Seroepidemiological study on *Toxoplasma gondii* of aborted women at AL- Shatrah hospital of AL- Shatrah district/Thi Qar city – Iraq

دراسة مصلية وبائية عن طفيلي التوكسوبلازما كوندي *Toxoplasma gondii* في النساء المجهضات في مستشفى الشطرة ـ قضاء الشطرة / مدينة ذي قار – العراق

*Alaa M. Nasser *Sukaina R. Neamah *Noor K. Matar
Ahmed N. alamiry * Prof .Dr. Ihsan Mohammed S. *** Rana Abd-Ameer Jawad
* General Directorate of Education in Thi-Qar , Ministry of Education .
**Cancer Research Unit, College of Medicine, University of Thi-Qar- Iraq
***University of Kerbala / College of Veterinary Medicine – Iraq
Key words:- ***Toxoplasma gondii* , ELISA , IgG, IgM, Aborted women , Iraq**

Summary

Toxoplasma gondii has a worldwide distribution and it is one of the most prevalent infectious agents in aborted Iraqi women. The present study was performed in Nasiriya city (AL-Shatrah) among different groups of women to estimate the seroprevalence of toxoplasmosis .The data were collected from AL-Shatrah hospital through a period from 01-January – 2013until 31-December-2013 by testing 928 serum samples during this period.

This study revealed high rates of infection by toxoplasmosis at 2013 , the number of women examined in that year were seropositive for toxoplasmosis which were 928 serum samples recorded from January – 2013, 132 (14.2 %) positive samples and 20 (2.2%) positive samples in the end of December of this year, The high rate of infection by toxoplasmosis was 134(14.4 %) in the March of 2013 while the lowest rate record 20 (2.2%) positive samples in the end of December / 2013. The results revealed the percentage of antibodies class IgG was 184 (19.83%) positive sample while the percentage of antibodies class IgM was 281(30.28%) positive sample **.**The age among aborted women in that study was between 12-48 years ,The results revealed (86 %) of aborted women were seropositive for *T . gondii* at age of (12-35) years , (12%) seropositive sample at age (36-40) years , and (2%) seropositive sample at age group (41- 48) years .The study showed high significant differences between aborted women age groups at ($p>0.05$) .

الخلاصة:-

الاصابات بطفيلي التوكسوبلازما كوندي تنتشر في جميع أنحاء العالم ، وهي واحدة من أكثر العوامل المعدية الاكثر انتشارا في النساء المجهضات في العراق. أجريت هذه الدراسة في مدينة الناصرية (الشطرة) بين مجموعات مختلفة من النساء لتقدير انتشار داء المقوسات باستخدام الفحص المناعي. تم جمع البيانات من مستشفى الشطرة خلال الفترة من 01 - يناير - 2013 حتى 31 - ديسمبر - 2013 بفحص 928 عينة مصل خلال هذه الفترة.

كشفت نتائج هذه الدراسة عن ارتفاع معدلات الإصابة بعدوى داء المقوسات خلال عام 2013 ، وكان عدد النساء اللائي تم فحصهن في تلك السنة إيجابياً للمصل من داء المقوسات 132 عينة (14.2 ٪) خلال شهر كانون الثاني من سنة 2013 وعينات إيجابية (20.2 ٪) في نهاية شهر ديسمبر من هذا العام ، كان معدل الإصابة بعدوى داء المقوسات 134 (14.4 ٪) في مارس من عام 2013 في حين سجل أدنى معدل 20 عينة إيجابية (2.2 ٪) في نهاية ديسمبر / 2013. كشفت النتائج كانت النسبة المئوية لفئة الأجسام المضادة IgG 148 عينة (19.83٪) موجبة بينما

كانت النسبة المئوية لعينة IgM من فئة الأجسام المضادة 281 (30.28٪) عينة إيجابية.
يتراوح عمر النساء المجهضات في تلك الدراسة بين 12-48 سنة ، وبينت النتائج ان نسبة (86 ٪) من النساء المجهضات كانت إيجابية المصل ل. gondii في سن (12-35) سنة ، (12 ٪) عينة إيجابية المصل في سن (36-40) سنة ، و (2 ٪) عينة إيجابية المصل في الفئة العمرية (41-48) سنة ، وأظهرت الدراسة فروق ذات دلالة إحصائية عالية بين الفئات العمرية للإجهاض عند ($p> 0.05$).

