Socio-economical Risk Factors Associated with *Toxoplasma gondii* Infection in Rural Women at Childbearing Age in Sudan

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Abstract

The main objective of this study was to determine the risk factors involved in toxoplasmosis transmission. The prevalence rate of 253 plasma samples using Complement Fixation Test (CFT) was 72.3%. The association of the risk factors to the disease discussed in this study. The results showed that the prevalence rate had increased with age (p<0.001). Infection rate was higher in EL Massoudia village (p<0.001). Uneducated women were more exposed to infection (p<0.001). Married women were more positive (p<0.001). Women unemployed were more exposed (p<0.001). The present study indicates that there is a considerable rate of Toxoplasma infection among rural women and support the concern that rural women in Sudan may be vulnerable to that infection.

Keywords: Risk factors, *Toxoplasma gondii*; Rural women; Sudan

Introduction

*Toxoplasma gondii* is widely spread throughout the world. The prevalence estimated for human population varies greatly among different countries, among different geographical areas within one country, and among different ethnic groups living in the same area. Seroprevalence of *Toxoplasma gondii* infection in women at childbearing age is found to be between 4%-100% [1]. In recent years, the importance of congenital toxoplasmosis has been increasingly recognized. Further, patients with a variety of neoplastic diseases as well as patients receiving immunosuppressive therapy are at risk of reactivation of *Toxoplasma gondii* infection. Furthermore, the incidence of toxoplasmosis and especially toxoplasmic encephalitis has risen dramatically with the increasing population of patients with AIDS [2].

Environmental conditions, differences in the type of food consumed, animal species used in food industry, and the number of cats are examples of factors that may influence the spread of the parasite. Water-borne transmission of *Toxoplasma gondii* has earlier been considered uncommon, but recently human outbreaks connected to water reservoirs have been reported [3,4].

There is little, if any, danger of *Toxoplasma gondii* infection by drinking cow’s milk which, in any case, is generally pasteurized or even boiled, but infection has followed drinking unboiled goat's milk. Raw hens, eggs, although an important source of Salmonella infection, are extremely unlikely to harbour *Toxoplasma gondii*. Transmission by sexual activity including kissing is probably rare and epidemiologically unimportant.

Blood transfusions and organ transplants, transplantation being the more important epidemiologically; this is a recent development. In people undergoing transplantation toxoplasmosis may arise either from implantation of an organ or bone marrow from an infected donor into a non-immune immunocompromised recipient or from induction of disease in an immunocompromised latently infected recipient. The tissue cysts in the transplanted tissue or in the latently infected person are probably the source of the infection. In both cases the cytotoxic and immunosuppressive therapy given to the recipient is the cause of the induction of the active infection and the disease [5,6].

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The complement fixation test is the basic method in the diagnosis of toxoplasmosis [7]. Due to the latter and due to the availability of the antigen it has become the "golden standard" in the diagnosis of toxoplasmosis despite the fact that this attribute is usually associated with Sabin and Feldman dye test [8,9]. The results of both reactions correlate well and their standards and reproducibility are comparable. In addition to other facts the diagnostic advantage resides in the possibility of quantification of the detected levels of antibodies [7]. The significance of the CFT for the diagnosis and evaluation of *Toxoplasma* infection is sufficiently known particularly in Europe [10].

From the aspect of the determination of *Toxoplasma* infection from a single serologic examination, the CFT test yields more relevant prognostic values when compared with IgG antibodies when the same requirement is involved. Specific IgG antibodies detected by ELISA techniques are a reliable substitution of quantitative results that can be assessed by CFT. On the other hand despite the fact that the dynamics of CFT antibodies are more significantly associated with the course of the disease, the assessment of the phase of infection must be supported also by parallel examination of IgM [11] or IgA antibodies and in indicated cases also by that of the avidity of IgG antibodies [12].

The overall aim of this study was to determine the prevalence of toxoplasmosis in childbearing women in North Geizera State and to study the risk factors contribute to the infection.

Materials and Methods

Study area and Population

The study was performed in two villages; EL Massoudia and EL
Nuba lay in the north of EL Geizera State (middle Sudan) located near Blue Nile, these villages belonging to EL Kamleen province about 50 kilometers south of the capital Khartoum. Most of population in these villages belongs to the same ethnic group. People in these areas presented low socio-economic status, thus people are farmers, animal breeders, or workers particularly after a big industry city was established near this area. Women live in simple life way and although most of them were not working but they lend a hand to improve the economic situation by bringing the water from river, or bringing the wood for cooking or take care of animals including contribution in birth of animals or participate in farms and agricultural process.

