Levels of Immunoglobulins and complements in sera of patients with toxoplasmosis

Wafaa Sadoon Shani*, Bushra Hussein Shnawah*, Nabel Elia Wahedah**

Dept. of Biology, College of Science, University of Basrah, Basrah – Iraq*.
College of Scientific Medical Techniques, Baghdad**

Abstract

Eighty Seven sera of infected women with toxoplasmosis was tested to determined the levels of IgG, IgM, IgA, C3 and C4 by using single radial immunodiffusion technique and compared with the control sera of non infected subjects. The results showed that there was arising in the titer of IgG, IgM, C3 and C4 whereas IgA titer not recorded any arising when it was compared with the sera of control group.

Introduction

Toxoplasmosis, a coccidian infection, is caused by obligate intracellular parasite Toxoplasma gondii. Although generally benign for healthy people, infection can result in stillbirth, blindness, mental retardation and occasionally the death of congenitally infected infants (Frenkle, 1988). The parasite is globally distributed and can be found within many different species of mammals and birds. It is estimated that up to 5 * 10^8 people worldwide are infected with T. gondii (Denker’s and Gazzinelli, 1998).

There is an important immunological situation where toxoplasmosis is of clinical concerns. That is in the case of a pregnant mother with newly contracted acute toxoplasmosis. In this situation, the female does not have acquired immunity to the parasite and tachyzoites can cross the placenta to infect the fetus (Kaufmann, 1997). However, chronically infected mothers rarely pass the infection to their unborn child, because maternal immunity maintains tachyzoites replication at subclinical levels.
The outcome of a primary infection is dependent on the ability of the host to mount an immune response capable of controlling the multiplication and spread of the parasites. The response to *T. gondii* is complex and involves humoral as well as cellular mechanisms (Gustafsson, 1997). So in this study we try to determined the titer of antibody in sera of infected women and correlate these titers with titer of complement component specially C3 and C4 by using single radial immunodiffusion technique in order to explain the role of antibody during the infection with *T. gondii* and the role of complement. Also we want to know which class of antibody rise in sera of infected women in this instance.

**Materials and Methods**

**A- Collection of samples**

Eighty seven sera from infected women were collected from the department of Primary Health Care in Basrah city. These samples considered as a positive sera by using indirect fluorescent antibody test (IFAT) whereas other twenty five sera where used as a control according to the result of (IFAT).

**B- Single radial immunodiffusion assay (SRID)**

The (SRID) test is performed by using biomaghrebi kit.

The plates were opened and left to stand at room temperature for a few minutes to allow any condensed water in the wells to evaporate. The wells were filled with 5µl of testing sera (infected and control). Then we left a plate to stay at room temperature for about (48) hour in the case of IgG, IgA, C3 and C4 and for (72) hour in the case of IgM.

**C- Statistical analysis**

The data was statistically analyzed by ANOVA test (Walpole, 1982).

**Results**

Serum IgG, IgM and IgA levels in women infected with *T. gondii* are shown in table (1). The results indicated that there was a difference in the titer of IgG between the infected women and control group. The infected group has a mean of about (1361.2) but the IgG mean of control group was (992.3), statistically there was a significant differences between two groups (P<0.0001).

The same is true in the case of IgM where the results showed a significant differences between the infected and control group (P<0.0001), the mean level of IgM was about (174.7) in infected women whereas its value reached (111.44) in control group.

No statistical difference was found in serum IgA levels in women with toxoplasmosis (233.9) when compared with normal control (190.68) (P<0.052).
Table (1) Immunoglobulins levels in sera of infected women with toxoplasmosis and control group

<table>
<thead>
<tr>
<th>Ig class</th>
<th>Ig concentration mg/dl of serum</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Patients</td>
</tr>
<tr>
<td></td>
<td>M ± SD</td>
</tr>
<tr>
<td>IgG</td>
<td>1361.2 ± 222.8</td>
</tr>
<tr>
<td>IgM</td>
<td>174.7 ± 52.74</td>
</tr>
<tr>
<td>IgA</td>
<td>233.9 ± 86.75</td>
</tr>
</tbody>
</table>

Where : M = Mean : SD = Standard Deviation

The mean levels of C3 and C4 in serum are shown in table (2). Serum C3 mean levels in infected women was differ (147.93) from that of control group (119.89).

