SEROPREVALENCE OF *TOXOPLASMA GONDII* AMONG PREGNANT WOMEN IN BAGHDAD CITY [+]

نسبة الاصابة بداء المقوسات ضمن النساء الحوامل باستخدام الاختبارات المصلية في مدينة بغداد

Taghreed Kheder Mohammed [*]

Abstract:

The current study is one of the prime investigations to evaluate the prevalence rate of *T. gondii* among pregnant women in Baghdad city and to consider some of the environmental and personal factors that may contribute to infection. Serum samples of 212 pregnant women of 18 to 33 years of age attending the Maternity hospital in Baghdad were tested for anti-*Toxoplasma* IgG and IgM antibodies using enzyme-linked immunosorbent assay (ELISA). Serological results, reflecting *T. gondii* prevalence rate, were statistically analyzed and linked to epidemiological data collected through a standard questionnaire. The seroprevalence of anti-*Toxoplasma* IgG was ٢٨,٧٧% (61 out of 212), whereas IgM seropositivity was 23.8 %. The highest IgG and IgM seroprevalence were among participants aged 26 to 33 years (31.3 % and 9.1 % respectively). No statistically significant relation was observed between *T. gondii* seroprevalence and the other variable factors studied. The current study indicates that there is a considerable rate of *Toxoplasma* infection among pregnant women in Baghdad and support the concern that Iraqi women may be vulnerable to that infection. Moreover, it shows the need to provide health education to pregnant women in order to prevent primary infection during pregnancy.

المستخلص:

تهدف هذه الدراسة إلى تقييم نسبة الإصابة بطفيلي التوكسوبلازما بين النساء الحوامل في مدينة بغداد و كذلك دراسة العوامل البيئية المحيطة و كذلك العادات الشخصية لهؤلاء النسوة والتي قد تساهم في انتشار المرض .تم اختبار عينات دم (٢١٢) من النساء الحوامل في المرحلة العمرية ما بين ١٨ – 33عامًا و اللاتي يترددن على مختبرات الصحة المركزي لمعرفة اسباب الاجهاض المتكرر. وأخضعت كل العينات لاختبار الادمصاص المناعي المرتبط بالانزيم للكشف عن الأجسام المضادة لطفيلي التوكسوبلازما من النوعين"IgG" و"IgM"و أجريت الاختبارات الإحصائية اللازمة لتحليل هذه النتائج وبيان مدى ارتباطها بالمعلومات البيئية والعادات الشخصية والتي سيقت من خلال نموذج للإستبيان الذي تم إستيفاؤه من النسوة المشاركات في الدراسة. وقد أظهرت النتائج أن النسبة المئوية لوجود الأجسام المضادة من النوع" IgG " ، والتي تدل على نسبة الإصابة بطفيل التوكسوبلازما بين عينات البحث بلغت 76.3 % (61 سيدة من إجمالي ٢١٢) بينما كانت نسبة تواجد الأجسام المضادة"IgM" ، والتي تعبر عن الحالة الحادة للمرض، لا تتعدى 23.8 % كما أوضحت الدراسة أن نسبة تواجد الأجسام المضادة"IgG" و"IgM "كانت الأعلى بين المصابات من المرحلة العمرية الأكبر (26 – 33) عاما حيث كانت 31.3 % ، 9.1 % على

[+] **Received on 15/6/2010 ,Accepted on 24/2/2011.**

[*]**Institute of medical technology/ Al - Mansour**

التوالي .كذلك لم تكن هناك أي علاقة إحصائية دالة بين نسبة الإصابة بطفيل التوكسوبلازما بين عينة البحث وأي من المتغيرات البيئية والشخصية التي تم دراستها.

تدل نتائج هذه الدراسة على أن هناك نسبة عالية نسبيا من الإصابة بطفيلي التوكسوبلازما بين النساء الحوامل في مدينة بغداد ، مما يحث على البدء في برامج للتثقيف الصحي للنساء الحوامل لمنع الإصابة الأولية بطفيل التوكسوبلازما أثناء فترة الحمل والتي قد تؤدى إلى مضاعفات سلبية على الجنين.