Study design
The study was cross-sectional study for toxoplasmosis in women at the child bearing age. The sample size was calculated as 255 on a prevalence of 20% obtained from first 10 samples collected, d = 0.05 at a confidence level of 95%. A total of 5% of the sample population was added to the sample size. Two samples were missed due to the lack of plasma.

Data collection
Consent form and questionnaire were filled for each individual. Consent form was signed and fingerprinted by each one after agreed participation in the study. Data were collected after convenient interview. The questionnaire was performed. There were questions eliciting socio-demographic data including age, education (illiteracy, primary school, secondary school, university or postgraduate), occupation, residency and related risk factors.

Samples collection
The blood samples were collected under direct medical supervision by medial venipuncture using 5 ml syringe into heparinized tubes, plasma was obtained by centrifugation of the blood at 5000 rpm for 10 minutes. Plasma was kept in different labeled cryo tubes in -20°C till used. Plasma samples were sent in dry ice to National Reference Laboratory for Toxoplasmosis, Prague, Czech Republic where more investigations were done.

Detection of Toxoplasma gondii Antibodies
Complement Fixation Test (CFT) (Test-line) was used for detection of specific antibodies to Toxoplasma gondii antigens in human plasma. It can give indication of differentiation of acute Toxoplasmosis.

Data analysis
Statistical evaluation was done by the data obtained into the personal computer (PC) using two different programs:

1. Statistical Package for Social Science (SPSS) version 13.0 (SPSS Inc. Chicago, IL. USA) was used to calculate the descriptive statistics to obtain specified statistics on the variables for numerical (mean and median) and categorical (frequency or percentage), also we used this package to calculate the prevalence rate of the disease with different screening tests used. In addition, Chi2 test was used for ordinal variables to find the significant difference between the infection and risk factors.

2. Statistical analysis was performed by statistical software Stata, version 9.2 (Stata Corp LP. College Station, TX). We used Fisher’s exact test (when cells values were less than 5), for comparison of frequencies among groups. Adjusted odd ratio (OR) and Relative Risk (RR) and 95% Confidence Interval (CI) were calculated by multivariate analysis using multiple, unconditional logistic regression. A p-value less than 0.05 were considered statistically significant.

Results
Prevalence of Toxoplasma gondii using CFT
A total of 253 plasma samples were tested by complement fixation test to detect antibodies against Toxoplasma gondii in those women. The result showed 183 (72.3%) of women from the two villages had antibodies against Toxoplasma gondii. The intensity of reaction was found to be 15 (8.2%), 52 (28.4%), 72 (39.3%), 23 (12.6%), 9 (4.9%), 10 (5.5%), 1 (0.6%), 1 (0.6%), and 0 (0.0%) at the dilution of 1:4, 1:8, 1:16, 1:32, 1:64, 1:128, 1:256, 1:512, and 1:1024, respectively. The prevalence rate of Toxoplasma gondii infection in women in EL Nuba village using complement fixation test was found to be 80 (59.3%). The intensity of infection was found to be 5 (6.3%), 27 (33.8%), 30 (37.7%), 11 (13.8%), 2 (2.5%), 5 (6.3%), and 0 (0.0%) at the dilution of 1:4, 1:8, 1:16, 1:32, 1:64, 1:128, and 1:256, respectively as shown in (Table 1). In EL Massoudia village the prevalence rate using CFT was 103 (87.3%). The intensity of infection was found to be 10 (9.7%), 25 (24.3%), 42 (40.8%), 12 (11.7%), 7 (3.8%), 5 (4.9%), 1 (1.0%), 1 (1.0%), and 0 (0.0%) at the dilution of 1:4, 1:8, 1:16, 1:32, 1:64, 1:128, 1:256, 1:512, and 1:1024, respectively as elucidated in table 1.

