

Evaluation of Some Serological Study in Pregnant Women Infect With Toxoplasma Gondii

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Abstract:

Toxoplasmosis is an endemic infectious disease, very widely distributed in all parts of the world and called a disease of the five continents and depending on the geographical location since 15% to 85% of the human population asymptotically infected. The present study was carried out on total number one hundred and thirty three women suspected of having toxoplasmosis with and without abortion. The titers of serum immunoglobulin M and G (IgM and IgG) of anti- Toxoplasma antibodies were detected by enzyme linked immunosorbent assay (ELISA) according to the final diagnosis of toxoplasmosis. It was found that women with toxoplasmosis had higher levels of median values of IgM and IgG Abs of T.gondii that were 0.420 ± 0.035 and 5.461 ± 0.39 respectively, while women that did not have toxoplasmosis, had 0.161 ± 0.019 and 0.159 ± 0.018 respectively . Serum levels of IL-18 and IL-5 detected by ELISA were 87.01 ± 10.02 pg/ml and 805.70 ± 122.30 pg/ml respectively in women with toxoplasmosis , while in women that did not have toxoplasmosis the levels were 67.59 ± 10.21 pg/ml and 126.62 ± 35.96 pg/ml respectively .

Introduction :

Toxoplasmosis is a zoonotic disease of animals caused by the protozoan parasite Toxoplasma gondii, human and other warm blooded animals are its hosts [1].

The infection has a world wide distribution. Approximately one –third of all humanity has been exposed to this parasite, Toxoplasmosis has a wide range of prevalence and this variability is related to various factors such as, age, sociocultural and nutritional habits, contact with domestic cats, climatic and geographical conditions [2]. The name Toxoplasma (toxon = arc, plasma = form) is derived from crescent shape. T.gondii was

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first discovered by Nicolle and Manceaux in 1908 in small North African rodent *Ctenodactylus gundii*. At about the same time Splendore (1908) independently described *Toxoplasma* in a laboratory rabbit in Sao Paulo, Brazil [3]. Then *Toxoplasma gondii* was described by (Nicolle and Manceaux) in 1909 as an intestinal coccidium of felids with a usually wide range of intermediate hosts , infection by this parasite is prevalent in many warm –blooded animals including humans [4].

During the first few weeks post-exposure, the infection typically causes a mild flu-like illness or no illness. Thereafter, the parasite rarely causes any symptoms in otherwise healthy adults. However, those with a weakened immune system, such as pregnant women, may become seriously ill, and it can occasionally be fatal. The parasite can cause encephalitis (inflammation of the brain) and neurologic diseases, and can affect the heart, liver, inner ears, and eyes (chorioretinitis) .

In addition patients with congenital infection, since 5- 24% of children becoming ill and dying during neonatal period [5]. Primary acquired infection during pregnancy may cause miscarriage, permanent neurological damage, premature birth and visual impairment [6]. It is well established that *T.gondii* infection induce strong cell-mediated immune response type-1- cytokines such as gamma interferon (IFN- γ), interleukin-12 (IL-12) and tumor necrosis factor- α (TNF- α) are crucial in protective immunity, while the type-2- cytokines IL-5 is homodimeric glycoprotein produced predominantly by activated cell surface molecules (CD-4) T-cells [7]. since IL-5 enhances B-cell - IL-12 receptor expression and promotes B- cell proliferation and differentiation . Also, IL-5 have protective role against *T.gondii* and may play role in the production of IL-12 . IL-5 has also essential role for production and function of eosinophils and serves as an anti apoptotic factor for the latter cells [8].

Materials and

Instruments:

Samples Collection has been from 197 women doubtful toxoplasmosis collected in Baghdad city from Ibn-AL- Balady hospital and Fatima- AL- Zahraa hospital during the period from October 2013 to February 2014.