Introduction:

Toxoplasma gondii is an obligatory intracellular protozoan parasite which appears to have broad host specificity. Cats and wild Felines are the only definitive host while all other worm-blooded animals including humans are intermediate hosts [1]. Infection is acquired by ingestion of viable tissue cysts in meat or oocysts excreted by cats that contaminate food or water [2]. Congenital transmission may occur when an uninfected mother acquires primary infection during pregnancy [3]. Even though,pregnant women are often asymptomatic or have only mild symptoms, infection may cause spontaneous abortion, still birth, or serious foetal damage. The gestational age at which the infection is contracted is a key variable affecting the clinical foetal outcome [3, 4, and 5].

Serological testing for anti-*Toxoplasma* antibodies is the mainstay for the diagnosis of toxoplasmosis. Diagnosis of acute maternal infection is mainly based on detection of seroconversion or fourfold rise in IgM antibodies level which appear sooner after infection than IgG antibodies and disappear faster than IgG after recovery [2].

Acute infection in pregnant women was caused by eating raw or under cooked meat and soil contact [6]. Toxoplasma gondii infection during pregnancy means either primary infection or risk of congenital toxoplasmosis or acute retinochoroiditis in a pregnant woman . Ingestion or manipulation of raw or undercooked meat is responsible for most contaminations (one- to two-thirds) in pregnant women. Toxoplasmosis seroprevalence is high in Europe, up to 54 % in southern European countries. Primary prevention advice is proposed to immunocompetent pregnant women who are seronegative for toxoplasmosis [7]. The Possible routes of infection were contaminated soil, drinking rainwater and eating raw vegetables rather than eating uncooked meat or contact with cats [8].

Ayi *et al.*, 2009 [9] found that a total of 294 Ghana pregnant women aged 15–42 years (mean age: 28.06 years; std. dev.: 5.528 years) volunteered to participate in the study after informed consent. Two pregnant women (0.7%) could not give their ages. The 25–29 years age group recorded the highest participation, 37.1% (109/294) and the 40–44 year-olds had the least of 2.0% (6/294). All the 294 pregnant women responded to over 90% of question items in a questionnaire interview, however, not all of them volunteered to give blood for testing. About 10.2% (30/294) of the pregnant women were in their first trimester with 50.7% (149/294) and 38.4% (113/294) in the second and third trimesters, respectively, while 0.7% (2/294) could not give the age of their pregnancy. Only one woman had some knowledge about Toxoplasmosis because she had been tested for it in the past.

There have been a few reported cases of outbreaks in some areas implicating drinking unfiltered water with possible oocyst contamination [9,10,11]. Outbreaks involving a group of students of the Cornel University Medical College implicating eating of undercooked hamburgers and, in Sao Paulo 110 persons admitting to eating undercooked meat after being diagnosed with toxoplasmosis have been reported [12,13].

Toxoplasmosis may be diagnosed by direct and indirect methods. Direct methods include Polymerase Chain Reaction (PCR) for the detection of *T. gondii* DNA from samples of body

fluids, demonstration of the organism through mouse inoculation, tissue culture, ophthalmic testing and radiological studies (14). Indirect diagnosis is mainly serological methods for the detection of anti-*T. gondii* antibodies. The most commonly used serologic tests to detect the presence of anti-*T gondii* IgG and IgM antibodies are the Sabin-Feldman dye test, indirect fluorescent antibody (IFA), and agglutination tests, or ELISA [15 , 16 , 17 ,18] .

The objective of the current study is to identify the seroprevalence of anti- *Toxoplasma* IgG and IgM antibodies among pregnant women in Baghdad city, and to determine the environmental, personal and behavioral factors associated with infection in women.

Patients and methods:

Study group

A total of 212 pregnant women attending the Maternity Hospital in Baghdad, participant consent and hospital approval were obtained. The age of the study groups ranged from 18-33 years. All of them have a history of spontaneous abortion.