Socio-demographic and economic risk factors
Location: General socio-demographic characteristics of 255 women have been studied. The interview of the suspected women for infection with toxoplasmosis was done in the two villages EL Nuba and EL Massoudia in North Geizera. Women who participated in this study belong to eight blocks according to administrative division; five of them belong to EL Nuba village, most of them were in Kamir block (39.7%) whereas three blocks belong to EL Massoudia village, most of them were in Elshargi block (61%). The majority of women were born and resident in these villages which belong to rural area.

The prevalence rate among the villages was highly significant (p<0.001) and women who live in EL Massoudia village were under the risk of infection by Toxoplasma gondii. The risk ratio was 1.4 at 95%, CI: 1.2-1.7 these findings were confirmed by calculating the OR= 4.4 at 95% CI: 2.3-8.4 (Table 2).

Age: The mean age of women was 32.9 ± 11 years (median: 35 years). The mean age of women in EL Nuba village was 32.4 ± 13 (median: 32 years) while the mean age of women in AL Massoudia was 33.6 ± 9 (median: 35 years).

Prevalence rate increased with age (p<0.001) as women who were...
more than 45 years were more infected than women in age group 35-44 and youngest women were less infected than oldest. This result was confirmed by calculating the odd ratio which also increased with age (Table 2). Comparing the age group and location with infection, we found significant difference in EL Massoudia village.

### Education

About (90.2%) of women who participated in this study were educated, most of them (44.6%) finished the secondary school, (33.8%) completed their primary or intermediate schools, and (21.6%) graduated or still studying at the university level and merely (9.8%) of them were uneducated. The women educational status in EL Nuba village was 56 (40.3%), 38 (27.3%), 33 (23.7%), and 12 (8.6%) in secondary, primary and intermediate, graduated, and uneducated, respectively. Similar situation of women educational status in EL Massoudia villages; most of women finished their secondary school 47 (40.2%) while 40 (34.2%) had primary or intermediate studies and 17 (14.5%) at the university level or graduated and 13 (11.1%) are uneducated.

Relationship between infection with *Toxoplasma gondii* and the education show significant (p<0.001) (Table 2), women who were uneducated were more expected to infection than women who learn at any level. This result was confirmed by calculating OR 4.0 at 95% CI 0.47-33.7 (Table 2).

### Married status

The married status of women in this study showed 157 (61.3%) were married, 82 (32.0%) were single, and 17 (6.6%) were divorced or widows. In EL Nuba village 72 (51.8%) of women were married, in EL Massoudia village 85 (72.6%) were married. Single women were more in EL Nuba 60 (43.2%) than in AL Massoudia 22 (18.8%). The group of divorced and widows were 7 (5%) and 10 (8.5%), respectively.

Married women were found to be more positive to *Toxoplasma gondii* infection (p<0.001) (Table 2). The OR was 2.7 at 95% CI: 0.35-22.3 (Table 2).

### Family size

The overall number of individuals in the family in this study group ranged between 1 and 14, the mean was 6.2 ± 2.7 and the median was 6. In EL Nuba village the range was 1-13 and the mean was 6.1 ± 2.7 while the median was 6. In EL Massoudia village the range was 2-14 and the mean was 6.3 ± 2.7 and the median was also 6.

The relationship between mother infection with *Toxoplasma gondii* and the number of her children was observed (p=0.007) (Table 2).

### Occupation

The whole study group of women in the villages were housewives 191 (74.6%), students were 48 (18.8%) and few of them were working as teachers 9 (3.5%), workers 5 (2.0%), and 3 (1.2%) were employees.

In EL Nuba village 93 (66.9%) were housewives, 32 (23.0%) were students, 9 (6.5%) were teachers, 3 (2.2%) were workers, and 2 (1.4%) were employees. In EL Massoudia village 98 (83.8%) were housewives, 16 (13.7%) were students, workers 2 (1.7%), and 1 (0.9%) was employee.

Infection with *Toxoplasma gondii* and occupation were compared. Although the result showed significant difference between housewives and students (p<0.001) (Table 2), this result was not valid when calculated the odd ratio 0.2 at 95% CI: 0.07-0.3 (Table 2).

### Husband job

The relationship between woman infected by *Toxoplasma gondii* and the job of her husband was found. Women whose husbands were workers, employees or farmers were more infected than others (p<0.001)

### Income

The minimum income per month was 120 $ and the maximum was 240 $. About 95.7% of those women had no permanent income and they were depending on their husbands’ income if they were married or family if they were students. Most of their husbands are workers (37.1%) in the industrial city established near these villages; some of them were employees (11.7%) and few of them were drivers (4.3%) or farmers (3.5%).

