

EVALUATION OF SOME IMMUNE PARAMETERS FOR PREGNANT WOMEN IN THE FIRST THREE MONTHS WHO ARE INFECTED WITH TOXOPLASMOSIS

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Abstract:

The aims of this study is to determine the prevalence of toxoplasmosis infection in pregnant women during the first three months of pregnancy in Salah Al-Din, Iraq, from August 1, 2023, until February 1 2024. The sample examined by studying 203 sample, 123 samples (60.59%) were positive, and 80 (39.04%) samples were negative for antibodies. The current study examined 90 optical ELISA techniques, The highest percentage of IgM infection was recorded when there were no cats (43.75%), while the lowest percentage of infection was recorded (56.25%). As for the IgG, the highest percentage reached when cats were present (74.66%), and the lowest percentage reached when cats were not present (25.33%). the epidemic it was recorded from the current study that the highest percentage of IgM (81.25%) for infection with *Toxoplasma gondii* was in women who had miscarried, while the percentage decreased to (18.75%) in non-aborted women. As for IgG, the percentage also increased in women who had miscarried (72%). IFN- γ Its concentration increased (20.6 ± 85.9 pg/ml) in infected women while its decreased (22.0 ± 63.8 pg/ml) In uninfected women. CCR5 Its concentration decreased (185.4 ± 19.5 pg/ml) in infected women, while its concentration increased (185.4 ± 19.5 pg/ml). (284.0 ± 28.1 pg/ml) in uninfected women

Keywords: Gondii, *Toxoplasma gondii*, Interferon gamma, chemokine receptor typr5 ,ELISA IgG ,IgM.

Introduction

Toxoplasma gondii is a parasitic protozoon (Protozoa Parasite). This parasite causes toxoplasmosis, or what is known as cat disease, which is one of the diseases shared between humans and animals, in addition to being one of the global diseases with opportunistic spread, especially in individuals. Immunocompromised, while in immunocompetent individuals, it is asymptomatic, and the global infection rate ranges from less than 10% to about 90% depending on the diagnosis of anti-parasite antibodies (Anti-*Toxoplasma* antibodies) in the serum [1]. Toxoplasmosis can have more severe complications and consequences such as birth defects, eye infections, or fatal toxoplasmic encephalitis in immunocompromised individuals [2].



The infection is transmitted to the fetus of a mother infected with *Toxoplasma* for the first-time during pregnancy, and the most serious infection is congenital infection [3]. *Toxoplasma* is classified within the sporozoa, the order Euccoidirida, and the Sercostidae family, under the *Toxoplasma* family. The name refers to its crescent-shaped or arc-shaped shape in Greek and Gondian for a rodent native to North Africa. *Toxoplasma* are parasites that are forced to live inside the cell and are diploid [4].

Humoral immunity plays an important role in acquired resistance against toxoplasmosis, as immunity develops as the infection progresses, and this leads to the formation of immunoglobulin antibodies. The first antibody formed is IgA, which appears for a short period[5], and begins to appear when the parasite enters the intestinal lining and is one of the most important antibodies found in the intestinal lining. It is unstable at the beginning of infection with toxoplasmosis and is considered an indicator of acute infection. In the event that the parasite spreads to the rest of the body, immune globulin begins to appear about 1-2 weeks after infection, and after about 4-8 weeks of infection, IgM begins. It decreases and remains for 6 months while IgG appears after the fourth week of infection in people with complete immunity, IgG levels remain high for several months, and a recent study in the city of Riyadh confirmed that up to 32.5% and 6.4% of pregnant women were positive for *T. gondii* IgG and IgM antibodies on their testicles. respectively [6].

2. Materials and method

2.1. Study Design: The current study was completed in Salah al-Din Governorate and some of its affiliated districts. Samples were collected randomly from pregnant women in the first three months who arrived at government hospitals such as Tikrit Teaching Hospital and some private medical clinics, whose ages ranged between 17 to 39 years.