Introduction

Toxoplasmosis is a parasitic disease caused by the protozoan Toxoplasma gondii.This parasitic protozoan is one of the most widespread parasites with a broad geographic range that is nearly worldwide and a broad host range including many birds and mammals [1and 2].The humeral immune response is characterized by the production of anti-Toxoplasma antibodies (IgM and IgG),which aid in determining if the infection is acute or chronic [3]. In both cases rapid and accurate diagnosis is required .

Epidemiology It has been estimated that about one third of the world population is infected with T .gondii [4].Environmental conditions, differences in the type of food consumed , animal species used in the food industry , and the number of cats are examples of factors that may influence the spread of parasite. Water- born transmission of T. gondii has earlier been considered uncommon ,human outbreaks connected to water reservoirs have been reported [5]. In Iraq toxoplasmosis was recorded by many studies [6 ; 7; 8; 9 ; 10 and 11] have reported different prevalence rates in different regions at Al- Nasiriya city.

The aims of this study was conduct to determinate the prevalence rates of T. gondii among aborted women of AL-Shatrah hospital at Al-Nasiriya city , by using ELISA and LAT/MLAT to detect the antibodies (IgG and IgM) against this parasite through the period of the study.

Materials and Methods

The data collection continued for one year from 01- January – 2013 until 31- December - 2013. Patients diagnosis depend on the presence of specific IgM and IgG anti-Toxoplasma antibodies by using ELISA kits (BioChik Diagnostics Company ,USA) according to the manufacturer's instructions

Samples collection :

Five ml of venous blood sample was obtained from 928 aborted women under sterile condition . For serum collection the tubes were let to stand for half to one hour at room temperature, and then centrifugation was done at 3000 rpm for 5 minutes. Then the serum was collected into Eppendorf tubes and was kept at -20° C until use. Freezing and thawing of the sera was avoided and writing the descriptive sample sequence on the tube was performed.

Determination of IgG and IgM Toxoplasma antibodies

For the qualitative and quantitative detection of IgM and IgG *Toxoplasma gondii* antibodies in serum samples of aborted and healthy pregnant and non-pregnant women used two types kits *Toxoplasma* IgM ELISA (Enzyme Linked Immuno Sorbent Assay) Test Kit and *Toxoplasma* IgG EIA (Enzyme Immuno Assay)Test Kit according manufactured.

Statistical analysis

Statistical analysis was performed with SPSS software package (Version 12) for chi-square.A probability value of $P<0.05$ indicated a statistically significant difference [12] .

Results

The results of this study revealed that aborted women serum samples which were collected from AL- Shatraa hospital were high rates of infection by toxoplasmosis at year 2013 . This results explained the numbers of women examined in that year was seropositive for toxoplasmosis of 928 samples recorded from the January of this year which was 132 (14.2 %) samples while 20 (2.2%) samples in the December of this year as shown in table (1).The high rate of infection by toxoplasmosis was 134 (14.4 %) in March of 2013 followed by 132 (14.2 %) in the January of 2013 while the low rate record 20 (2.2%) positive samples in the December of 2013 figure (1).

Table (1): Frequency distribution of toxoplasmosis infection in women with a history of abortion in 2013 of AL- Shatraa hospital .

Months (2013)	N	No. of samples	% of Total N	% of Total samples
January	7	132.00	16.3	14.2
February	3	92.00	7.0%	9.9
March	4	134.00	9.3%	14.4
April	3	70.00	7.0%	7.5%
May	3	79.00	7.0%	8.5%
June	3	58.00	7.0%	6.2%
July	6	93.00	14.0%	10.0%
August	4	86.00	9.3%	9.3%
September	5	72.00	11.6%	7.8%
October	3	69.00	7.0%	7.4%
November	1	23.00	2.3%	2.5%
December	1	20.00	2.3%	2.2%
Total	43	928.00	100.0%	100.0%
Statistical analysis	Chi-square =16.24 , df=11 .p< 0.05			

