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The effect of toxoplasmosis infection in the concentration of interferon-ɣ and complementary proteins C3,C4 for parasite infected woman *Toxoplasma gondii*

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ABSTRACT

The current study deals with measuring the level of interferon-ɣ for 96 sample of infected women with *Toxoplasma gondii* , whose attended Salah-El-Deen Teaching Hospital in Salah- El-Deen Governorate from 13/10 /2016 to20/4/ 2017. The samples have been divided into 50 infected pregnant women with *Toxoplasma gondii* and 46 women as a control group which included 23 pregnant women and 23 non – pregnant whose as not infected. The study shows significant difference in the level of interferon ɣ between the infected women group and control group .The level of interferon ɣ in infected women was higher compared with the control group 1.658± 0.977 and 0.765 ± 0.524 pg. / ml, respectively, at the level of probability p< 0.01. concerning on to the measurement of the level of the complementary proteins C3, C4, It was shown the result exist variances moral if were valued in infection women of 0.3708± 0.0107 and 0.3851± 0.0306 mg/dl, respectively, compared to the valued in non-infection women 0.2528± 0.0463 and 0.2142±0.0608 mg/ dl, respectively, at the level of probability p< 0.01.

Introduction

Toxoplasmosis disease is a zoonosis disease. It is widespread all over the world which by the *Toxoplasma gondii* intracellular parasite. It can infect all the warm-blooded animals, most mammals, and birds as an lnter mediate [1]. The infection with toxoplasmosis differs according to region and age as it affects third of the world population. In the acute cases, the symptoms are implicit or trivial [2] Wild and domestic cats represent the final hosts through which oocysts of *Toxoplasma*, are through with feces. So it a vital role in distribution of the parasite [3] .The infection can be occurs when eating poorly cooked meat which contains cysts, contact with infected cat feces, contaminated food wate, dust during organ transplantation, blood transfusion, infection by transmitting through placenta or by inhaling contaminated air [4] . Severe symptoms may occur in those with immune deficiency. The disease is also spread by an infected mother to her child when pregnant leading to abortion or abnormal fetus [5] . Adult patients often show no symptoms, but 10 %of the disease accompanies the clinical toxoplasmosis connected with lymph node disease [6]. The incidence of toxoplasmosis caused an immune response to the production of antibodies, we note the high level of IgM within 1-2 weeks at the start of injury [7]. The IgA level is also observed with an increase against IgM in the acute phase of the infection and then the onset of IgG against all these antibodies will be directed against the parasite antigens[8] After 2-8 months, the IgM level begins and IgA gradually decreases [9]. IgM is often present for nearly a year [10]. Antibodies work with the complement to the analysis of the rapid phase of reproduction outside cells. When this phase is exposed to these objects it becomes unable to enter the cells and throws them through the phagocytosis [11] The infection of cells and tissues of the body with the parasite leads to the alteration of some values of the physiological norms such as Alanine amino transferees enzyme and aspartate amino transfers enzyme found in muscles and liver [12].the study aimed are the effect of *T. gondii* parasites on the level of interferon- ɣ in infected women , and the effect of parasitic *Toxoplasma gondii* on the formation of IgG, IgM, IgA antibodies, as well as on

the C3 and C4 protein complements in blanched women.

Material and Methods

Sample Collection:

The current study includes 96 patients of women attended to Saladin General Hospital in Tikrit City. The study starts from October to the end of March 2017. The studied samples had been divided as follows:

First group: 50 patients of women apportion Infected with Toxoplasmosis between 19-42 years ago.

Second group: This includes 23 pregnant women not infected with *Toxoplasma* between 15-40 years aged.

Third group: This includes 23 aborted women for other reasons,17-40 years aged.

Blood samples are collected from the selected women by taking 5ml vein blood by using tourniquet and 5ml medical syringes. Then the blood is divided into two types of test tubes. A readymade tube contains (EDTA) to test blood image by the CBC device. Empty disposable tube to be left 10 minutes, then it is to be put in the centrifugal accelerator at 3000 RPM for five minutes to separate serum from the blood. The serum is extracted by a Micropipette. Then it is placed in small pipes and kept in the refrigerator for future serum tests.

Measuring the concentration of interferon - ɣ by ELASA .

