Relationship between *Toxoplasma gondii* and abortion in aborted women in Najaf province

العلاقة بين الإصابة بطفيلي مقوسات كوندي والإجهاض بين النساء المجهضات في محافظة النجف

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Abstract

Background: Toxoplasmosis, caused by the protozoan parasite *Toxoplasma gondii*, is one of the most common parasites of man; Toxoplasmosis may causes severe symptoms in the congenitally acquired form with severe complications such as miscarriage, fetal developmental retardation, intracranial calcification, hydrocephalus, encephalitis, neurological, mental illnesses and visual disorder.

Methods: A case-control study was conducted to estimate role of Toxoplasmosis in occurrence of abortion among pregnant women in Najaf province. The present study was carried out on 228 aborted women, and 127non-aborted (pregnant women at 20th week gestation and had more than one successful birth as a control). All of these cases were limited to females only in the reproductive age (15-50 years). Toxo-IgM and Toxo-IgG antibodies were detected with Enzyme-Linked Immunoadsorbent Assay (ELISA) (Human/ Germany) in both groups.

Results: It was found that IgG positive cases were 112 (31.5%), IgM positive cases were 27 (7.6%) and both IgM and IgG positive cases were 9 women (2.5%). The present study showed that toxoplasmosis positive women were about 4.8 folds more likely to have abortion (OR= 4.8, P=0.0001).

Conclusion: There is association between infection with *Toxoplasma gondii* and occurrence of abortion.

المستخلص

داء المقوسات الناجم عن الإصابة بطفيلي التوكسوبلازما, وهو واحد من الطفيليات الأكثر شيوعا في الإنسان, المرأة الحامل أذا اكتسبت الإصابة لأول مرة أثناء الحمل يسبب لها أعراض حادة ومضاعفات خطيرة للجنين مثل الإجهاض, والتخلف التنموي للجنين, والتكلس داخل الجمجمة, استسقاء الدماغ, والتهاب الدماغ, والأمراض النفسية واضطرابات البصرية. أجريت هذه الدراسة لتقييم دور داء المقوسات في حدوث الإجهاض بين النساء الحوامل اللواتي عانين من الإجهاض المتكرر في محافظة النجف, وقد شملت هذه الدراسة على 228 امرأة عانت من الإجهاض و 127 امرأة مجموعة سيطرة (هن النساء الحوامل في الاسبوع20 من الحمل، ولديهن أكثر من ولادة ناجحة ولم يعانين أي حالة إجهاض). اقتصرت جميع هذه الحالات للإناث فقط في سن الإنجاب (15-50 سنة). تم التحري عن وجود الأضداد لطفيلي التوكسوبلازما في عينات الدم المأخوذة من النساء IgG, IgM باستخدام تقنية الاليزا (الادمصاص المناعي المرتبط بالانزيم) وكانت ايجابية الغلوبولين المناعي المرتبط بالانزيم) وكانت ايجابية الغلوبولين المناعي IgM و IgM و IgM و اللها و (2.5٪): النتائج. وأظهرت الدراسة أن النساء المصابات بداء المقوسات كن حوالي 4.8 أضعاف أكثر عرضة للإجهاض OR (2.5٪): النتائج. وأظهرت الدراسة أن النساء المصابات بداء المقوسات كن حوالي 4.8 أضعاف أكثر عرضة للإجهاض OR التوكسوبلازما وحدوث الإجهاض.

1. Introduction

Toxoplasma gondii is a single-cell protozoan that belongs to the family Coccidia. It is an obligatory intracellular protozoan with a heterogeneous life cycle in humans and other vertebrates [1].

The transmission of the disease to human is either by eating raw or uncooked meat, blood transfusion and organ transplantation or through ingestion of oocysts introduced into the environment by cats or congenitally during pregnency [2].

Toxoplasmosis is important due to the possibility of transplacental transmission and harming the fetus[3]. When primary infection is acquired by a pregnant woman, tachyzoites can colonize

placental tissues during the dissemination process and from there can gain access to the fetal compartment may lead to abortion, stillbirth or severe disease of fetus [4, 5].