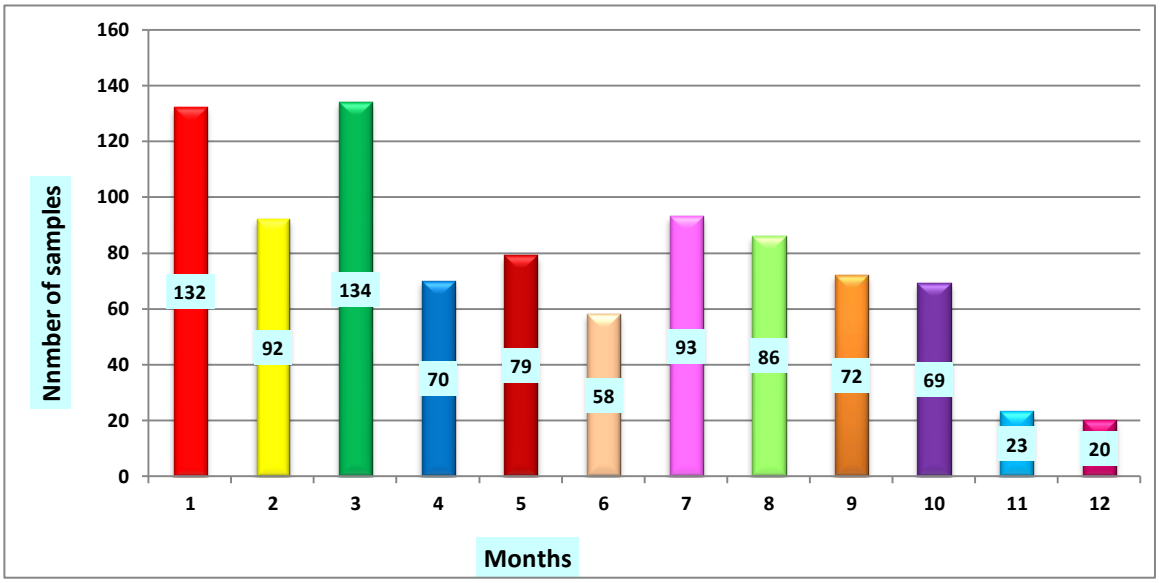

Figure (1): Distribution of toxoplasmosis infection in aborted women in (2013)of AL- Shatraa hospital

In 928 seropositive women the results revealed the percentage of positive sample antibodies class IgG 184 (19.83%), while the percentage of negative sample of IgG antibody was 744 (80.17%) as shown in table (2).

Table (2): Frequency of T. gondii IgG antibody in (2013) of AL- Shatraa hospital .

Months (2013)	No. of samples	IgG+	% For positive	% For negative
January	132.00	24.00	18.18182	81.81818
February	92.00	13.00	14.13043	85.86957
March	134.00	31.00	23.13433	76.86567
April	70.00	22.00	31.42857	68.57143
May	79.00	18.00	22.78481	77.21519
June	58.00	8.00	13.7931	86.2069
July	93.00	15.00	16.12903	83.87097
August	86.00	20.00	23.25581	76.74419
September	72.00	15.00	20.83333	79.16667
October	69.00	13.00	18.84058	81.15942
November	23.00	3.00	13.04348	86.95652
December	20.00	2.00	10	90
Total	**928.00**	**184.00**	**19.83**	**80.17**
Statistical analysis	Chi-square =16.24 , df=11 .p> 0.05			

In 928 seropositive women the result showed the percentage of antibodies class IgM were 281(30.28%) positive samples , while the percentage of negative sample of IgG antibody was 647 (69.72%) as shown in table (3) .

Table (3): Frequency of T. gondii IgM antibody in 2013 of AL- Shatraa hospital .

Months (2013)	No. of samples	IgM+	% For positive	% For negative
January	132.00	52.00	39.39	60.61
February	92.00	34.00	36.96	63.04
March	134.00	29.00	21.64	78.36
April	70.00	37.00	52.86	47.14
May	79.00	8.00	10.13	89.87
June	58.00	11.00	18.97	81.03
July	93.00	30.00	32.26	67.74
August	86.00	26.00	30.23	69.77
September	72.00	28.00	38.89	61.11
October	69.00	9.00	13.04	86.96
November	23.00	7.00	30.43	69.57
December	20.00	10.00	50.00	50.00
Total	928.00	281.00	30.28	69.72
Statistical analysis	Chi-square =16.24 , df=11 .p> 0.05			

The ages among aborted women in this study was between 12-48 years, the women were divided in to three groups according to their age. The results showed 86% were seropositive for *T . gondii* at age of (12-35) years , (**12**%) seropositive sample at age (36-40) years , and (**2**%) seropositive sample at age group (41 - 48) years .