The test tools used in the research are the products of SHANGHAI YEHUA Biological Technology Co. Ltd. China.

Results are as shown in the Figure 1.

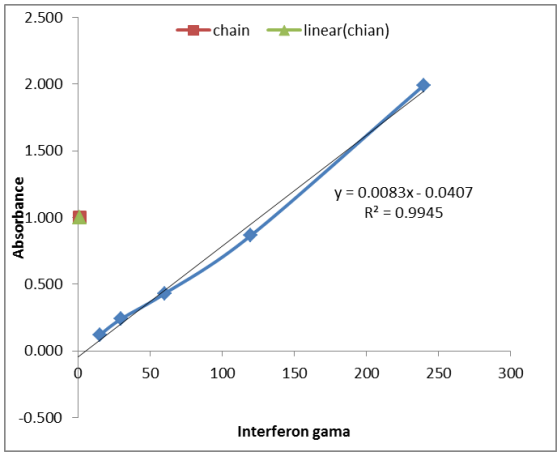

Figure (1) The standard curve of interferon -ɣ

A readymade set to test the concentration of the proteins of the third complementary C3.

according to the Spanish company Bio systems A. The absorption rate is measured at a wave length at 340 nm after 8 minutes of adding the sample. Results are as shown in the Figure 2 .

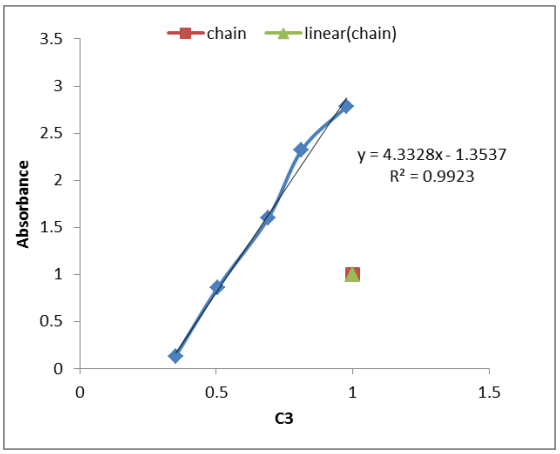

Figure (2) The standard curve of C3

A readymade set to test the concentration of the proteins of the forth complementary C4, and The kit used comes from the Spanish company Bio System S –A, and the results are as shown in the Figure 3.

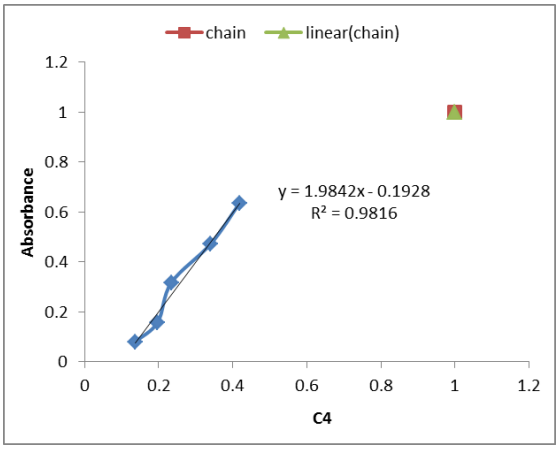

Figure (3) The standard curve of C4

Calculation

Standard curves are drawn showing the concentrations of standard solutions and values of absorption rates according the accompanied instructions, interferon concentrations, IgA and the complementary proteins C3 and C4. The calculations of their concentrations are based on the resulting equation from drawing the standard curve.

Statistical Analysis

The results are analyzed by the statistical program Mini tab (ver. 17) according to T- test for two samples.