The incidence of infection depends on the immunological state of the population and favorable environmental conditions, such as hot weather for the survival of the oocysts, which are discarded in cat feces, also genetic factors [6, 7].

The clinical implications of infection due to *Toxoplasma* in pregnant patients are manifold. Such patients may have spontaneous abortions, stillbirth, intrauterine growth retardation, preterm deliveries or fetal anomalies. In addition to the risk of gestational complications and congenital infections, it has been suggested that toxoplasmosis has some unfavorable effects on reproductive capacity in both men and women [8].

In women during toxoplasmosis, the proliferative tachyzoites may invade female reproductive organ specially the oviducts, subsequently evoke tissue cysts formation in it, that may produce tubule dysfunctions, the latter action can leads to hormonal abnormalities which give rises into secondary infertility sequels [9].

The aim of the research for the study possible association between infection with *Toxoplasma gondii* and abortion in Najaf .

2. Materials and methods

This case-control study was performed on 355 women, The mean age of the participant women was 29.5±7.6 (range:16-50) years. The women in the current study were classified into two groups; First group: involved 228 women with bad obstetric history (BOH), who suffered from previous abortions, Second group: included 127 non aborted women without obstetric problems and had more than one successful birth. They were attending to AL-Zahraa Maternity and Pediatric Teaching Hospital from March, 2013 to March, 2014.

This study was approved before its commencement by the ethical committee of the Faculty of Medicine, University of Kufa, and informed consent was obtained from all individuals.

Four milliliter of venous blood was collected from patients and control in sterile serum tube and left for one hour at room temperature to allow the clot to form. Then, centrifuged at 3000 rpm for 15 min. to separate the serum which divided in eppendorf tubes ($200\mu l$) and kept at deep freeze (-20 C°) until used.

The search for anti-*T. gondii* antibodies in serum was performed by Enzyme Linked Immunosorbent Assay (ELISA) by using Toxo IgM ELISA and Toxo-IgG ELISA (Human-Germany). It was done according to manufacturer's instructions

Data of the studied group were checked for any error or inconsistency, entered and analyzed by using the statistical package for social sciences (SPSS) version 22, 2014. Chi square test was used to compare frequencies. Level of significance, P. value set at ≤ 0.05 to be considered as significant difference or correlation.

3. Results

Seroprevalence data obtained are shown in table (1). It had been found that the positive IgG only detected in 112 women (31.5%), IgM only in 27 (7.6%) and both Igs were detected together in 9 women (2.5%), these findings revealed an overall 148 positive toxoplasmosis and 207 negative toxoplasmosis women.

Table 1: Distribution of studied groups according to the anti-Toxoplasma- antibodies.

		p					
Parameter	Aborted		Non-aborted		Total		
	No.	%	No.	%	No.	%	
IgG positive	87	38.2	25	19.7	112	33.4	0.00001
IgM positive	27	11.8	0	0	27	8.1	
Both Ig positive	9	3.9	0	0	9	2.7	
Negative	105	46.1	102	80.3	207	61.8	
Total	228	100	127	100	355	100	

The demographical distribution of the study groups show no statically significant among aborted and non aborted women in regard to age and occupation, table (2). The majority of infection with toxoplasmosis was (67.6%) among women of residence urban and (83.8%) seropositive among housewife occupation table (3).

Table 2: Demographical distribution of the studied groups (N=355).

Variable		Aborted women		Non aborted women		Total		p
		No.	%	No.	%	No.	%	
	≤ 20	33	64.7	18	35.3	51	100	0.45
Age (year)	21 - 30	98	60.5	64	39.5	162	100	
	31 - 40	75	70.1	32	29.9	107	100	
	41 - 50	22	62.9	13	37.1	35	100	
	Mean	29.9±7.5		28.6±7.8		29.5±7.6		0.12
Occupation	Housewife	193	66.3	98	33.7	291	100	0.05
	Employed	29	60.4	19	39.6	48	100	
	Student	6	37.5	10	62.5	16	100	

Table 3. Demographic characteristics of the studied group distributed according to the toxoplasmosis infection (N=355).