This study showed high significant differences between age groups of aborted women at (p>0.05) as demonstrated in the figure (2) .

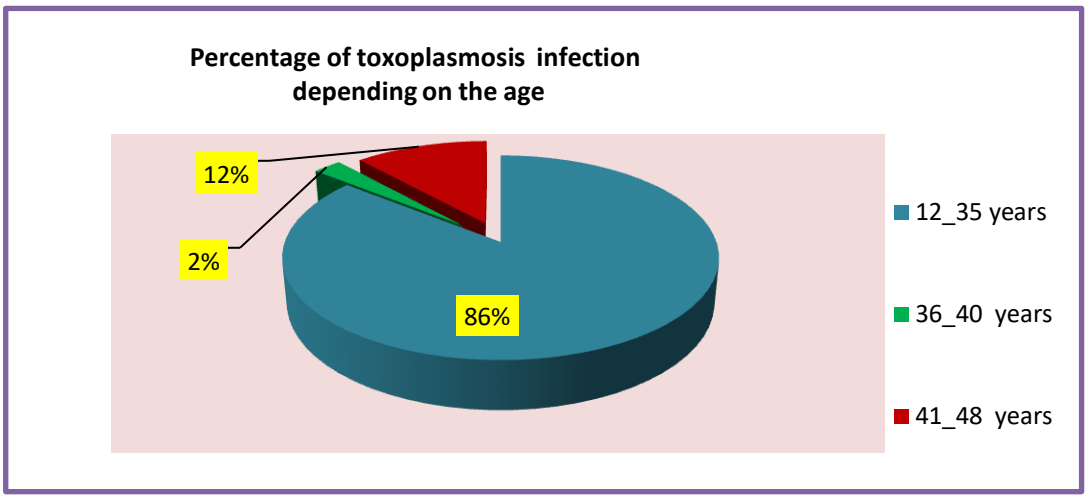

Figure (2): Percentage of Toxoplasmosis infection in women with a history of abortion in AL- Shatraa hospital depending on the ages of patients .

Discussion

The results showed high significant differences between aborted and healthy women at ($p<0.05$).This explains the effect of the parasite *(T.gondii*) on the pregnant women. The high rates of infection by Toxoplasmosis in women in this study is mainly may be acquired by ingestion of undercooked or raw meat containing viable tissue cyst or by ingestion of food or water that contaminated with oocyst shed by cats such as similar study [13] .

The reasons of these results may be due to the fact that women infected with Toxoplasmosis during early ages, so that the immune system will respond to the infection by formation of anti-*T. gondii* antibodies IgM and IgG. The IgG antibody will persist for several months or for life in some patients and small titter of IgM may remain in blood as residual as demonstrated in the similar study [14] .

This differences in seroprevelance between results of this study and other related studies may be due to the seroprevalence estimated for human populations which vary greatly among different countries , different geographical areas within one country , and among ethnic groups living in the certain areas [15] , or may be attributed to several other factors including cultural level , nutritional habits, age or rural and urban areas [16] .

These results were in agreement with several previous studies from different regions [17 and 18], that indicated the percentage of anti-*T . gondii* antibodies was a higher with the increase of age group in Iraq While the results disagrees with the studies that revealed the higher percentage of seropositive women to *T. gondii* between (21-25) years age group [19].

The results also differed with other studies who observed that the main age of seropositive for Toxoplasmosis cases are between (11-20) years old [20; 21] who concluded that the main age of seropositive of Toxoplasmosis cases were between (20-30) years old .

The high occurrence of Toxoplasmosis with this age may be due to marriages of this age group, which is considered the age of higher fertility so we found that most of pregnant women were young 16-19 years old. However ,when IgG antibodies against *T. gondii* were present in the blood they indicate that *T.gondii* cyst were already present in the tissue [22] .