Results and discussion

The level of interferon for proteins is 1.658 ±0.977 (table1) in comparison with the control group 0.765 ± 0.524 the current results show a significant rise in the level of interferon- ɣ in the serum of the infected persons with *Toxoplasma*. This clarifies the role of interferon-ɣ in the immune response against toxoplasmosis parasite. The interferon-ɣ resists and abolishes the parasite through stimulating a lot of mechanisms that contribute in the annihilation of the parasite or changing it from the fast reproducing phases to the slow reproducing phases under the

effect of many factors, including Nitric Oxide [13] and increasing the efficiency of 3-dioxygenase, Indoleamine 2 which dissolve the tryptophan amino acid vital for the life of the parasite [14] These results agree with the study of each of [15] and [16] in Iraq who assure the rise of the level of interferon-ɣ in comparison with the control group. In spite of the importance of interferon- ɣ in resisting parasites, producing it abundantly and uncontrolled response may cause illness to the host. So, the host needs a balanced immune response between removing the causes of the disease and decreasing the damages affecting the host [17]. Interferon-ɣ also plays an important role in stimulating the macrophage cells to swallow and kill the parasite. Interferon is produced from dendritic cells, Natural killers cells and leukocytes [18].

Table 1: Estimation of interferon -ɣ concentration (INF-Y) in healthy and infected patients.

Featuresgroup	Values
Non - pregnant patients	1.658±0.977
Control of pregnant and non – pregnant	0.765±0.524
t-Test value	**4.68
p- value	0.0002

** The high significantly of p-value

The results of statistical analysis C3 shows significant differences between the infected patients and a control group. Table 4 shows that the rate of the concentration of complementary protein C3 in the patients infected with Toxoplasmosis reaches 0.3708 ± 0.0107, whereas it reaches 0.2528 ± 0.0463 in the control group. As for protein C4, the calculation averages for this protein indicate significant differences for infected women in comparison with the control group. The protein's rate in the infected women 0.3851 ± 0.0306 and in the control group 0.2142± 0.0603 . These differences may be attributed to the collaboration of the complementary proteins in the activity of the complementary against infection with the parasites which are produced in the liver stimulated by tumor necrosis factor type TNF-a and interleukin in the stage of primary infection so these proteins are counted among acute phase proteins [19] The results of this study does not agree with [20] who makes it clear that there are no significant difference for the C3 and there are significant differences for C4 among the infected women with toxoplasmosis. Higher integrals may also be due to the complement system being the first mediator of various inflammatory processes and also a mediator of interactions between antibodies and antigens and the best stimulant of the IgM complement system at the end of the first week after infection[21].

Table 2: Estimation of the concentrations of the proteins C4, C3 (mg/dl) for healthy persons and infected patients with *T.* gondii.

Features group	C3	C4
Non - pregnant patients	0.3708±0.0107	0.3851 ±0.0306
Control of pregnant and non-pregnant	0.2528±0.0463	0.2142±0.0608
t-Test value	**15.09	**3.80
p-value	0.0005	0.004

** The high significantly of p-value

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تأثير الخمج بداء المقوسات في تركيز الانترفيرون كاما والبروتينات المتممة C3,C4 للنساء المخمجات بطفيلي *Toxoplasma gondii*

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الملخص

تناولت الدراسة الحالية قياس مستوى الانترفيرون كاما ل 96 عينة من النساء المخمجات بطفيلي المقوسات الكوندية المراجعات لمستشفى صلاح الدين التعليمي في محافظة صلاح الدين للفترة 2016/10/13 لغاية 2017/4/20 وقد قسمت العينات الى 50عينة من النساء الحوامل المخمجات بطفيلي *Toxoplasma gondii* و46 عينة كمجموعة سيطرة. اشتملت على 23عينة من النساء الحوامل و23 عينة من النساء غير الحوامل من غير المخمجات ب *T.gondii*. واظهرت الدراسة وجود فرق معنوي في مستوى الانترفيرون كاما بين النساء المخمجات ومجموعة السيطرة اذ ارتفع مستوى الانترفيرون كاما في النساء المخمجات مقارنة بمجموعة السيطرة 1.658 ± 0.977 و0.765 ± 0.524 بيكوغرام / لتر على التوالي عند مستوى احتمالية $p<0.01$. وفيما يتعلق بقياس مستوى بروتينات المتمم C3,C4 فقد اظهرت النتائج وجود فروق معنوية اذ كانت قيمها في النساء المخمجات 0.3708 ±0.0107 و0.03851 ± 0.0306 ملغرام / ديسلتر على التوالي مقارنة بقيمها بالنساء غير المخمجات 0.2528 ± 0.0463 و0.2142 ± 0.0608 ملغرام / ديسلتر على التوالي عند مستوى احتمالية $p<0.01$.