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Variable		Toxoplasmosis Positive N=148		Toxoplasmosis Negative N= 207		Total		p
		No.	%	No.	%	No.	%	
	≤ 20	16	31.4	35	68.6	51	100	0.07
	21 - 30	66	40.7	96	59.3	162	100	
Age (year)	31 - 40	45	42.1	62	57.9	107	100	
	41 - 50	21	60	14	40	35	100	
	Mean	30.7±7.8		28.6±7.4		29.5±7.6		0.009
Residence	Urban	100	41	144	59	244	100	0.68
	Rural	48	43.2	63	56.8	111	100	
Occupation	Housewife	124	42.6	167	57.4	291	100	0.16
	Employed	21	43.8	27	56.2	48	100	
	Student	3	18.8	13	81.2	16	100	

The cross tabulation between toxoplasmosis infection and abortion revealed that toxoplasmosis positive women were about 4.8 folds more likely to have abortion (OR= 4.8, P=0.0001). Table (4)

Table 4. Distribution of women according to the abortion and toxoplasmosis infection

Toxoplasmosis		Total				
	Abo	orted	Non-A	Aborted	1 0 111	
	No.	%	No.	%	No.	%
Positive	123	53.9	25	19.7	148	41.7
Negative	105	46.1	102	80.3	207	58.3
Total	228	100	127	100	355	100
Odd's ratio (95% CI) = 4.8 (2.9-7.9) P. value =0.0001						

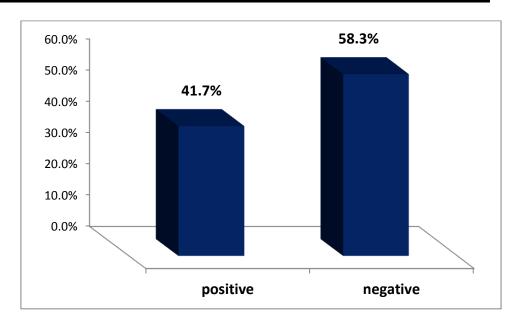


Figure 1. Distribution of the studied group according to the toxoplasmosis

4. Discussion

The extent to which toxoplasmosis causes habitual abortion is still controversial. Therefore, this study was achieved for the study possible association between infection with *Toxoplasma gondii* and abortion.

Toxo-IgM and Toxo-IgG antibodies were detected with Enzyme-Linked Immunoadsorbent Assay (ELISA) in both groups. It was found IgG positive cases were 112 (31.5%), IgM positive cases were 27 (7.6%) and both IgM and IgG positive cases were found in 9 women (2.5%).

Most cases with positive IgG titer are enough to establish that the patient has been infected with *Toxoplasma gondii* and indicate chronic infection, while positive IgM indicate recent infection [10].

The results of the present study lower than results of the previous studies conducted in Najaf city by [11] who mentioned that, out of 201 aborted women only 36%, 5% and 13.45% were positive for IgG, IgM, and positive both IgM and IgG respectively. [12] found that, the positive *Toxoplasma* results were 48% and 19.6% by IgG-ELISA and IgM-ELISA respectively. [13] revealed that, the percentage of toxoplasmosis by IgM-ELISA was 13.95%. Whereas [14] recorded that, 4.25% was positive by IgM-ELISA.

The results of those studies in comparison with current study in spite of there is variation in percentage of infection indicate that the toxoplasmosis is common among women in Al-Najaf and that encourage to know the actual or real relationship between the toxoplasmosis and occurrence of abortion.

The finding of the present study incompatible with [15] in Mid Euphrates area in Iraq who revealed that 46.75%, 5% and 47.4% were positive by IgG, IgM and IgG & IgM- ELISA test respectively. The seropositive percentage in this study was nearly similar to results in Wassit by [16] who found that, the seroprevalence of anti-*Toxoplasma gondii* IgG was 31.6%.

