Seroprevalence of toxoplasma in BOH cases in a tertiary care hospital
Ponugoti Munilakshmi, T.Kasturi and Munagala venkata Krishna
Dr. NTR University of Health sciences, Vijayawada, Andhra Pradesh.
E-mail:lakshmiponugoti@yahoo.in

ABSTRACT:
OBJECTIVE: This study is conducted to assess the seroprevalence of toxoplasma in antenatal women and correlate with BOH. BACKGROUND: Toxoplasma one of the important parasite causing recurrent pregnancy losses. Prevalence of toxoplasmosis in Indian pregnant women is 7.7%. By ELISA we can identify the antibodies against toxoplasma and we can prevent the infection as well as we can reduce the severity of disease. MATERIALS AND METHODS: A total of 174 samples were collected from antenatal women who attended Narayana Medical College & Hospital, Nellore, during the period from November 2009 to August 2010. RESULTS: Among 174 cases 134 were BOH cases, 40 were controls. In 134 cases 11.9% positive for IgM, 20.9% for IgG. In controls 20% positive for IgM and all controls are negative for IgG. CONCLUSION: This study concluded that there is correlation between toxoplasma infection and Bad Obstetric history.

1. INTRODUCTION
Pregnancy is one of the most desirable events of a woman. But pregnant are more prone to lot of infections due to temporary immunosuppression leading to recurrent pregnancy loss. Out of many aetiological agents responsible for this loss, Toxoplasma gondii is one among them.

BOH implies previous unfavorable foetal outcome in terms of two/more consecutive spontaneous abortions, history of intra uterine foetal death, intrauterine growth retardation, still births, early neonatal death and congenital anomalies[1]

Serological surveys indicate that human infections are common in many parts of the world. In some countries more than third of the human population have antibodies against T.gondii. This high prevalence of infection in man proves the importance of toxoplasmosis as a zoonotic disease, particularly in pregnant women and immunocompromised patients. Usually it doesn’t cause serious illness, but it can cause mental retardation in congenitally infected children and immunocompromised individuals [2].

Infection is mainly acquired by ingestion of food or water that is contaminated with oocysts shed by cats or by consuming contaminated meat containing tissue cysts. In addition, infection may be acquired by contact with cat feces containing oocysts. Toxoplasmosis can be transmitted to the fetus inutero through transplacental transmission [3].

Other modes of transmission are by transfusion of blood or blood products, organ transplants, ingestion of milk or saliva and by eating eggs [4].

Seroprevalence of Toxoplasma antibody in Europe and USA varies between 50-60% and ranges between 4-9% in India. In pregnant women on the worldwide scale, there are seroprevalence from 7%-51.3% and in women with abnormal pregnancies and abortions, the seroprevalence vary from 17.5-52.3%. Prevalence of toxoplasmosis in Indian pregnant women is 7.7% [5].
The disease is associated with HIV infection and considered as indicator disease for AIDS. It also associated with teratogenic infections and congenital infections mainly in the form of chorioretinitis. Most of the infections are associated with bad obstetric history in symptomatic cases and associated with still births [6].

The detection of antibodies IgM, IgG to T.gondii is helpful for the diagnosis of acute and chronic infections [7]. As parasite can lead to severe birth defects detection of antibodies during pregnancy is useful to know the chronicity and risk of symptomatic disease.

So previous history of pregnancy, detection of antibodies during pregnancy should be done in routine obstetric practices to reduce adverse foetal outcome.

2. MATERIALS AND METHODS

A total of 174 blood samples were collected from antenatal women who attended Narayana Medical College & Hospital, Nellore during the period from November 2009 to August 2010.

A total of 134 samples were collected from the antenatal women with Bad Obstetric History as test group and 40 samples were collected from antenatal women without any pregnancy wastage.

Among the test group samples 76 were from antenatal women with history of repeated abortions, 15 from Intra uterine death, 9 from Intra uterine growth retardation, 15 from congenital anomalies and 19 from pre-term deliveries.