Group I: Patients The study included 70 women tests proved they are infected toxoplasmosis and women aged between 18-45 years have been diagnosed with infection through laboratory testing , which included screening for antibodies specific IgG, IgM technique using Enzyme Linked Immunosorbent Assay

Group II: control (healthy) Group: which were collected 20 blood samples Other women in the age group 18-45 years has been making sure not to infect

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with toxoplasmosis . Sample of venous blood was collected from these women for serum collection.

This study was carried out to asses the presence of Anti-Toxoplasma gondiiIgG and IgM. ELISA was use for detection of the antibodies in serum samples using commercial kits, (Bio Kit,Spain).

ELISA was performed by the use of two kits (Omega Diagnostica company,Scotland).Human IL- 5 kit by ELISA Cusabio (china) and Human IL- 18kit by ELISA Cusabio (china).

Results & Discussion

Serum levels of IgG in patients women with Toxoplasmosis infections

IgG levels were assayed by ELISA method in the sera of the patients women with toxoplasmosis infections and compared to the healthy control group. Result showed a statistically significant elevation in the concentration of the IgG in the sera of infected patients women in comparison to those of healthy control group. The concentrations of IgG were significantly elevated ($P<0.05$) the levels were(mean \pm SD) 5.461 ± 0.39 IU /ml in comparison to healthy control group 0.159 ± 0.018 IU /ml.

Serum levels of IgM in patients women with Toxoplasmosis infections IgM levels were assayed by ELISA method in the sera of the patients women with toxoplasmosis infections and compared to the healthy control group. Result showed a statistically significant elevation in the concentration of the IgM in the sera of infected patients women in comparison to those of healthy control group. The concentrations of IgM were significantly elevated ($P<0.05$) the levels were (mean \pm SD) 0.420 ± 0.035 ./ml in comparison to healthy control group 0.161 ± 0.019 .

Table 1. Compare between patients and control in IgG and IgM

Group	No.	Mean \pm SE	
		IgG	IgM
Patients	70	5.461 ± 0.39	0.420 ± 0.035
Control	20	0.159 ± 0.018	0.161 ± 0.019
LSD Value	---	1.463 *	0.134 *
* ($P<0.05$).			

The present results indicated that there was an increase in the titer of IgG and IgM antibody in the sera of infected women in comparison with the titer of these antibodies in control sera, that mean there is a defined role of IgG and IgM during the infection with T. gondii which is insured by [9]. who was revealed that the immunoglobulins belonging to class IgG, IgM, IgA and IgE is produced in response to infection. This finding is also similar with the results of [10]. in indicating the

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association between the infection and strong humoral response involving IgM, IgG, IgA and IgE. [11] noted that B-cells are required for vaccination – induced resistance to virulent tachyzoites. In addition to these finding, the presence of high level of IgG indicate that the person has had toxoplasmosis at some time in their life (Internet) because IgG can persist for many decades and is, therefore, not an indicator of recent infection [12] . Also raised IgM may indicate a current or recent infection because this immunoglobulin typically persist for(6- 9) months after infection and is helpful in diagnosing acute infection [13].

effect of infection toxoplasmosis in the level of IL-18, IL-5

The results suggest that reached from measuring the concentration IL-5 in ELISA'S way for the group infected(70 infected) to obvious raise in concentration rate of the interleukin for this group($805.70 \pm 122.30\text{pg/ml}$) compared to what it is in the control group($126.62 \pm 35.96\text{pg/ml}$) which counted(20) so this increase records significant difference($p<0.05$) as shown in the table(3-2) this corresponds to what (Zhang and Denkers,1999) and(Matowicka-Karna et al,2005) and(Lang et al.,2007) and(Joanna et al.,2009) and (Suha,2010) reached that toxoplasmosis infection results increase in IL-5 level. IL-5 produced from Th2 cells and play's an important role in immunological response where it work's on differentiation of acidic cells plus it increases Cytotoxic activity in collaboration with TNF-a, and also work's on differentiation and growing lymphocytes .B. kind(David et al.,2008) and activate gene expression of antibody(Murray et al.,1987) which work's to protect mucosal surface which is the entry of the parasite sites(Yap&Sher,1999) and (Filisetti et al.,2004) have confirmed that IL-5 urges to increase the production of IL-12 where it work's(the last one) to stimulate the NK cells for the production of motor cell IFN-y which work's with TNF-a to induce gene expression to produce Nitric Oxide(NO) which work's to kill parasite inside the cell(Robert et al.,2001) and also the table (3-2) shows results that have been reached in the measurement of the concentration of IL-18 in ELISA way for the patient group, which point to a raise in the rate of interleukin this group($87.01 \pm 10.02\text{pg/ml}$)compared to what it is in the control group($67.59 \pm 10.21\text{pg/ml}$) however this increase did not occur significant changes($P<0.05$)and this is consistent with what(Bhopale , 2002) reached, who stressed the role of IL-18 during the acute and chronic phases, where this interleukin stimulates the production of IFN-y which urges to produce (NO) capable of killing the parasite inside the cell.