The seropositivity of present study higher in regard to IgG and lower in regard to IgM than results by [17] in Baghdad they showed that, 25(20.5%) women were positive for IgG, and 17(13.9%) women were positive for IgM, while 9 (7.4%) women were positive for both. [18] in Baghdad showed the seroprevalence (IgG) toxoplasmosis was 48% in recurrent spontaneous abortion (RSA) group, while for (IgM) toxoplasmosis was 4%. [19] revealed that, 9(26.5%) women were positive for specific IgG anti-toxoplasma antibodies, while 4(11.8%) women were positive for both specific IgG and IgM anti-toxoplasma antibodies out of 34. In Salah-Adden, Toxoplasma prevalence among pregnant women with single or multiple abortions was 26.1% and 3.1% for IgG and IgM respectively [20]. In Erbil [21] said that, seropositivity for toxoplasmosis by ELISA IgG was 37.5%. In Kirkuk [22] showed that, out of (319) of pregnant women only 54 (16.9%) was IgM-

ELISA positive for Toxoplasma gondii. These variations may be due to sample size in addition to different circumstances and times of sample collection and processing.

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 [23].

In the present study, the infected aborted women were 83.1% and infected non-aborted women were 16.9%. It had been significantly found that the toxoplasmosis positive women were about 4.8 folds more likely to have abortion (OR= 4.8, P=0.0001).

This result was supported by others researchers. [24] in Najaf, [25] in Yemen, [26] in Erbil, [27] in Sudan and [28] who indicated correlation between positivity of Toxoplasma IgG antibodies and past history of abortion and congenital anomalies. So the conclusion referred to that Toxoplasmosis is suggested to be the major parameter in infectious causes of spontaneous abortion.

The significant correlation between toxoplasmosis and abortion in the present study confirms and extends with previous findings by [18, 29] and [30] who showed that, the risk of developing habitual abortion among women who were seropositive was 3.6 times (OR 3.6, P=0.01). Nevertheless, these findings are not consistent with other studies which didn't confirm this association [31, 32].

In the current study the distribution of toxoplasmosis according to the age revealed that the majority of toxoplasmosis patients were in age group 41- 50 years (60%). Several studies have indicated an increase in seroprevalence with age [33, 34, 35, 36, 37, 38, 39]. [40] recorded that, the highest level of toxoplasmosis was among 35-45 age group, in comparison with low rate of infection at 15-19 age group [41, 42].

These results may be due to the cumulative effect of exposure to the infective stages of the parasite.

The distribution of toxoplasmosis according to residence showed no significant differences, however the higher percent of toxoplasmosis in study groups was (67.6%) in the urban regions compared with the rural ones. These findings are corresponding to results by [43, 13] in Najaf province. [44] recorded nearly similar results in Yemen among women in urban areas. The findings of present study may be due to the dealing with high number of cases in urban area comparing with rural area and their different habits in eating poultry and junk food from restaurants which may be a major source for *T. gondii* transmission.

The present study showed that there were no significant differences between prevalence of infection with toxoplasmosis in study groups in regards to occupation. However the majority of toxoplasmosis was (83.8%) among housewives. This result consistent with other studies like [15] in Najaf, [21] in Erbil, [44] in Mexico and [45] in Saudi Arabia, [41, 46] who said that, the high percentages of toxoplasmosis were recorded among housewives.

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. The infection in housewives may be attributed to meat processing and working in home garden [47].

The results of the present study in contrary with [12] who said that, the officials were more affected with toxoplasmosis. Likewise, Mwambe said that, business women and employed pregnant women had higher infection rates with *T. gondii* than housewives.

The occurrence of congenital toxoplasmosis is still a problem in our community and that the best prevention is the prompt and adequate examination of pregnant women for the presence of *Toxoplasma gondii* infection and required more attention from the health institution.

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