Cases from other causes of abortions like cervical incompetence, Rh incompatibility, Diabetes, Syphilis, HIV, Hepatitis were not included in this study.

2.1. Collection & Transport of specimen:

2-3 ml of venous blood was collected under aseptic precautions in vaccutainers with no additives and kept at room temperature for 30 minutes to clot. Serum was separated by centrifugation and transferred to plastic aliquots. The aliquots were labeled with information of patient and were stored at -20°C until tested.

Serum samples from both groups of subjects were tested using the commercial kits qualitative immune enzymatic determination of IgM and IgG antibodies for T.gondii. The commercial kits used were EUROIMMUN. They are stored at 2-8°C as per manufactures instructions.

Other tests used for the diagnosis of toxoplasma are Sabin-Feldman’s dye test, IgG avidity, indirect immunofluorescence test, Polymerase chain reaction.

3. RESULTS

A total number of 174 blood samples were collected from antenatal women who attended Narayana Medical College & Hospital, Nellore during the period from June 2009 to April 2010. The blood samples were processed in the Department of Microbiology, Narayana Medical College, Nellore.

Samples were screened for IgM and IgG Antibodies against Toxoplasma gondii using EURO IMMUN kit by ELISA, as per the manufactures instructions. Among 174 cases 134 were BOH cases, 40 were controls. In 134 cases 11.9% positive for IgM, 20.9% for IgG.
### TABLE - 1
SEROPREVALENCE OF TOXOPLASMA ANTIBODIES AMONG 2 GROUPS

<table>
<thead>
<tr>
<th>Group</th>
<th>No of sera tested</th>
<th>Seropositivity</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (BOH)</td>
<td>134</td>
<td>16 (11.9%)</td>
<td>28 (20.9%)</td>
<td></td>
</tr>
<tr>
<td>2 (CONTROL)</td>
<td>40</td>
<td>2</td>
<td>NIL</td>
<td></td>
</tr>
</tbody>
</table>

### TABLE - 2
SERO PREVALENCE OF TOXOPLASMA ANTIBODIES IN RELATION TO CLINICAL PRESENTATION

<table>
<thead>
<tr>
<th>Clinical presentation</th>
<th>No of sera tested</th>
<th>Seropositivity</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Abortions</td>
<td>76</td>
<td>11(14.4%)</td>
<td>16 (21.0 %)</td>
<td></td>
</tr>
<tr>
<td>Congenital anomalies</td>
<td>15</td>
<td>02 (13.3%)</td>
<td>04 (26.6%)</td>
<td></td>
</tr>
<tr>
<td>Preterm delivery</td>
<td>19</td>
<td>Nil</td>
<td>Nil</td>
<td></td>
</tr>
<tr>
<td>Still birth (IUD)</td>
<td>15</td>
<td>02 (13.3%)</td>
<td>08 (53%)</td>
<td></td>
</tr>
<tr>
<td>IUGR</td>
<td>09</td>
<td>01(11.1%)</td>
<td>Nil</td>
<td></td>
</tr>
</tbody>
</table>

### TABLE - 3
AGE RELATED SEROPOSITIVITY OF TOXOPLASMA IN BOH CASES

<table>
<thead>
<tr>
<th>Age in years</th>
<th>No of sera tested</th>
<th>Seropositivity</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>18-23</td>
<td>34</td>
<td>03 (8.82%)</td>
<td>10 (29.4%)</td>
<td></td>
</tr>
<tr>
<td>24-29</td>
<td>87</td>
<td>09 (10.3%)</td>
<td>17 (19.6%)</td>
<td></td>
</tr>
<tr>
<td>30-36</td>
<td>13</td>
<td>04(30.8%)</td>
<td>01 (7.7%)</td>
<td></td>
</tr>
</tbody>
</table>