Table(3-2): Concentration of level IL-5 and IL-18 in the studied group.

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Group	No.	Mean \pm SE	
		IL-18	IL-5
Patients	70	87.01 \pm 10.02	805.70 \pm 122.30
Control	20	67.59 \pm 10.21	126.62 \pm 35.96
LSD Value	---	38.921 NS	458.11 *
NS: Non-significant. * (P<0.05)			
*: significant			

References

- [1] Steven, E., Schmitt, B., Golovko, A., Mehdi, E. and Santanu, K.(2008): Toxoplasmosis. Chapter 2.9.10. In: Barry, O.N.(ed.).Terrestrial Manual. OIE Scientific Publications, 6th ed.
- [2] Ichhpujani, R.L.; Bhatia, R.(eds.).(1994): Toxoplasmosis. p.144-151.In: Medical Parasitology.(1st ed.). Jaypee Brothers Medical Publisher(P)Ltd.India .
- [3] Dubey, JP.(1993): Toxoplasma, Neoplasma, Sarcocystis, and other tissue cyst-forming coccidian of human and animals.pp.1-156. In :Kreier, P.J.(ed.) Parasitic protozoa ,vol.62nd ed. Academic Press, Inc., San Diego, California .
- [4] CDCP: Centers for Disease Control and Prevention.(2002);Guidelines for the Control of Sexually Transmitted Diseases. Morb. and Mort. Weekly Report.: 51(No.RR-6).
- [5] Singh, S.(2003): Mother to child transmission and diagnosis of Toxoplasma gondii infection during pregnancy. Indian. J. Med. Microbiol. ;21(2):69-76.
- [6] Frenkel, K.J.(1973): Toxoplasma in and around us.Biol.Sc.;23: 343-352.
- [7] Karlen, S., DeBoer, M.L., Lipscombe, R.J., Lutz, W., Mordvinov, V.A. and Sanderson, C. J.(1998): Biological and molecular characteristics of interleukin-5 and its receptor. Int. Rev.Immunol.;16:227-247.
- [8] Zhang ,Y. and Denkers, E. Y.(1999): Protective role for Interleukin-5 during chronic Toxoplasma gondii infection. Infect. Immun.; 67(9):4383- 4392.
- [9] Mandell, J. L.; Bennett, J. E. and Dolin,R. (2000). Fth ed. Vol. 2. Principles and practice of infectious diseases. Churchill. Livingstone.
- [10] Chardes, T.; Bourguin, I. And Mevelce, Mavie-Noelle (1990). Antibody responses to Toxoplasma gondii : in sera, intestinal secretions and milk from infected mice and characterization of target antigens. Infect. and immun., 58 (5) : 1240 – 1246.
- [11] Sayles, P. C.; Gibson, G. W. and Johnson,L. L. (2000). B-cells are essential for vaccination – induced resistance to virulent Toxoplasma gondii. Infect. and immun., 68 (3): 1026 – 1033.
- [12] Internet (1997) . Drinking water inspectorate fact sheet . Toxoplasma gondii . Fact sheet No. 15: issue No.1 March (1997) .
- [13] Zhang ,Y. & Denkers , E. Y. (1999). Protective role for interleukine -5 during chronic Toxoplasma gondii infection . J . Inect . Immun. 67(9) : 4383-

Evaluation of Some Serological Study in Pregnant Women Infect With Toxoplasma Gondii Dr. Bedir. M. AL - Azawi , Ola Hawwal AL-Timeme

4392.

