Study of Some Immunological aspects of Diabetic type1 Infected with Toxoplasmosis

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Abstract

The current study was performed on diabetic type1 patients infected with \textit{Toxoplasma gondii} parasite in order to investigate the role of interleukin IL-2 and IL-17 in immune response against the parasite. A total of 160 serum samples were collected from Thi-qar hospitals and private laboratories divided into four groups: diabetic type1 infected with toxoplasmosis 40 sample, patients with diabetic type1 only 40 sample, patients with toxoplasmosis only 40 sample and control group 40 sample. The results of this study founded higher concentration serum level of IL-2 in diabetic type1 infected with toxoplasmosis, then diabetic type1 only group, then toxoplasmosis infected group while the lowest concentration was in control group. The results of IL-17 serum level recorded higher concentration in toxoplasmosis infected group, then diabetic type1 infected with toxoplasmosis while the lowest concentration was in control group.

Key words: IL-2, IL-17, Diabetes type1, Toxoplasmosis

How to cite this article: Hadi NA, Kodair AM (2020): Study of some immunological aspects of diabetic type 1 infected with toxoplasmosis, \textit{Ann Trop Med & Public Health}; 23(S16): SP231613.

DOI: \url{http://doi.org/10.36295/ASRO.2020.231613}

Introduction

Toxoplasmosis is one of most common disease caused by \textit{Toxoplasma gondii} an apicomplexan protozoan intercellular parasite that infected more than third of world’s population (Al-Shamma, 2014). \textit{Toxoplasma} infect all warm blooded mammalian especially Felida family (Tenter et al., 2000).

The disease occur by ingested Oocyst with food or water, by tissue cysts in raw meat and tachyzoites may infect embryo from pregnant mothers that infected with toxoplasmosis (Mahami \textit{et al.}, 2017; Master, 2015).

In most individuals acute infection with toxoplasmosis is asymptomatic. acquired infections are usually associated with reticular cell hyperplasia and lymphadenopathy (Torgerson and Mastroiacovo, 2013). Toxoplasmosis can also cause severe disease in immunocompromised patients such as HIV positive or patients treated with immunosuppressive drugs (Alavi and Alavi, 2016; Mohraz \textit{et al.}, 2011).

\textit{Toxoplasma gondii} infection leads to induce the production of a many proinflammatory cytokines (TNF-\textalpha; IL-1; IL-15; IL-17), anti-inflammatory cytokines (TGF-\textbeta; IL-4; IL-10),reactive oxygen synthesis, nitric oxide synthase, that related with inflammatory responses in various sites and cells, including the brain, astrocytes, microglial cells and infiltrating CD4+ and CD8+ T cells (Henriques \textit{et al.}, 2009; VanWormer \textit{et al.}, 2014).

\textit{Annals of Tropical Medicine & Public Health} \url{http://doi.org/10.36295/ASRO.2020.231613}
Insulin-dependent diabetes mellitus (IDDM) or type I diabetes is a debilitating chronic disease that impairs production and secretion of the key hormone insulin and alters blood sugar metabolism. Insulin is synthesized and secreted by pancreatic islet cells of Langerhans (WHO, 2014).

Type 1 diabetes mellitus is caused by loss the insulin-producing beta cells of the pancreatic islets, that is leading to insulin deficiency. The major individual of type I diabetes is caused by immune-mediated nature, in which a T cell-mediated autoimmune attack, thus, leads to the loss of beta cells and insulin. Type 1 diabetes can affect children or adults so termed “juvenile diabetes” because a most cases of these diabetes were in children. (Rother, 2007).

Interleukin-2 (IL-2) is an interleukin, a type of cytokine signaling molecule in the immune system. It is a 15.5–16 kDa protein that regulates the activities of white blood cells (leukocytes, often lymphocytes) that are responsible for immunity. IL-2 is part of the body's natural response to microbial infection, and in discriminating between foreign (“non-self”) and "self". IL-2 mediates its effects by binding to IL-2 receptors, which are expressed by lymphocytes. The major sources of IL-2 are activated CD4+ T cells and activated CD8+ T cells. (Liao et al., 2011).

IL-2 has acritical roles in immune system, tolerance and immunity, via its direct effects on T cells In thymus, where T cells mature, it can prevents autoimmune diseases by promoting the differentiation of certain immature T cells into regulatory T cells,. IL-2 enhances activation-induced cell death (AICD). (Arenas-Ramirez et al., 2015) IL-2 also induce the differentiation of T cells into effector T cells and memory T cells when the initial T cell is also stimulated by an antigen, thus helping the body fight off infections. (Liao et al., 2011) Together with other cytokines, IL-2 stimulates naive CD4+ T cell differentiation into Th1 and Th2 lymphocytes while it impedes the differentiation into Th17 and Th lymphocytes. (Liao et al., 2013).

