

Toxoplasmosis Sero- Prevalence among Aborted Women by Different Techniques in Atbara Area- River Nile State, Sudan

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Abstract

This cross-sectional study was conducted in River Nile state (Atbara locality) during the period from February to May, 2017. The aim of this study was to determine the prevalence of toxoplasmosis among aborted women. One hundred fifty two aborted women were enrolled in this study with the age ranged between 20-49 years old. Blood samples were taken from all participants. All samples were examined to detect *Toxoplasma gondii* IgG and IgM antibodies by using LATEX, ICT and ELISA techniques. The study showed that, the overall prevalence of *Toxoplasma* parasite were 35.5%, 33.6% 22.4% by ELISA, LATEX agglutination and ICT respectively. The study showed that, the prevalence of *Toxoplasma* infections was highest (54.6 %) among the age group (41-49) years old, followed by 31-40 age group (33.5%) and the lowest (11.8%) among age group 20-30 years old. Assuming ELISA technique as the gold standard method, the sensitivity and specificity of the LATEX technique were 94 % and 100% while for ICT technique were 64 % and 100 %. The study concluded that, the toxoplasmosis was more prevalent among pregnant women in Atbara locality in River Nile state.

Keywords: toxoplasmosis; Latex; Elisa; Ict; Atbara

Abbreviations: DT: Dye Test; ELISA: Enzyme-Linked Immunosorbent Assays; MAT: Modified Agglutination Test; ISAGA: Immunosorbent Agglutination Assay; IFAT: Indirect Fluorescent Antibody Test; IHAT: Indirect Hemagglutination Assays Test; TSP: Toxoplasma Serological Profile; ICT: Immuno chromatography test.

Introduction

Toxoplasma gondii is a protozoan parasite of the family Coccidia. It is intracellular obligatory protozoan

with a heterogeneous life cycle in vertebrates and humans [1-3]. Domestic cats and other felidae are the definitive hosts of *T. gondii* while intermediate hosts are other non-feline and human [4]. Infection of humans with *T. gondii* acquired by ingestion of under cooked meat of animals that had tissue cyst, or environmental contaminated by infected sample via food or water, also congenital and blood transfusion or organ transplantation from individuals harboring tissue cyst [5-7]. Toxoplasmosis is usually an asymptomatic disease, but often takes a severe course in immune compromised hosts [8,9]. Many type of

disease may occur as congenital, acquired and reactivated toxoplasmosis [10]. The congenital toxoplasmosis has a serious complication and is acquired congenital from infected mother to her fetus through placenta. It may lead to severe damage or abortion [11,12].

Diagnosis of toxoplasmosis through serological tests, such as dye test (DT), enzyme-linked immunosorbent assays (ELISA), modified agglutination test (MAT), immunosorbent agglutination assay (ISAGA), indirect fluorescent antibody test (IFAT) and indirect hemagglutination assays test (IHAT), to detect different antigens or antibody classes [13-16]. The toxoplasma serological profile (TSP) is very important to differentiate between past and early infection, the presence of IgG antibodies does not give timing of infection, IgM detectable about one week after infection, IgA antibodies may persist for several months and produced earlier than IgM, therefore, considered as markers of acute infection, IgE provides a greater indication of current infection due to their shorter period [16,17]. Several a studies from different part of world enrolled to detect difference between IgG and IgM antibodies levels against *T.gondii* in abortions compared to normal pregnancies by different techniques [18-20].

Materials and Methods

It's a cross-sectional study was performed in Atbara locality which located about 350 km North of capital Khartoum, between longitude 33.59 East and Latitude 17.14 North.

The study was carried out among aborted women. The study was conducted during the period from February to May 2017.

Sample Collection

A total of 152 venous blood samples were collected from aborted women using 5 ml sterile syring. The collected blood samples were placed into plain vacutainer. Serum were separated after centrifugation for (15) minutes in 3000 rpm. Then were stored in (-20°C) till used.