Epidemiological assessment

A questionnaire sheet was designed to assess some of the main risk factors which may influence the prevalence of *Toxoplasma* infection among the pregnant women with recurrent abortion. These data were intended to be completed by interviewing each participant during her hospital visit followed by home visits to central health laboratory to validate questions related to environmental factors. The influential risk factors considered in the study include: maternal age, gestation period, owning cats, sources of drinking water, and the nature of cooking and eating habits such as: eating raw or undercooked meat, and eating in restaurants.

Samples

Double blood samples were taken from each pregnant woman. Samples from all patients were collected over six-month from January to June, 2009. Blood samples were collected in a 10 ml tubes. Samples (sera) were separated by centrifugation and stored in small tubes at -20°C till testing.

Serological testing

Serum specimens were tested by ELISA to detect anti-*Toxoplasma* IgG and IgM antibodies using Euroimmun , (U.K.) according to manufacturer's instructions. The cut-off value of the assay was calculated and results were expressed in an index by dividing sample absorbance by the cut-off value. The test was considered negative if the index was <0.8, the result was equivocal when index was from 0.8 to <1.1, while the positive result was if index was ≥ 1.1. A negative reaction indicates absence of significant *Toxoplasma* antibodies. A positive *Toxoplasma* IgG reaction was interpreted as an indication of either a past or recent infection.

Statistical analysis

A descriptive statistical analysis of the serological data in relation to the epidemiological variables, used in the study, was done. Chi-square test and t-test were used and appropriate p values of <0.05 were considered significant. Data were analyzed using SPSS version 12 software.

Results:

The questionnaire data revealed that (75 %) of participants have never heard or seen information about toxoplasmosis prior to the interview. Out of 212 sera tested, 61 samples were IgG positive (76.3 %), while 19 were IgM positive (23.8 %).A history of spontaneous abortion was reported in (54 %) of the IgG positive cases and (3 %) of the IgM cases.

Seroprevalence in relation to gestational age

There was no statistical significant correlation between IgG or IgM seropositivity and the gestational age of the fetus. Comparable results of seropositivity of both IgG and IgM were obtained in both first trimesters (76.3 % & 23.8 % respectively) (Table 1).

Seroprevalence in relation to women age

The seroprevalence of *Toxoplasma* IgM level according to maternal age was 9.1 % with no significant difference among age groups, whereas, the IgG level was significantly higher ($p<0.05$) in older age groups. Among expectant females aged 26 - 33 year old was (31.3 %) compared to (26.5 %) of younger age group 18-25 years old. While, the seropositivity of IgM increased with age reaching its highest level (9.1 %) in the oldest age group (26 - 33 year old), its correlation to the different age groups was not statistically significant (Table 2).

Seroprevalence in relation to educational background

Results showed that IgG seroprevalence tends to be lower in educated participants (from primary school education up to university graduates) (26.9 %) compared to the uneducated group (30.2%). However, this tendency falls short of being statistically significant. On the other hand, the IgM seropopositivity levels were comparable between educated and uneducated groups (8.6% and 9.2% respectively) (Table 3).

Seroprevalence in relation to other influence factors

There were no significant associations between seroprevalence of anti-*Toxoplasma* IgG and IgM antibodies and other risk factors considered in the study. These influential factors include: water supply, cat exposure, consumption of undercooked meat, being exposed to any kind of knowledge about toxoplasmosis and history of previous abortion (Table 4).

Discussion:

The current study is one of many studies in Baghdad to explore the prevalence of *Toxoplasma gondii* infection in pregnant women. If a woman gets infected with *T. gondii* for the first time in her life during pregnancy, she may pass infection to her fetus; a situation that ultimately could lead to a very serious fetal damage. Most previous studies in Baghdad have concentrated on the prevalence of *Toxoplasma* infection among general population. The current study is also one of the leading studies that evaluate some environmental and behavioral factors that may influence the infection rate of *T. gondii* in Baghdad city. The *Toxoplasma* seroprevalence obtained in this study among pregnant women in Baghdad was 37, 7 % which is comparable to results previously reported in Makkah 35.6% [19]. A comparable seroprevalence data was also obtained from healthy blood donors from two urban

areas in the Eastern Region (25- 26.4) % [20]. Low prevalence rates of 10% were reported in the United Kingdom [21] and rates as high as around 55% were reported in France [22]. Higher prevalence rates were also reported in some neighbouring Arab countries like Kuwait (58.2%) [23].