Interleukin (IL-17) is one of pro-inflammatory cytokine which produced by a group of T helper cell known as T helper 17 cell in immune response to stimulation with IL-23. Th17 was identified in 1993 by Rouvier et al. who isolated IL-17 transcript by a rodent T-cell hybridoma. (Starens et al., 2002) After binding to the receptor, IL-17 activates several signaling cascades that is leading to the induction of chemokines. Acting as chemoattractant, these chemokines recruit the immune cells, like neutrophils, lymphocytes, monocytes and innate immune cells to the site of inflammation. All the signaling events follow an invasion of the body by pathogens and inducing the inflammation and antimicrobial functions. (Miossec et al., 2009; Chiricozzi et al., 2011; Hajishengallis, 2014).

Although IL-17 can induce the production of epithelial cell-derived antimicrobial molecules many studies current evidence from human and animal model studies that suggests the net effect of IL-17 signaling promotes disease development. (Iwakura et al., 2011; Eskin et al., 2012; Miossec and Kolls, 2012).

Aim of the study:

The aim of this study was to detection the role of IL-2 and IL-17 among diabetic type1 patients compared with other groups and control to understand the possible relationship between Toxoplasmosis and diabetes type1 by using ELISA technique.

Materials and Methods

1-Study Groups:
The Study groups divided into four (a group of Type1 diabetic patients infected with Toxoplasmosis 40 sample, a group of Type1 diabetic only 40 sample, a group of Toxoplasmosis patients only 40 sample and control group 40 sample).

2-Collection of samples:

The study samples were collected from Thi-Qar hospitals and private laboratories for the periods started from 1st of September 2018 to the end of July 2019. A total of 60 samples of type1 diabetic were diagnosed infected with *Toxoplasma gondii* parasite from 350 samples, while 260 samples were infected with type1 diabetes only, 30 samples collected from patients infected with Toxoplasmosis only and 30 samples as a control. We collected blood samples from each patients then separated serum by using gel tubes and centrifuge for (5-10)minutes on (2500-3000)c/min ,Then freeze samples under -20c° , we used 30 sample from each group to detection the concentration of IL-2 and IL-17 by ELISA technique.

3-Using Enzyme-linked Immuno Sorbent Assay (ELISA) to calculate concentration of Cytokines (IL-2 and IL-17):

Assay procedure:

The main steps of this technique include adding 100 μL standard or sample to each well. Incubate for (90) min at 37°C, remove the liquid, Adding 100 μL HRP Conjugate. Incubate for (30) min at 37°C. Aspirating and washing 5 times, Adding 90 μL of Substrate Reagent. Incubate for (15) min at 37°C, Adding 50 μL Stop Solution and Reading at 450 nm immediately, finally calculation of results.

Results

1-Interleukine- 2 result:

The current study recorded highest concentration of IL-2 in type1 diabetic patients infected with Toxoplasmosis group which recorded (343.02 ± 59.30) pg/ml, then type1 diabetes mellitus only group which recorded (289.59 ± 52.92) pg/ml, then Toxoplasmosis infected group which recorded (197.86 ± 53.98) pg/ml, compared with control group which recorded the lowest concentration of IL-6 which recorded (87.12 ± 17.55) pg/ml.

Table (1): Interleukin -2 concentration in study groups.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Interleukin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diabetic 1+toxoplasmosis</td>
<td>343.02 ± 59.30</td>
</tr>
<tr>
<td>Diabetic type1</td>
<td>289.59 ± 52.92</td>
</tr>
<tr>
<td>Toxoplasmosis</td>
<td>197.86 ± 53.98</td>
</tr>
<tr>
<td>Control</td>
<td>87.12 ± 17.55</td>
</tr>
<tr>
<td>L.S.D</td>
<td>35.67</td>
</tr>
</tbody>
</table>
Figure (1): The Standard curve of Interleukin -2 concentration in study groups.

2-Interleukine -17 result:

The study recorded highest concentration of IL-17 in Toxoplasmosis infected group which recorded $(426.09 \pm 21.59)$ pg/ml, then type1 diabetes mellitus infected with Toxoplasmosis group which recorded $(405.21 \pm 43.66a)$ pg/ml, then type1 diabetes mellitus only group $(367.48 \pm 34.78b)$ pg/ml, compared with control group which recorded the lowest concentration of IL-12 $(299.09 \pm 18.35c)$ pg/ml.