- [14] **Matowicka** - Karna , J. ; Kemoni , H. & Dymika-Piekarska , V. (2005) . Some parameters of immune response in patients infected with Toxoplasma gondii .Pol. Merkur Lekarski .; 18(103):85-7.
- [15] **Joanna**, M. ; Violetta , D.P. & Halina , K . (2009). Dose Toxoplasma gondii infection Affect the level of IgE and cytokines (IL-5,IL-6,IL-10, TNF- α . J. Clin . Immunol . V(2009) : p 4 .
- [16] **Suha** , A. A. (2010) . Laboratory Diagnostic Evaluation of Toxoplasmosis in Abortion women from Baghdad. Degree of Doctor .Thesis .College of Medicin . baghdad University .
- [17] **David**, F.L.; Jason, S . S.; Andew , E .G. ; Adeeb , H .R.; Devon,K.T.; Christopher,A.H. & Laurence ,A.T.(2008): T cell expression of my D88 is required for resistance to Toxoplasma gondii.(<http://www.pnas.org/content/105/10/3855.full>).
- [18] **Murray**, PD. ; Mckenzie , DT.; Swain ,SL. & Kagnoff ,MF.(1987) : Interleukin 5 and interleukin 4 produced by peyers patch T cells selectively enhance immunoglobulin A expression .J. Immun. 139(8): 2669-2674.
- [19] **Yap** , G. ; Sher , A . (1999). Cell mediated immunity to Toxoplasma gondii: initiation regulation and effector function. Immunol .; 201: 240-247.
- [20] **Filisetti** ,D. & Candolfi , E. (2004). Immune response to Toxoplasma gondii. Ann Ist Super Sanita. 40(1) : 71-80 .
- [20] **Robert**, C. ; Walker ,W. & Alexander , J .(2001).Sex Associated hormone and immunity to protozoan parasites . J . Clin . Microbiol . Rev. 14(3) 476-488.
- [21] **Bhopale** , G. M. (2002) . Pathogenesis of toxoplasmosis. J . Immun . 26(4): pp 213-222.

دراسة مناعية للنساء الحوامل المصابات بداء المقوسات

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المستخلص

داء المقوسات (داء القطط) مرض معدي مستوطن وواسع الانتشار في كل أنحاء العالم يدعى ((مرض القارات الخمس)) ويتسبب عن الإصابة بطفيلي التوكسوبلازما كوندوي T.gondii، ويعتمد انتشاره على الموقع الجغرافي حيث ان هناك 15%-85% من المجتمع البشري مصابون بالمرض وبدون أعراض. جربت الدراسة الحالية على 133 مريضة مشكوك بإصابتهم بداء القطط لديهن واللاتي يعانين من حالات الاجهاض او بدونه. معيارية الاضداد نوع M و G لطفيلي التوكسوبلازما تم التحري عنها في المصل بطريقة الامتصاص المناعي المرتبط الانزيمي ال ELISA ، حيث وجد ان النساء المصابات بداء المقوسات لديهن أعلى قيم متوسطة من الاضداد M و G والتي بلغت 0.420 ± 0.035 و 5.461 ± 0.39 . اما النساء غير المصابات والتي بلغت قيمتها 0.161 ± 0.019 و 0.159 ± 0.018 على التوالي. مستويات الانترليوكينات (IL-5 و IL-18) والتي تم التحري عنها بطريقة الامتصاص المناعي المرتبط الانزيمي ال ELISA في مصل النساء المصابات بداء المقوسات وكانت 10.02 ± 87.01 و 122.30 ± 805.70 على التوالي. بينما كانت في النساء غير المصابات بداء المقوسات 10.21 ± 67.59 و 126.62 ± 35.96 على التوالي.