2.2. Blood samples and Immunological tests

5 ml of venous blood was drawn using a tourniquet and a 5 ml medical syringe equipped with a 23 mm plier. The blood samples were placed in clean plastic plan tubes devoid of any substance, then left for 15-30 minutes, after which they were placed in a centrifuge for 5 minutes/3000 rpm for the purpose of separating the blood serum and kept frozen (20C) until laboratory tests were performed. ELISA test was conducted for 90 samples to detect the infection and determine the concentration of IgG specific antibodies. IgM, and immune indicators (INF-Y, CCR5) were also evaluated using ELISA technology for 60 positive samples and 30 control samples (uninfected women).

2.3. Statistical analysis:

Results were analyzed statistically using analysis of variance test-ANOVA by using the statistical program Minitab. Averages were compared to calculations of the characteristics of the application Duncan's Multiple Range Test by probability level $P \leq 0.05$.



3.Results and Discussion

During the study of 203 samples serum from pregnant women in the first three month using Rapid test, 123 samples (60.59%) were positive for IgM or IgG antibodies, and 80 (39.04%) samples were negative for antibodies showed a positive result for (IgG) (46.29%)75 And (29,625) 48 samples out of the total samples showed the acute infection and it was (IgM) as shown in (Table 1).

Table 1. Diagnosis of the *Toxoplasma gondii* parasite.

Method	Number of samples tested%	+ve %	-ve%
Toxo-IgG/IgM Rapid test kit	203 (100%)	123 (60.59%)	80 (39.04%)

It was observed that there was a variation (Table 2) in the percentages of infection with *Toxoplasma gondii* according to the presence or absence of cats in the home of the infected women under study. The highest percentage of IgM infection was recorded when there were no cats (43.75%), while the lowest percentage of infection was recorded (56.25%). (In the case of the presence of cats in the house. As for the loss of IgG, the highest percentage reached when cats were present (74.66%), and the lowest percentage reached when cats were not present (25.33%). The results of the current study were consistent with the findings of [7],[8], and [9], as they concluded that the highest rate of infection with *Toxoplasma gondii* was among women who owned cats.

What has been shown from the results of the current study and previous studies is that dealing with animals and cats increases the rates of infection with *Toxoplasma gondii*. This is due to the fact that these animals and their waste are considered a fertile medium for the parasite's reproduction and transmission, especially cats and the feline family, as they are the final or intermediate hosts for the parasite and exposure to it and its waste.

This may be due to the fact that cats are among the most important animals that contribute to the occurrence of *Toxoplasma gondii* infection, as they are the final host that sheds the infectious stages represented by egg sacs [10].

Cats can infect themselves by licking the exit area after defecation and when women touch cat fur contaminated with egg sacs (as a result of cats coming into contact with soil contaminated with their feces), and not washing hands is a cause of infection, in addition to the fact that lack of attention to the semen of cat homes plays a major role. In the incidence of toxoplasmosis *gondii*.



Table 2: Percentages of infection with toxoplasmosis gondii according to the presence cat.

Presence of cats	IgM %	IgG %
Yes	21(43.75%)	56(74.66%)
No	27(56.25%)	19(25.33%)
Total	48(100%)	75(100%)

In (Table 3) it was recorded from the current study that the highest percentage of IgM (81.25%) for infection with *Toxoplasma gondii* was in women who had miscarried, while the percentage decreased to (18.75%) in non-aborted women. As for IgG, the percentage also increased in women who had miscarried (72. %) and decreased among non-aborted women (28%). The results converged with [11], where the highest percentage of miscarriages was recorded among women infected with *Toxoplasma gondii* during the first three months of pregnancy, while a lower percentage of miscarriages was recorded during the second and third periods of pregnancy, respectively, and converged with [12] where 17 cases of miscarriage were recorded in women infected with *Toxoplasma gondii* in the first three months of pregnancy.

The cause of miscarriage in the first three months of pregnancy may be due to the placenta being weak at the beginning of pregnancy compared to the advanced stages of pregnancy, so any defect in these stages leads to miscarriages [13]. The *Toxoplasma gondii* parasite works to destroy placental tissue as a result. The process of rapid reproduction of the parasite's individuals and the formation of spaces between the tissues of the placenta as a result of the formation of tissue cysts and that the secretions secreted by the parasite lead to the death of the cells of the tissues that make up the placenta [14].

