to T. gondii infections.

2. In Table 2, do the authors group seropositive individuals together in the different geographical settings. However, a breakdown on the type of seropositivity i.e. IgG only, IgM only and both IgG and IgM may be more beneficial to the readers for a critical understanding as to which geographical setting is recording more acute cases and which is showing more in exposure to T. gondii

3. In Table 3, the authors provide details of seropositivity in pregnant women. In total, 110 pregnant women comprising of 37 in first trimester, 41 in second trimester and 32 in third trimester are
presented. Although this information may have a degree of importance, the gestation period alone is scanty to make any meaningful deductions. It will be more beneficial if the authors can add the gravidity and parity status of the pregnant women to show whether they have had previous live births or abortions. This additional information coupled with the specific seropositivity i.e. IgG only, IgM only and both IgG and IgM will provide information to discuss the significance of past or acute infections have on seropositivity.

4. In Table 5, the authors provide data on the haematological parameters analyzed and the seropositivity of the participants. Although this seems to be the crux and novelty of the paper, the manner the data has been presented provides the reader with less clarity. Because the participants were drawn from different population groups, it will be more prudent to provide a breakdown of the haematological data in sub-populations. This is so because the haematological parameters of pregnant women will be significantly different from children and also different from general hospital attendees. Thus, whiles providing a composite data is good, it does little to clearly draw out the differences between the various study groups. Additionally, I will suggest that the seopositivity data is clearly distinguished between IgG only, IgM only and both IgG and IgM. This provides greater clarity and brings out the different nuances in the data set.

5. The authors provide no data on the risk factors in this manuscript although it is an integral part in their title. I suggest they add the data or modify their title to reflect the data that is presented.

Minor Comments:
1. In paragraph 4, of sub-title “Seroprevalence of T. gondii” the authors state discuss results pertaining to risk factors and incorrectly attribute it to Table 3.

2. On the haematological data discussion, the authors should discuss the differences in mean corpuscular volumes. I am highly convinced that the differences observed could be due to large percentage of the participants being pregnant women.

3. A major limitation to this study is the use of a qualitative commercial kit for determining seropositivity. An additional method such as an avidity ELISA or molecular identification of specific genes of T. gondii such as the Dense granules (GRA) or surface antigens (SAG) may have provided more information whether the infections were latent or acute. I suggest the authors add the limitations of this study into their conclusions.
Are the conclusions drawn adequately supported by the results?
Partly

**Competing Interests:** No competing interests were disclosed.

**Reviewer Expertise:** Molecular Parasitology and Microbial genetics

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.

Reviewer Report 14 January 2020
https://doi.org/10.21956/aasopenres.14110.r27328

© 2020 Jafari R. This is an open access peer review report distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Rasool Jafari
Department of Medical Parasitology, School of Medicine, Urmia University of Medical Sciences, Urmia, Iran

I studied the manuscript with interest. The manuscript studied the prevalence and related risk factors of infection with *T. gondii* in Ghana. There are some issues that need to be addressed as below.

1. There is extensive research on the seroprevalence of toxoplasmosis, and most of the risk factors and epidemiological aspects of the infection are relatively known. The findings of the manuscript are not so novel, mostly known; however it has regional importance in the studied area of Ghana.

2. The method for determining anti-*Toxoplasma* IgG and IgM is not quantitative, and I do not suggest rapid tests for research. If the author may have used ELISA, ECLA or CLA, data may have been more accurate and they may analyze the quantitative concentration of antibodies among risk factors.

3. For studying the hematological parameters, authors should be aware of the fact that the study population must be similar in seropositive and seronegative individuals. In women, anemia is prevalent and the authors could not compare the parameters between men and women; they may analyze it separately among both sexes. Pregnancy and nutritional status during pregnancy (food, vitamin and iron supplements) are other issues affecting blood parameters, that are not addressed, which can bias the results.

4. The authors could discuss other risk factors reported in the literature, such as having contact with soil and food habits such as eating raw vegetables, etc.

5. Why are different mean corpuscular volumes among seronegative and seropositive cases? The authors should discuss this issue and I think it may reflect the fact that the majority of the studied group were female and pregnant.
6. The language is relatively good; however, few misspellings and punctuation-errors need to be corrected.

Is the work clearly and accurately presented and does it cite the current literature?
Partly

Is the study design appropriate and is the work technically sound?
Partly

Are sufficient details of methods and analysis provided to allow replication by others?
Yes

If applicable, is the statistical analysis and its interpretation appropriate?
Yes

Are all the source data underlying the results available to ensure full reproducibility?
Yes

Are the conclusions drawn adequately supported by the results?
Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Molecular and serological epidemiology of parasitic diseases (e.g., toxoplasmosis, cryptosporidiosis and hydatid disease)

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.