Regional variations in the incidence of *Toxoplasma* infection rates from one country to another or even within the same country, has been well documented. This variation has been attributed to climate, cultural differences regarding hygienic and feeding habits [1,11,12,13].

The frequency of stray cats in a humid rainy climate favouring the survival of oocysts has contributed to the high *Toxoplasma* prevalence in Central America [1]. Stray cats are widely spread in Baghdad city, however, the hot and dry weather conditions are not ideal for oocyst survival, compared to cooler and more rainfall environmental conditions in the north regions of Iraq, which are in favor of a higher prevalence. Farming and animal rearing are also common.

The significant relation showed in the current study between *Toxoplasma* prevalence rate and the mother's age confirms the fact that seroprevalence of *Toxoplasma* is well known to increase with age; the greater the prevalence, the earlier the rise [1,13,14] .This association does not mean that older age is a risk factor predisposing to infection but might be explained by the older the person the longer time being exposed to the causative agent and may retain a steady level of anti-*Toxoplasma* IgG in serum for years. A contradictory result was reported in the Eastern Region where seropositivity declined with age [7, 15].

The current results suggested that the educational level of pregnant women, in spite of being not statistically significant, may present possible protective measures against *Toxoplasma* infection. High education level may reduce risk exposure and increase awareness to adopt appropriate hygienic measures regarding food and cooking habits such washing chopping boards with soap or bleach, using different chopping board for meats and vegetables, the frequent washing of knifes and hands while cooking and avoiding contamination of food by protecting it from flies and dust.

33% of the Indian women of low socioeconomic group and 22% of the women of high socioeconomic group were seropositive for *Toxoplasma* specific IgG antibodies possible acute infection as indicated by IgM positivity alone, was higher 18.3 % in women of high socioeconomic group, compared to women from low socioeconomic group 5%. This indicates that the incidence of primary infection is higher in women of high socioeconomic group and these women are at a greater risk of foetal loss if they conceive during the acute phase [18].

Al-Harthi *et al*. , 2006 in Makkah observed that *T. gondii* infection for the first time in the life of pregnant women .Serum samples of 197 pregnant women aged 17- 45 years were tested for anti-*Toxoplasma gondii* IgG and IgM antibodies using ELISA . The seroprevalence of anti - *Toxoplasma gondii* IgG was 29.4 % , whereas IgM seropositivity was 5.6% [19]. Abu-Madi *et al* .,2008 in Qatar showed that overall prevalence of IgG responses was 29.8% and this did not differ between the sexes nor between the three years of the study although there was a marked age effect. Among children less than 1 year old prevalence was 22.9%, but then dropped to <4% in the age group up to one year , indicating that these antibodies were most likely acquired in uterus from immune mothers [24]. Prevalence then increased steadily to peak at 41.2% among the oldest age class (>45 years). The prevalence of IgG antibody also varied significantly with region of origin, with higher rates for subjects from Africa, followed by those from the Eastern Mediterranean or Asia and lowest rates for subjects from the Arabian Peninsula. No IgM antibodies were detected in any subjects younger than 19 years, but prevalence increased to plateau at 7 – 9% in subjects aged over 20 years, and also varied with region of origin. In this case prevalence was highest among subjects from the Arabian Peninsula and least among those from Asia. Prevalence of IgM was higher among male subjects but did not vary between the three years of the study.