References

1. Singh , S. (2003) . Mother-to-child transmission and diagnosis of *Toxoplasma gondii* infection during pregnancy. India . J. Med . Microbial. 21 (2):69-76 .
2. Jones , J. L. and Dubey. J. P.(2010) .Water borne toxoplasmosis Recent developments . Experi . Parasitol . 124:10-25 .
3. Li, S . ; Galven .G. and Araujo .F. G .(2000). Serodiagnosis of recently acquired *Toxoplasma gondii* infection using an Enzyme Linked Immunosorbent Assay with a combination of recombinant antigens . Clin . Diag . Lab . Immunol . 7:781-787 .
4. Hill ,D. E. and Chirukandoth. S . (2005). Biology and epidemiology of *Toxoplasma gondii* in man and animals . Anima . Health . Res Rev . 6:41-61 .
5. Hill, D . and Dubey .P. (2002). *Toxoplasma gondii*: transmission , diagnosis and prevention . Clin Microbial Infect. 8: 684-690 .
6. Niazi , A . D . ; Nasiaf .W . M .; Abbas .A . and Gzar. S . F . (1992). Prevalence of *Toxoplasma* antibodies in Iraqi population . J . Fao. Med . (Bagh.) 34:255-261 .
7. AL-Sabbak , M . (1999) . The rate of affected toxoplasmosis out come on pregnancy . Basra . J . Soc. 51:58-92 .

8. Abdul-Ridha, R. A. H. (2000). Biochemical changes in the aborted toxoplasmosis seropositive women .M.Sc. Thesis. College of Science. AL-Mustansiriyah University . pp 70.
9. AL-Deen, M . M. (2002) . Seroepidemiological study on toxoplasmosis in women with history of abortion ,. M.Sc. Thesis College of Medicine. AL-Nahrain University . pp 139
10. Kareem, S. S. (2008) . Prevalence, serodiagnosis and some immunological aspects of toxoplasmosis among women in Baghdad province . M.Sc. Thesis .College of health and Medical Technology. Technical Foundation. (Citied by AL-Shikhly, 2010)
11. AL-Shikhly , M . A . H .(2010) . Early detection of toxoplasmosis percentage in pre-marital females by immunological methods . M.Sc. Thesis . College of Science .Baghdad University .pp 147
12. SPSS for Windows, Rel. 11.0.1 (2001) SPSS Inc., Chicago Thompson RCA
13. Fabio , A.; Jose .E .;Vidal .A.; Penalva .D.; Francisco .B.; Roberta .S. and Roberto .F. (2005). Diagnosis of cerebral toxoplasmosis in ADIS patient in Brazil :Importance of Molecular and Immunological Methods using peripheral blood samples. J . Clinical Microbial . 39: 5044-5047 .
14. Giannoulis, C. ; Zournatzi .B.; Giomisi A .; Diza .E. and Tzafettas .I. (2008) . Toxoplasmosis during pregnancy: a case report and review of the literature , 2nd Department of Obstetrics and Gynecology, Aristotle University of Thessaloniki Greece . 3:139-143 .
15. Tenter, A . M . ; Heckeroth .A . R . and Weiss .L . M .(2000) . *Toxoplasma gondii* : from animals to humans . Int. J. Parasitol. 30:1217-58
16. Etheredge , G. D. and J. K . Frankel (1995) . Human *Toxoplasma* infection in Kuna and Embera children in the Bayano and San Blas , eastern Panama . Am . J. Trop . Med . Hyg . 53:448-457 .
17. Karem , L. O. M .(2007). Serological study of *Toxoplasma gondii* antibody from aborted women in Sulaimania city by using ELISA and Minividas test .M.Sc. Thesis . College of Science University of Baghdad .pp 126 (In Arabic).
18. Khalil , H . I . (2008) . Som aspects in seroprevalence , diagnosis and influence of sex hormones on immunity during human toxoplasmosis.Ph.D.Thesis.College of Medicine.AL-Mustansiriyah University . pp 173.
19. AL-Dujaily, K. Y. (1998). Seroepidemiological study of toxoplasmosis among aborted women in Baghdad. M.Sc. Thesis. College of Veterinary Medicine . Baghdad University. (Citied by Karem , 2007).
20. Abdullah. A. (2001). Toxoplasmosis in Mosul-Iraq. J. SC. AL-Rafidain . 12(2):1-4.
21. AL-Rawi , K . H . Z . (2009) . Detection of B1 gene from blood of pregnant and aborted women infected with *T . gondii* . Ph. D. Thesis. College of Science . Baghdad University . pp 125.
22. Dupon, M.;Cazenave .J.; Pellegrin .J.; Ragnaud .J.;Cheyron .A.; Fischer .I.; Leng .B. and Lact .J. (1995). Detection of *T. gondii* by PCR and tissue culture in cerebrospinal fluid and blood of human immunodeficiency virus-seropositive patient . J. Clin. Microbial . 33(9): 2421-2426 .