

## STUDIES ON THE EFFECT OF INFECTED CASES OF PARASITE *TOXOPLASMA GONDII* ON SOME SEX HORMONES

Dr. Bedir. M. AL Azawi and Farah M. Shtaib AL-Waeli

Biology Department, College of Science, University of AL-Mustansiriyah.

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### \*Correspondence for

#### Author

Dr. Farah M. Shtaib AL-  
Waeli

Biology Department,  
College of Science,  
University of AL-  
Mustansiriyah.

### ABSTRACT

Toxoplasmosis is considered to be an epidemic disease which spreads very widely all over the world. The cause of it is the infection by the parasite *Toxoplasma gondii*. Its spread depends on the sociocultural and nutritional habits, climatic and geographical condition. The present study was conducted on 130 women infected with toxoplasmosis and who was suffering from abortion or without it. It was found that 90 patients who were sent by the specialist to Fatima- AL- Zahraa Hospital and Ibn- AL- Balady Hospital in Baghdad had antibodies against toxoplasmosis parasite. And 90 men infected by toxoplasmosis from National Blood Transfusion Center their ages ranged between 15-45 years for the period from December 2014 to March 2015 Parallel to that, 60 healthy women and 30 healthy men were chosen as a control

group. The results showed that the percentage of infections by the parasite for women and men were 69.23% and 66.67% respectively using ELISA and Agglutination methods. The serum concentrations of several hormones were studied such as (Estradiol, progesterone and Prolactin). The results of these trials revealed a significant ( $p < 0.05$ ) increase in level of Estradiol hormone in women ( $674.42 \pm 137.78$  ng/ml) and non-significant in infected men ( $501.32 \pm 82.09$  ng/ml) and a significant ( $p < 0.05$ ) decline in progesterone level in infected women ( $7.56 \pm 1.61$  ng/ml) and ( $5.51 \pm 1.25$  ng/ml) in infected men when compared with control. However, no significant differences were noticed in the level of prolactin where it registered significant decrease in infected patients in women and men comparison to control subjects.

**KEYWORDS:** *Toxoplasma gondii*, Estradiol, progesterone and Prolactin.

## INTRODUCTION

Toxoplasmosis is a zoonotic disease caused by the protozoan parasite *Toxoplasma gondii*, human and other warm blooded animals is its hosts.<sup>[1]</sup>

The infection has a worldwide distribution. Approximately one-third of all humanity has been exposed to this parasite, but the seroprevalence varies considerably between countries (from less than 10% to more than 90%) and population group.<sup>[2]</sup>

All mammals, including humans, and birds are intermediate hosts, whereas Felidae (cats) are intermediate and definitive host, they are the only animals that pass oocyst in their feces. Sheep and goat meats are important infection sources of toxoplasmosis.<sup>[3]</sup>

The parasite is transmitted to humans mainly by contaminated food and drinks.<sup>[4]</sup> Claudia and his colleagues confirmed in (2010) 43% of those infected because of direct exposure or indirect of oocyte and 57% by eating meat containing oocyst, Kasper explained in (1998) that eating one mature Oocyst able to cause disease in humans, as well as the blood transfusion a means of transmitting the disease and it is the same in the operations of organ transplant and tissue infected to healthy people who are given drugs immunosuppressive which play an important role in for the disease.<sup>[5]</sup>

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.

Primary acquired infection during pregnancy may cause miscarriage, permanent neurological damage, premature birth and visual impairment.<sup>[6]</sup>

Lower cellular immunity, which is associated with high levels of steroid hormones contributes to the survival of the parasite in the body.

Such these people, cause of steroid hormones increase with weak immune system.<sup>[7]</sup> Other reports indicate that *Toxoplasma* can increase the number of sons, height in infected men and change personality factors in men and women.<sup>[8,9]</sup> It is likely that sexual hormone changes can play an important certain role in relation with *Toxoplasma* and a fore mentioned phenomena.

Sex differences in infection are mediated, in part, by the effects of androgens as an example, testosterone, the steroid hormone that is the predominant circulating in the bloodstream and is produced by Leydig cells in the testicles, and plays an important role in normal development of male characteristics and demonstrate the male Low levels of testosterone occur slightly with age It promotes the development of secondary sexual, Characteristics in men with facial hair growth in the axilla, and scrotum, the growth and development of the prostate in men, the cartilage growth of the larynx and the depth of the sound, causing the baldness.<sup>[10]</sup>

## **MATERIALS AND METHODS**

### **A- Collection of samples**

Samples collected from 220 women and men infected with toxoplasmosis collected in Baghdad city from Fatima - AL- Zahraa Hospital, Ibn- AL- Balady Hospital and National Blood Transfusion Center during the period from December 2014 to March 2015.

#### **The group I**

##### **Patients Group**

This group included 90 women and 60 men that the mastery tests proved they are infected with Toxoplasmosis disease, ranging in age from 15-45 years and have been diagnosed with the disease by: - laboratory test which included screening for antibodies quality IgG, IgM is using technique Enzyme Linked Immunosorbent Assay.