Table 3: Percentage of infection with toxoplasmosis gondii among abortionists.

Case	IgM%	IgG%
Abortion	39(81.25%)	54(72%)
Not Abortion	9(18.75%)	21(28%)
Total	48 (100%)	75(100%)

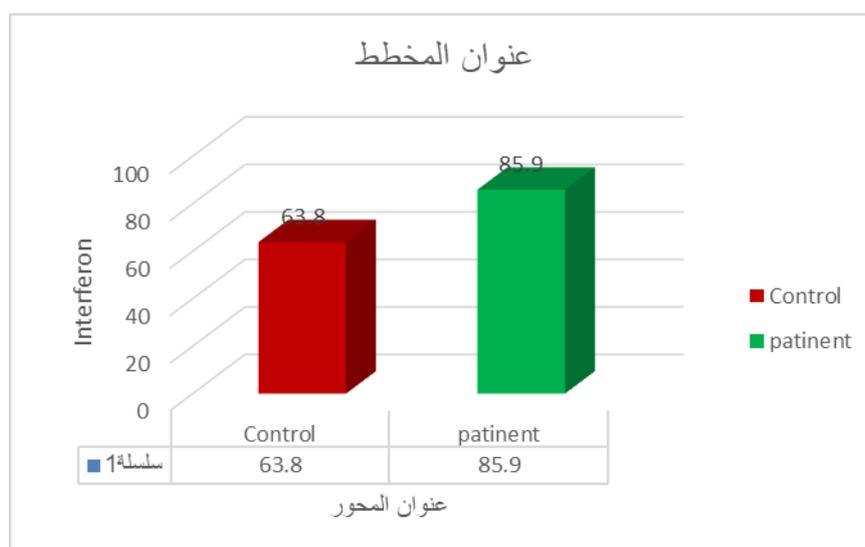
In table 4 There was a significant difference in the concentration of interferon gamma IFN- γ in both women infected and without *Toxoplasma gondii* during pregnancy in the first three months. Its concentration increased (20.6 ± 85.9 pg/ml) in infected women while its concentration decreased (22.0 ± 63.8 pg/ml) In uninfected women, interferon gamma has a role in the immune response against the *Toxoplasma gondii* parasite, as CD4 and CD8 positive T lymphocytes have an important role in the production of INF, and this kinetic has a role in resisting and inhibiting the parasite by stimulating many mechanisms according to the oxygen-dependent mechanism, which produces peroxide. Toxic hydrogen It also secretes Nitric oxide synthase (NOS2) to form nitric oxide, which in turn works to destroy the parasite or transform it from the rapidly reproducing tachyzoite phase to the slow-reproducing bradyzoite phase to



prevent the reactivation of the tissue cyst and works to stimulate macrophage cells to present the antigen. It also increases the effectiveness of lysosomes in macrophage cells and stimulates The effectiveness of NK and inhibits the effectiveness of Th2 helper lymphocytes [15] and also increases the effectiveness of Indoleamine 2,3-dioxygenase, which works to break down the amino acid tryptophan, which is important for the survival of the parasite. The results agreed with [16] and [17], who confirmed an increase in the level of interferon gamma IFN- γ compared to the control group through their study that they conducted in Iraq. Despite the importance of interferon gamma in resisting parasitic infection, the production Too much of it and an uncontrolled response may contribute to causing diseases in the host. The host needs a balancing act in the immune response between reducing damage and eliminating pathogens that infect the host [18].

Table 4: Interferon gamma IFN- γ kinetics in women with toxoplasmosis gondii.