Table(2): Interleukin -17 concentration in study groups.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Interleukins</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>IL-17</td>
</tr>
<tr>
<td>Diabetic 1+Toxoplasmosis</td>
<td>$405.21 \pm 43.66a$</td>
</tr>
<tr>
<td>Diabetic type1</td>
<td>$367.48 \pm 34.78b$</td>
</tr>
<tr>
<td>Toxoplasmosis</td>
<td>$426.09 \pm 21.59a$</td>
</tr>
<tr>
<td>Control</td>
<td>$299.09 \pm 18.35c$</td>
</tr>
<tr>
<td>L.S.D</td>
<td>23.04</td>
</tr>
<tr>
<td>P-Value</td>
<td>0.0001</td>
</tr>
</tbody>
</table>
Figure(2): The Standard curve of Interleukin -17 concentration in the study groups.

Discussion

Toxoplasma infection is common among opportunistic patients (Dubey, 2005) Diabetic patients were included in a classification of the immuno-compromised patients (Olefsky, 1985), thus Toxoplasmosis in type1 diabetic individual may cause sever or chronic infection. Toxoplasmosis in most immunocompetent individuals occur with nonspecific sign and asymptomatic and Most of them recover without any treatment (Kankova, 2015) by an efficient immune system but in immunocompromised individual Toxoplasma can replicate in any nucleated cell leading to induce various inflammatory marker via innate acute inflammatory response and by an antigen –specific adaptive immunity ,thus lead to chronic infection. (Prandota, 2013)

This study founded significant increasing of interleukin -2 in the group of type 1 diabetes infected with Toxoplasmosis ,then the group of type1 diabetic only ,then the group of Toxoplasmosis only ,compared with control which recorded lowest concentration.

IL-2 has acritical role in immune response against Toxoplasma gondii because it enhance the activation of induced cell death (Arenas-Ramirez, 2015 ), IL-2 also induce the differentiation of T cells into active (effector) T-cells , memory T-cells when its stimulate by antigen ,so IL-2 helping the body to fight any infection by invading organisms (Liao, 2011) , together with cytokine , IL-2 can stimulate naïve CD4+ T-cells into Th1 and Th2 , also involve in activation of Th17 lymphocytic response. (Liao, 2013).

The results of the current study agreement with Al-Obaidy, 2019 and Saeed, 2018 those found increasing in serum level of IL-6 among diabetic patients infected with toxoplasmosis. Similarly, Targher et al., 2000; Choudhary and Ahlawat, 2008; Shelbaya et al., 2012; He et al., 2014) found higher serum IL-6 levels in patients of diabetes type I compared to control group. In addition Reis et al., (2012) analyzed plasma samples of 42 diabetic type(1) patients and 24 healthy samples as a control, finding higher circulating levels of IL-6 in diabetes type1 patients than in the control group.

The present study founded significant increasing of interleukin -17 in the group of type 1 diabetes infected with Toxoplasmosis ,then the group of type1 diabetic only ,then the group of Toxoplasmosis only ,compared with control which recorded lowest concentration.
The IL-17 has been linked with many immune and autoimmune diseases, also has a critical role in immune regulatory function. IL-17 is involving to inducing and mediating proinflammatory responses and induce the production of other cytokines like (TNF-α, TGF-β, IL-1β, IL-6, and GSF) which have an important role in immune response against Toxoplasmosis. IL-17, also can induce the production of many cells like macrophages and endothelial cells. IL-17 is essential to subset of CD4+ T cells that called Thelper 17. (Aggarwal and Gurney, 2002)

Increasing serum level of IL-17 in patients compared with healthy group are agree with those previous of studies saeed, 2018 which recorded high level of IL-17 among diabetic infected with Toxoplasmosis, also agree with Bradshaw et al., (2010); Honkanen, (2010) and Tuama et al., (2014) all they found that the level of IL-17 in D1M patients groups were a significant elevated in comparisons with healthy group.

References


Honkanen, J. (2010). "Studies of Immune Regulation in Type 1 diabetes" Academic dissertation for PhD, Faculty of Medicine/ University of Helsinki.


Tuama , A.; Hassan, H. and Rashid ,K. (2014). Estimation the level of il-17a in a sample of type i diabetes mellitus patients . IOSR Journal Of Pharmacy .57-60