#### **Group II**

##### **Control Group**

This group was used as a control group and that in evaluating the results of the study and to find the acute changes in the percentage of the ingredients serological components required investigation in the patients who under study, another 40 blood samples have been collected for women and 30 for men from ages between 15-45 years that had confirmed negative for toxoplasmosis use as a control group.

Five ml of peripheral venous blood samples had been collected from these women and men.

### **B- Serological analysis**

#### **1- Latex Agglutination test (LAT)**

For determining the presence of anti-Toxoplasma antibodies the latex agglutination test kits. It is a direct agglutination test performed on a card for serodiagnosis of toxoplasmosis.

## 2- Enzyme Linked Immunosorbant Assay (ELISA)

This assay was performed by using two commercial kits for the detection of IgG and IgM anti-*Toxoplasma gondii* antibodies in the serum.

## 3- Cobas e411 (Roch)

This technique is used to measure the proportion of hormones (Estradiol, Progesterone and Prolactin) in the current study and this device works by chemiluminescence technology (fully automated).

## 4- Statistical Analysis

The Statistical Analysis System- SAS (2012),<sup>[11]</sup> was used to effect of different factors in study parameters. Least significant difference –LSD test was used to significant compare between means and chi-square test was used between percentages in this study.

## The results

### 1- Comparison between patients and control in Estradiol in men and women

In Table (1) results showed significant differences in estrogen levels between infected and non-infected women, although high levels of estrogen were noticed in infected women with *T.gondii* compared with non-infected women ( $674.42 \pm 137.78$ ) and ( $237.60 \pm 95.42$ ) respectively. And non-significant between infected and non-infected men ( $501.32 \pm 82.09$ ) ( $398.60 \pm 18.01$ ) respectively.

**Table (1): Compare between patients and control in E<sub>2</sub> (ng/ml) in men and women**

Group	Sex		LSD value
	Men	Women	
Patients	$501.32 \pm 82.09$	$674.42 \pm 137.78$	271.03 NS
Control	$398.60 \pm 18.01$	$237.60 \pm 95.42$	238.55 NS
LSD value	232.74 NS	428.54 *	---

\* (P≤0. 05), NS: nonsignificant.

Estradiol, necessary for the beginning correlation of the blastocyst with progesterone, and induce the progesterone receptor, (IL-6 inhibits).<sup>[12]</sup>

As well as estradiol important in the division Mitogen and stimulate the split granulosa cell process.<sup>[13]</sup> And there during pregnancy immune regulation through the mother to the fetus, where there is a rise in the level of cytokines TH2 and hormones (progesterone, estradiol),

and in case of decline these hormones and TH2 cytokines and in the contrast rise TH1 cytokines abortion will occur.<sup>[14]</sup>

Our results agree with the result of Nawzad (2007), where he confirmed that the level of the hormone estradiol rise in people while reduced hormone progesterone level, and also agreed with the results which showed that estradiol had the ability to enhance parasite reproduction.<sup>[15]</sup> As well as estradiol decrease natural killer cell activity and the production of IL-, IL-2, IL-6, INF $\gamma$  and TNF $\beta$ .<sup>[16,17,18]</sup>

So the immune response will be lower than normal and the infection with *T.gondii* will exacerbate. In the men's results are not affected by the infection with *T.gondii* or is not infected.

## 2- Comparison between patients and control in progesterone in women and men

Progesterone, play an important role in reproduction, including the maintenance of pregnancy in mammals, and immune function. Progesterone can have both activated and suppressive effects on the immune system, but is typically regarded as immunosuppressive.<sup>[19]</sup> Progesterone receptors have been identified in epithelial cells, mast cells, granulocytes (e.g. eosinophil), macrophages, and lymphocytes, also it can bind to glucocorticoid receptors, which are more presence in the immune system than progesterone receptors, and may represent an alternative mechanism for progesterone-induced changes in immune function.<sup>[20]</sup>

In Table (2) the results of this study showed that the level of the progesterone hormone in infected women ( $7.56 \pm 1.61$  ng/ml), while in control ( $11.72 \pm 2.36$ ) either in men results were infected ( $5.51 \pm 1.25$ ), while the control ( $4.01 \pm 0.22$ ). The results of the current study showed that there were no significant differences in progesterone concentration in women or men. These results were approached with the results of (Oktenli, *et al.*, 2004).<sup>[21]</sup> that showed that acute Toxoplasma infection may cause temporary sex hormone insufficiency regardless of the course of the disease. In the men results are not affected by the infection with *T. gondii* or are not infected to the fact that the hormone progesterone of female hormones Proof of this there has not been a large significant effect on the experience results.

**Table (2): Compare between patients and control in progesterone (ng/ml) in men and women**

Group	Sex		LSD value
	Men	Women	
Patients	5.51 ± 1.25	7.56 ± 1.61	3.472 NS
Control	4.01 ± 0.22	11.72 ± 2.36	5.302 *
LSD value	3.543 NS	5.699 NS	---
* (P≤0.05), NS: Non-significant.			