Many of the risk factors examined, such as cat contact, handling or eating raw or undercooked meat and drinking unfiltered water have been documented to have an influence on *Toxoplasma* transmission in different parts of the world [16,17]. The absence of a statistically significant relationship between the prevalence of *Toxoplasma* infection among pregnant women in Baghdad and many of the factors explored in the study, does not indicate that these factors have no influence on the transmission of toxoplasmosis. However, it may suggest that such factors play a limited role in this region due to religious beliefs and cultural habits. Furthermore, weather conditions in Baghdad do not enable these factors to exert their full influence on *T. gondii* transmission.This study may also be informative and useful to the public health community. It revealed that only around 20 % of pregnant women were aware of toxoplasmosis and its association with congenital disease. Furthermore, all pregnant women were not aware of being previously tested for *Toxoplasma* nor were able to identify any risk factor by their gynecologist. This suggests that there is a need to launch an awareness program for the pregnant women and additional studies of this type should be encouraged to add to the knowledge of the community about the risks of exposure to *T. gondii* by pregnant women in Baghdad.

Table 1: Seropositivity of Anti-*Toxoplasma* IgG and IgM in relation to the first trimester

Antibody	First trimester		Total
	positive	Negative	
IgG	61 (76.25)	90	151
IgM	19 (23.75)	42	61
Total	80	132	212

Table 2: Seropositivity of Anti-*Toxoplasma gondii* IgG and IgM in relation to participant's age

Age group	IgG		IgM		Total
	+ ve	-ve	+ ve	-ve	
18 – 25	30 (26.5%)	48	10 (8.8%)	25	113
26 – 33	31 (31.3%)	42	9 (9.1%)	17	99
Total	61	90	19	42	212

Table 3: Seropositivity of Anti-*Toxoplasma gondii* IgG and IgM in relation to Educational background

Educational background	IgG		IgM		Total
	POSITIVE	NEGATIVE	POSITIVE	NEGATIVE	
Educated	25 (26.9%)	40 (43%)	8 (8.6%)	20 (21.5%)	93
Non educated	36 (30.2%)	50 (42%)	11 (9.2%)	22 (18.5%)	119
Total	61 (28.8%)	90	19 (9%)	42	212

Table 4: Seropositivity of Anti-*Toxoplasma gondii* IgG and IgM in relation to studied influence factors

Influence Factors		Total	IgG Positive cases	Percent	IgM Positive cases	Percent
Water source	Treated	190	41	21.6%	13	6.8%
	Untreated	22	20	90.9%	6	27.3%
Cat espouser	Yes	30	5	16.7%	1	3.3
	No	182	56	30.8%	18	9.9%
Uncooked meat	Yes	12	4	33.3%	1	3.3%
	No	200	57	28.5%	18	9.9%
Previous abortion	Yes	185	45	24.3%	3	1.6%
	No	27	16	59.3%	16	59.3%

References:

1-Remington JS, McLeod R, Thulliez P & Desmonts G.. Toxoplasmosis, infectious diseases of the fetus and newborn infant; in *In* Remington JS, Klein J (5th ed.). W. B. Saunders, Philadelphia, Pa. pp. 205. (2001).

2- Montoya JG & Liesenfeld O. Toxoplasmosis. "*Lancet*"; 363(9425): 1965.(2004).

3- Ades AE & Nokes DJ. Modeling age-and time-specific incidence from seroprevalence: toxoplasmosis. "*American Journal of Epidemiology"*.137 (9): 1022, (1993).

4- Lopez A, Dietz VJ, Wilson M, Navin TR & Jones JL. Preventing congenital toxoplasmosis. " *MMWR Recomm Rep*". 49(RR-2): 59.(2000).

5- Muna M. Al-Hamdani and Nadham K. Mahdi . Toxoplasmosis among women with habitual abortion." *Eastern Mediterranean Health Journal* ". (3), 2: 310-315, (1997).