Group	IFN- γ (pg/ml) Mean \pm S.E	P-Value
Patient	85.9 \pm 20.6	0.0001 **
Control	62.8 \pm 22.0	



The results of the current study (Table 5) recorded that there was no significant difference in the chemokine CCR5 in both women infected and without Toxoplasma gondii during pregnancy in the first three months. Its concentration decreased (185.4 \pm 19.5 pg/ml) in infected women, while its concentration increased (185.4 \pm 19.5 pg/ml). /ml 284.0 \pm 28.1) in uninfected women. This indicates that there are no significant differences, as $P \leq 0.05$. CCR5 is a G protein-coupled receptor (GPCR) that mediates the migration and activation of immune cells in response to the chemokines CCL3, CCL4, and CCL5 [19] There are other chemokine receptors that work together, CCR5, to stimulate T cells and release cytokines from CD4+ T cells [20]. During inflammation, the level of CCR5 expression in CD8 cells is regulated, allowing the cells to move towards the sites of CD4+ T cells and stem cells [21], this increases the chance



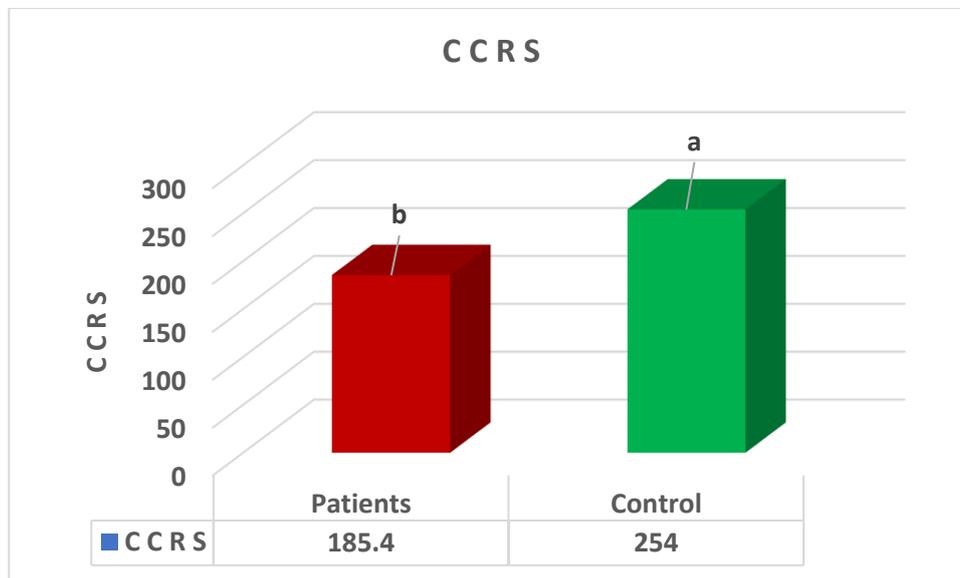
that CD8+ cells will encounter antigen-specific cells and thus, CCR5 enhances the adaptive immune response.

During *T. gondii* infection, TLRs and Chemokine receptors may work together to activate APC and produce IL-12, thus determining the outcome of the infection [22]. The molecular production of IL-12 comes from the interaction between chemokines such as CCL3, CCL4, and CCL5 that it produces. Intestinal epithelial cells after *Toxoplasma gondii* infection with CCR5 [23] These chemokines and chemokine-like molecules can stimulate the migration of CD8+ lymphocytes to the intraepithelial region of the small intestine by interacting with CCR5, and this may be a necessary signal. To control the exacerbated inflammatory response caused by the parasite through the production of TGF-β [24].

Furthermore, in other disease models, CCR5 also modulates the migratory and suppressive function of regulatory T cells and may interfere in pathogen control or regulation of the immune response [25].

Table 5: chemokine Receptor Type5 in women with toxoplasmosis gondii.

Group	CCR5 (pg/ml) Mean±S.E	P- Value
Patient	185.4±19.5	0.055
Control	284.0±28.1	*



Conclusion:

increase in the concentrations of the level of IgG immune antibodies in the blood serum of samples of the group infected with toxoplasmosis, 0.966±0.249 UI/ml, the presence of an increase in the level of gamma interferon in the blood serum of women infected with



toxoplasmosis, and the presence of a decrease in the level of IgM in the blood serum of the samples. Women with toxoplasmosis 0.339 ± 0.135 UI/ml compared to the control group.

The current study found a non-significant effect of toxoplasmosis infection among aborted pregnant women on the level of CCR5.

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