A data collected through questionnaires which include personal information, demographic data, socio-economic data, and health data.

Ethical Consideration

The approval was taken from research committee of college of medical laboratory science, Sudan University of

Science and Technology. Written informed consent was obtained from all study participants after explaining the study purpose.

Latex Agglutination Test

The reagents and samples were brought to room temperature, 100µl from sample, positive and negative control were placed into the black circle of the slide, latex reagent was mixed well and 50µl of latex were added for serum, positive and negative controls, then were mixed well with plastic stick and slide was tilt, the presence or absence of agglutination was observed within a period no longer than 3 minutes.

Immuno Chromatography Test (ICT)

Test cassette, specimen were allowed to room temperature (15-30° C) prior to testing, the test cassette was removed from the foil pouch and used immediately, the test device was placed on a clean and level surface. The dropper was holden vertically and 2-3 drops of serum or plasma (60 - 90µl) was transferred to the specimen well of the test device, the result was read in 15 minutes.

Enzyme linked Immuno-Sorpant Assay (ELISA)

The desired numbers of coated strips were placed into the holder, 1:40 dilutions were prepared by adding 5µl of the test samples, negative and positive control and calibrators to 200 µl of sample diluent and were mixed well, 100 µl of diluted sera, calibrators and controls were dispensed into the appropriate wells. For the reagent blank, 100 µl samples diluent were dispensed in 1A well position. The holders was tapped to remove air bubbles from the liquid and was mixed well, then were incubated for 30 minutes at room temperature.

Liquid was removed from all wells and washed three times with washing buffer, 100 µl of enzyme conjugate was dispensed to each well and incubated for 30 minutes at room temperature, enzyme conjugate was removed from all well by repeated washing three times with washing buffer, 100 µl of TMB chromogenic substrate was dispensed to each well and incubated for 15 minutes at room temperature, then 100 µl of stop solution was added to stop reaction. Finally, optical density was read at 450 nm with a micro well reader.

Sensitivity and Specificity

The sensitivity and Specificity were calculated using the two formulas below:

Sensitivity= (+ ve cases tested technique)/(+ve cases of reference technique) ×100

Specificity= (- ve cases tested technique)/(-ve cases of reference technique) ×100

Data Analysis

All information and data was analyzed by using statistical package of social science (SPSS) (version 20; corp., College station, Tex), using Chi square test, then data was presented in tables using excel.

Results

General Characteristics of the Study Population

This study was conducted on 152 aborted women, their ages ranged between 20-49 years old, the age groups were divided into 20-30, 31-40 and 41-49 years old. The frequency of each group as follow; 18 (11.8%), 51(33.5%) and 83(54.6%) respectively (Table 1). Three out of 152 tested women had never been to school (1.9%), 30 had elementary education (19.7%), 54 had ended high school (35.5%) and 65 had university education (42.7%) (Table 2). Eighty seven of tested women have house animal (57.2%).

Age groups (years)	Frequency	Percentage (%)
20-30	18	11.80%
31-40	51	33.50%
40-49	83	54.60%

Table 1: Frequency of age groups.

Level	Frequency	Percentage (%)
Never been to school	3	1.9
Elementary	30	19.7
High school	54	35.50%
University	65	42.70%

Table 2: Frequency of education.

The Prevalence Rate of *Toxoplasma* Parasite among Aborted Women According to Age Groups

The results showed that, the highest prevalence of *Toxoplasma* parasite infection was among age group (41-49) years old, which comprised 54.6% of all studied subjects, followed by 32.9% among (31-40) age group, then 12.5 % among age group (20-30) years old. The differences in rate was found to be statistically insignificant at P value=0.06 (Table 3).