Our results are mainly in agreement with those by Al-Warid and Al-Qadhi (2012),<sup>[22]</sup> result of these studies showed that there were no significant differences in progesterone levels between infected and non-infected pregnant women with *T. gondii*. On the other hand, when estimated levels of progesterone in pregnant women seropositive IgG *Toxoplasma* antibodies and in controls with regard to gestational age.

Occurs in pregnancy increase in the production of progesterone during pregnancy, the beginning of the corpus luteum formation then after 8 weeks of pregnancy is the source of the placenta, which produces about 250 mg of progesterone per day per beneficiary of cholesterol for the mother initial substrate for the construction of progesterone then progesterone enters the product circulation but some of the mother's move to the fetal circulation and used as raw materials corticosteroids of the fetus, Progesterone is working to transform the endometrium to secretory stage in order to prepare the uterus for implantation. If pregnancy did not happen the hormone progesterone low level, leading to the occurrence of menstruation (menstrual).<sup>[23]</sup> But if the pregnancy occurred, progesterone will serve to reduce the immune response of the mother to be allowed to accept the pregnancy, and prevents contraction of the muscles of the uterus. The low level of progesterone is the first phase of the beginning of labor. In addition progesterone works inhibitor to the process of milk production (Lactation) during pregnancy and the proof is that low progesterone after childbirth is the first step for milk production.

In the current study note the low of the hormone progesterone and the high of estradiol level has due that to one or more reasons, including pathological changes which lead to disorder in the level of hormones because of the damage and necrosis arising from the development of toxoplasmosis disease mediated stage of injury infective stage the parasite *T. gondii* (Oocyst, bradyzoite, tachyzoite) and establish in the placenta, uterus and trophoblast and this member

that participates in the production and manufacture of hormones.<sup>[24]</sup> In addition to the parasite's ability to invade the placenta and fetal tissue and all cell types.<sup>[25]</sup>

A section of the researchers believe that the cause of death of the fetus, abortion is a parasite impact on the level of the host hormones that play an important role in maintaining pregnancy.<sup>[26]</sup>

The second reason is the overlap immune hormone, as the sex hormones such as estradiol and progesterone relationship with the immune system because of hormone receptor-steroidal site on most immune cells that can directly change gene expression for the cells of the immune system.<sup>[27]</sup> Such as macrophage, Lymphocyte, Eosinophil, mast cell, NK cell.

### The third reason

Physiological, it is noted that certain hormones, especially estradiol and progesterone increase during pregnancy.<sup>[24]</sup>

The concentrations of hormones in the first three months have little with low Th2 and here is a chance abortion High.<sup>[28]</sup>

### 3- Compare between patients and control in Prolactin in men and women

In table (3) result show significant decrease in the levels of prolactin in seropositive patients in women and men comparison to control subjects.

**Table (3): Compare between patients and control in Prolactin (ng/ml) in men and women**

Group	Sex		LSD value
	Men	Women	
Patients	5.81 ± 0.82	29.24 ± 4.41	6.721 *
Control	7.65 ± 1.78	45.15 ± 6.99	11.694 *
LSD value	3.397 NS	16.024 NS	---
* (P≤0.05), NS: Non-significant.			

Prolactin (PRL) is one of the most important hormones involved in immune regulation in the host body.<sup>[29]</sup> Exogenic prolactin can induce anti parasitic activity in microglial cells as a reaction against the *T. gondii* infection.<sup>[30]</sup>

In the current study, we observed significant decrease in levels of prolactin in both positive IgG anti-Toxoplasma antibody men and women.

When compared with control groups, higher levels of prolactin hormones in seronegative groups (control) in opposition to seropositive groups (chronic infection) may indicate to the protective action of PRL in a host organism against *Toxoplasma* infection. It has been reported that the prolactin hormone increases the production of immune globulins, cytokines and autoantibodies.<sup>[31]</sup>

Also (Mavoungou, 2006),<sup>[32]</sup> reported that prolactin concentrations increase during pregnancy, regardless of parity. He showed that prolactin concentration did not differ according to *P. falciparum* status.

(Dzitko *et al.* 2008).<sup>[33]</sup> Have found increased prevalence of latent toxoplasmosis in women with an aberrant level of prolactin.

(Dzitko *et al.* 2010).<sup>[29]</sup> Revealed that pre-incubation of the *Toxoplasma* tachyzoites with the recombinant human prolactin (rhPRL) *in vitro* resulted in a significant reduction (up to 36.15%) in the replication abilities of the parasite. They suggested that the inhibition of replication was caused by a limited capacity of the parasites to penetrate host's cells as demonstrated by the reduced number of infected cells in their study.

(Dzitko *et al.* 2012).<sup>[34]</sup> suggest that a significant increase in the serum PRL level, during pregnancy for instance, might significantly limit the risk of *Toxoplasma* spreading and could play an important role in natural protection against toxoplasmosis, and they revealed that exogenous human recombinant prolactin (rhPRL) as well as autologous endogenous prolactin present in serum – sPRL from inactivated sera significantly restricted intracellular growth of *Toxoplasma* in peripheral blood mononuclear cells cultures. Moreover, analysis of IL-10 production by PBMC infected with *Toxoplasma* and cultured in the presence of sPRL showed a positive correlation between sPRL concentration and the level of IL-10.

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