Furthermore, significant differences was occurred in the serum C4 of infected women in comparison with normal subjects (P<0.0001) and the mean levels of patients (32.291) differ from that of control group (26.088).

Table (2) Complements (C3 and C4) levels in sera of infected women with toxoplasmosis and control group

<table>
<thead>
<tr>
<th>Complement</th>
<th>Complement concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Patients</td>
</tr>
<tr>
<td></td>
<td>M ± SD</td>
</tr>
<tr>
<td>C3</td>
<td>147.93 ± 27.28</td>
</tr>
<tr>
<td>C4</td>
<td>32.291 ± 8.432</td>
</tr>
</tbody>
</table>

Where : M = Mean : SD = Standard Deviation

Discussion

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 Mandlle et al. (2000) 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 Chardes et al. (1990) and Roberts and McLeod (1999) in indicating the association between the infection and strong humoral response involving IgM, IgG, IgA and IgE. Sayles et al. (2000) also 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 (Internet, 1997). 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 (Internet, 1997).

In the case of IgA there was no significant differences in the titer of this antibody between the infected sera and control sera and this may be due to the importance of the secretory IgA in the case of infection rather than the serum IgA because McLeod and Mack (1986) demonstrated the production of intestinal IgA antibody to T. gondii and later suggested that IgA may play a protective role against toxoplasmosis. The role of secretory IgA in reducing the initial infection of enterocytes was also reported by Mack and McLeod (1992). Furthermore it was necessary to note that some studies indicate the importance of IgA as an element of mucosal immunity to oral infection to Toxoplasma cysts (Chardes et al., 1990). Antibodies of this isotype may be important in avoiding reinfection with T. gondii, and their induction of IgA is a major strategy for vaccine development.

Although there was an increase in the titer of infected women and there was an arising in the IgG and IgM antibody, but there is a problem associated with serologic diagnosis is that antibody to T. gondii is present in relatively high numbers of individuals in most human populations. These antibody titers may persist at a high level for years in healthy people.

Number of serologic tests often measure different antibodies that posses unique patterns of rise and fall with time after infection, false positive and false negative results (or both) have been a problem with certain commercial kit and laboratories in the United States and Europe (Liesenfeld and Montaya, 1997; Wilson and et al., 1997).

Moreover, techniques demonstrating circulating T. gondii antigen and immune complexes have been developed but are not widely applied in routine diagnostics. One the limitation of serology in toxoplasmosis is that
antibody levels have no correlation with the severity of disease (Dubey and Beattie, 1988)

Significant differences were reported in the levels of C3 and C4 between the serum of infected women in comparison with non-infected one. These observation is in agreement with results of earlier study done by Sabin and Feldman in (1948) when they were described a mechanism of humoral immunity to this parasite, that required antibody and heat – labile accessory factor or activator present in fresh serum. Suzuki et al. (1971) reported a requirement for C4 and C2, which indicate that the classical complement pathway was involved in the killing of Toxoplasma.

This increasing in the titer of C3 and C4 may be contributed to the ability of the parasite to repair their membrane after the membrane attack complex of complement has been assembled on its surface (Schreiber and Feldman, 1980) and these factor may necessitate a high input of complement activity in order to cause cytolysis of the organism.

The cooperation between specific antibody and complement was also reported by Hammouda et al. (1995) in the case of killing or lysis of extracellular tachyzoites. Roberts and McLeod (1999) concluded that the extracellular tachyzoites coated with antibody and complement can be lysed via the classical complement pathway or killed within phagocytes.

References


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