6- **Tomita T. , Yamada T. , Weiss L. , and Orlofsky A.** Externally Triggered Egress Is the Major Fate of *Toxoplasma gondii* during Acute Infection "*The Journal of Immunology* ".October 21, (2009) .
7- Shin D., Cha D., Hua G., Cha G. and Lee Y. Seroprevalence of *Toxoplasma gondii* Infection and Characteristics of Seropositive Patients in General Hospitals in Daejeon, Korea *Korean . "J. Parasitol"*. 47 (2) : 125- 130, (2009).
8- K.I. *Nijem1 and S. Al-Amleh1*.Seroprevalence and associated risk factors of toxoplasmosis in pregnant women in Hebron district, Palestine " *La Revue de Sante de la Mediterranee orientale"*, 15 (5),(2009) .
9- AYI I. , EDU S. A.A , APEA-KUBI K. A. , BOAMAH D. , BOSOMPEM K. M. and DOH D..SERO-EPIDEMIOLOGY OF TOXOPLASMOSIS AMONGST PREGNANT WOMEN IN THE GREATER ACCRA REGION OF GHANA. "*GHANA MEDICAL JOURNAL*" . September , 43(3),(2009).
10- Isaac-Renton J., Bowie W. , King A. , Irwin G., Ong C., Fung C. , Shokeir O. , and Dubey J. .Detection of ***Toxoplasma gondii*** Oocysts in Drinking Water. "*Appl Environ Microbiol*". June; 64(6): 2278–2280, (1998).

11- Heukelbach J., Meyer-Cirkel V., Moura R., Gomide M. , Queiroz J. , Saweljew P. , and Liesenfeld O. Waterborne Toxoplasmosis, Northeastern Brazil . "*EID journal*", 13(2), February, (2007).

12- Molina E. and Ridley-Dash L. Toxoplasmosis in cats at Kabacan, Cotabato, Philippines . "*USM R & D J"*.16(1):53-55, (2008) .
13- www.parasitetesting.com , (2010).
14- www.lapcorp.com,Detection of Toxoplasma gondii Infection by PCR , (2004).
15- Ryu J.,Min D.,Ahn M.,Choi H. Rho S.,Shin Y. Choi B. and Joo H.*Toxoplasma* Antibody titers by ELIA and indirect latex agglutination test in pregnant women."*The Korean journal of parasitology "* . 34(4):233-238 , (1996).
16- Jones JL, Dietz VJ, Power M, et al. Survey of obstetrician-gynecologists in the United States about toxoplasmosis. *" **Infect Dis Obstet Gynecol".*** 9:23-31, (2001) .
17- Kook J., Lee H., Kim B. ,Yun C., Guk S. , Seo M. , Park Y., Hong S. , and Chai J. . Toxoplasma gondii antibody titers in sera of children admitted to the Seoul National University Children's Hospital . "*Korean J Parasitol*". March; 37(1): 27–32, (1999).
18- Abdi J., Shojaee S., Mirzaee A., Keshavarz H. Seroprevalence of Toxoplasmosis in Pregnant Women in Ilam Province, Iran." *Iranian J Parasitol*". 3(2): 34-37,(2008).
19- Al-Harthi A.; Jamjoom M.; Ghazi H. Seroprevalence of Toxoplasma Gondii Among Pregnant Women in Makkah, Saudi Arabia. "*Umm Al-Qura Univ. J. Sci. Med. Eng".* 18(2):217 -227 (2006).
20- Sundar P., Mahadevan A., Jayshree R. , Subbakrishna D. & Shankar S. Toxoplasma seroprevalence in healthy voluntary blood donors from urban Karnataka. "*Indian J Med Res* "126, July: 50-55,(2007).
21-Allain JP, Palmer CR & Pearson G.. Epidemiological study of latent and recent infection by Toxoplasma gondii in pregnant women from a regional population in the UK." *J. Infect.*" 36:189, (1998).
22-Robert-Gnansia E. Congenital toxoplasmosis. Orphanet Encyclopedia. http://www.orpha.net/data/patho/GB/uk-toxo. October (2003).
23- Iqbal J. and Khalid N. Detection of acute Toxoplasma gondii infection in early pregnancy by IgG avidity and PCR analysis." *J Med Microbiol* " . 56:1495-1499: (2007).

24- Abu-Madi M. , Al-Molawi N. and Behnke J. Seroprevalence and epidemiological correlates of Toxoplasma gondii infections among patients referred for hospital-based serological testing in Doha, Qatar . "*Parasites & Vectors"*. 1: 39 (2008).