People were very similar economically in their income and the variation was not detected. The results showed no relationship between income and infection with *Toxoplasma gondii* (p>0.06).

### Discussion

Using complement fixation test supported this study. CFT is considered as basic method in the diagnosis of toxoplasmosis.

<table>
<thead>
<tr>
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<th>EL Nuba</th>
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<th>No. of Positive</th>
<th>P-value</th>
<th>Adjusted OR</th>
<th>95% CI</th>
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</table>

Table 2: Association between Demographic as Risk Factors and Infection with *Toxoplasma gondii*.
Considering its high specificity it gives reproducible results and is rarely used in routine diagnosis of toxoplasmosis due to the risk of using life tachyzoites [7]. The prevalence obtained by CFT in this study was 72.3%. It was higher than with other studies done in women in Czech Republic where this test is used widely [13,14]. Differences in life style, food habitual, geographic and climate are the main purposes of this difference but the health education programme, education, awareness, and routine and compulsory investigation of infection, are the main reasons in reduction the prevalence rate to less than 30%.

The present study has revealed that the prevalence rate of toxoplasmosis vary remarkable in childbearing age in relation to locality. The risk ratio was high in EL Massoudia village and the odd ratio confirmed that women in EL Massoudia village are more than four times to be exposed to infection than women in EL Nuba village. However, Seroprevalence estimates for human population vary greatly among different countries, among different geographical areas within one country, and among different ethnic groups living in the same area [1]. For example, seroprevalences in Central European countries, such as Austria, Belgium, France, Germany, and Switzerland have been estimated to range between 37% and 58% in women of childbearing age comparable with seroprevalences have been observed in similar populations in Croatia, Poland, Slovenia, Australia, and Northern Africa [1].

In this study, we found that the prevalence increased steadily with age and this is presumably due to accumulated opportunities for exposure. The finding is symphonizing with finding in Europe [15] and with study done in Sudan [16] also, with recent study done in Mexico [17] and in Brazil [18] and in contrast with finding in women in Tanzania where they found the infection was concentrated in age group of women between 12-25 years of age [19].

The relationship between education and infection was recorded in this study, thus women uneducated were more infected than educated women. In the United States the risk of toxoplasmosis is associated with low level of education [20]. The OR confirmed that uneducated women have chance to be infected four times more than educated women. Although, 90% of women participated in this study are women have chance to be infected four times more than educated women. In the United States the risk of toxoplasmosis is associated with age and this is presumably due to accumulated opportunities for exposure. The finding is symphonizing with finding in Europe [15] and with study done in Sudan [16] also, with recent study done in Mexico [17] and in Brazil [18] and in contrast with finding in women in Tanzania where they found the infection was concentrated in age group of women between 12-25 years of age [19].

Married women are more suspected to infection by *Toxoplasma gondii* than single women. OR confirmed that married women were two times more infected than women unmarried, this might be due to that married women are matured than single and more suspected to sources of infection because they did all work in their houses including cleaning and food preparing.

Occupation plays a key role in the transmission of the disease when occupation is related to direct contact with meat, soil, and animals. The majority of women in this study were housewives and few of them are working in jobs not related to soil or meat but at last they are housewives. The infection in housewives may be attributed to meat processing while infection in students may be attributed to the contact with soil during houses cleaning as this is main job of girl students in Sudan particularly, in rural areas. These findings are similar to the finding recorded in camels, herders in Sudan [21] and in workers on swine farms in Illinois [22]. Although the significance was higher but the OR did not confirm the responsibility of cleaning and preparing meat.

Infection was increased by increasing the number of children, this is often due to mother age because a woman with many children was older than others and she has a big chance of exposure to the parasite. Family size in Sudan means that people live together not only father, mother, and their sons and daughters but also other relatives. Family size did not significantly confirm increasing of infection but age can do. These findings are in contrast to the findings in Brazil [23] this contrast may be due to poor living conditions and poor hygiene. The relationship between infection in women and the job of their husbands was observed. The or confirms that women who their husbands were employee were more infected than others. This may be due to the life style of women. There was no variation due to income because people belonged to poor and low socio-economic level.

References