Age groups	Number examined	Positive <i>Toxoplasmosis</i>	Prevalence (%)
20-30	19	7	12.50%
31-40	50	17	32.90%
41- 49	83	30	54.60%

P value=0.06

Table 3: Relationship between prevalence rate of *Toxoplasmosis* among aborted women and age groups

The Overall Prevalence Rate of *Toxoplasma* Parasite among Aborted Women

The result showed that, out of the 152 blood sample collected from aborted women, 54 were found positive for *Toxoplasma* IgG with prevalence rate (35.5%), while only one IgM positive with prevalence rate (0.6%) (Table 4).

Number examined	Positive IgG	Positive IgM
152	54 (35.5%)	1 (0.6%)

Table 4: The overall prevalence rate of *Toxoplasma* parasite among aborted women

The Prevalence Rate of *Toxoplasma* Parasite among Aborted Women According to the Technique Used

The prevalence rate of *Toxoplasma* parasite among aborted women by different techniques was as follows: 33.6 % by latex technique, 22.4 % by ICT technique and 35.5 % by ELISA technique. The differences in rate was found to be statistically highly significant at P value=0.00 (Table 5).

Sensitivity and Specificity Rates of LATEX and ICT

Assuming ELISA as the gold standard, the sensitivity and specificity of the LATEX technique were 94 % and 100% respectively and the sensitivity and specificity of the ICT technique were 64% and 100% respectively (Table 6).

		ELISA		Total
		+ve	-ve	
ICT	+ve	28(18.4%)	7(4.6%)	35(23.0%)
	-ve	26(17.1%)	91(59.9%)	117(77.0%)
Total		54(35.5%)	98(64.5%)	152(100%)
latex	+ve	37(24.3%)	14(9.2%)	51(33.6%)
	-ve	17(11.2%)	84(55.3%)	101(66.4%)
Total		54(35.5%)	98(64.5%)	152(100%)

Table 6: Sensitivity and Specificity rates of LATEX and ICT with ELISA.

Discussion

Toxoplasma gondii cause severe impairment and death to fetuses or newborns through congenital infection. In this study, the overall seroprevalence rate of *Toxoplasma* infection among aborted women 35.5% among the tested participants, all of them IgG positive, while only one IgM positive (0.65%), the findings of IgG were higher than those detected by Saja, (2010) in Al-Iraq 20 who was reported only 23% were IgG positive, but our findings in IgM was lower than his reported 3.5% IgM positive. Also, Adnan and Abdel mom'em, (2009) in Gaza [21] reported a lower rate (17.9% and 12.8%) for IgG and IgM respectively.

Our findings is dis agreed with studies in Sudan by Elhag and Elturabi, (2015) in Khartoum state [22], Abdel-Raouff and Elbasheir, (2014) in Khartoum and Omdurman [23], they were reported a lower rate of *T.gondii* IgG (28.4% and 20%) and higher rate of IgM (5.3%), none of the examined women had anti toxoplasma IgM positive. The differences may be due to study area and the number of subjects enrolled in each study.

Satti et al, (2011) in khartoum state [24] used ELISA technique to detect prevalence of IgM and IgG antibodies, they were obtained 23.1% and 16.4% respectively. Their findings were disagreed with our finding and this is may be due to differences in the study area and study population enrolled in each study.

The study done by Elsheikh (2015) in Wad madani [25] was reported seroprevalence of 41.7% using latex, this study nearly closed to our findings, although his studied the relation of some risk factors to the disease, and found that consuming raw meat and contact with pets other than cats and dogs were highly significant to infection. Also other a study done by Ahmed, (2016) in Soba Area, Khartoum State [26] was reported that, age groups of the participants ranged between (16-40) years old were 29% IgG +ve, while 4.6 % were IgM +ve by using ELISA. Our findings were disagreed to her findings in age group, the highest prevalence of toxoplasmosis in our study was in age group above 40 years age.

Conclusion

The study was concluding that, the infection of toxoplasmosis exists in Atbara locality, and positive seroprevalence of antibodies (IgG and IgM) was detected in pregnant women, and was prevalent among age group (20-49) years old.

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