### TABLE - 4
SERO POSITIVITY OF TOXOPLASMA ANTIBODIES IN RELATION TO PARITY

<table>
<thead>
<tr>
<th>Gravida</th>
<th>No of sera tested</th>
<th>Seropositivity</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Primi</td>
<td>36</td>
<td>03 (8.3%)</td>
<td>13 (36.1%)</td>
<td></td>
</tr>
<tr>
<td>Multi</td>
<td>98</td>
<td>13 (13.3%)</td>
<td>15 (15.3%)</td>
<td></td>
</tr>
</tbody>
</table>

### TABLE - 5
SEROPREVALENCE OF TOXOPLASMA ANTIBODIES IN RELATION TO GESTATIONAL PERIOD

<table>
<thead>
<tr>
<th>Gestational age</th>
<th>No of sera tested</th>
<th>Seropositivity</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;20 weeks</td>
<td>43</td>
<td>1 (2.32%)</td>
<td>7 (16.3%)</td>
<td></td>
</tr>
<tr>
<td>&gt;20 weeks</td>
<td>91</td>
<td>15 (16.2%)</td>
<td>21 (23%)</td>
<td></td>
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</tbody>
</table>

### TABLE - 6
SERO PREVALENCE OF TOXOPLASMA ANTIBODIES IN RELATION TO THE URBAN AND RURAL POPULATION

<table>
<thead>
<tr>
<th>Residential status</th>
<th>No of sera tested</th>
<th>Seropositivity</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Urban</td>
<td>79</td>
<td>9 (11.4%)</td>
<td>15 (18.9%)</td>
<td></td>
</tr>
<tr>
<td>Rural</td>
<td>55</td>
<td>7 (12.7%)</td>
<td>13 (23.6%)</td>
<td></td>
</tr>
</tbody>
</table>
TABLE - 7
SEROPREVALENCE OF TOXOPLASMA ANTIBODIES IN RELATION TO CONTACT WITH PET ANIMALS

<table>
<thead>
<tr>
<th>H/O contact with pet Animals</th>
<th>No of sera tested</th>
<th>IgM (%)</th>
<th>IgG (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes</td>
<td>45</td>
<td>5 (11.1%)</td>
<td>10 (22.2%)</td>
</tr>
<tr>
<td>No</td>
<td>89</td>
<td>11 (12.3%)</td>
<td>18 (20.2%)</td>
</tr>
</tbody>
</table>

Fig 1: CASE CONTROL DISTRIBUTION

Fig 2: SEROPREVALENCE OF TOXOPLASMA ANTIBODIES AMONG 2 GROUPS

Fig 3: SERO PREVALENCE OF TOXOPLASMA ANTIBODIES IN RELATION TO CLINICAL PRESENTATION
Fig: 4
AGE RELATED SEROPOSITIVITY OF TOXOPLASMA INBOH CASES

Fig: 5
SERO POSITIVITY OF TOXOPLASMA ANTIBODIES IN RELATION TO PARITY

Fig: 6
SEROPREVALENCE OF TOXOPLASMA ANTIBODIES IN RELATION TO GESTATIONAL PERIOD
In controls 20% positive for IgM and all controls are negative for IgG as in table1.BOH cases are further studied depend upon clinical presentation, age, parity, gestational age, relation to the urban and rural population and contact with cats.

**Clinical presentation:** High prevalence of Toxoplasma IgM were seen in abortion cases(14.4%) and IgG were seen in IUD cases(53%) followed by congenital anomalies(IgM 13.3% and IgG 26.6%) as in table2.

**Age:** highest seropositivity for IgM antibodies in the age group of 30-36 years whereas for IgG in the age group 18-23 years as in table3.

**Parity:** seropositivity of IgM antibodies in primiparous women is 8.3% and a multiparous woman is 13.3% whereas IgG antibody in primi and multiparous women shows 36.1% and 15.3% respectively as in table4.

**Gestational age:** 2.32% seropositivity of IgM antibodies in women with <20 wks gestation, whereas16.2% in >20 wks gestation. IgG seropositivity was 16.3 % and 23% in < 20wks and >20 wks respectively as in table5.

**Relation to the urban and rural population:** seropositivity of IgM antibodies in urban and rural is 11.4% and 12.79% whereas IgG antibodies18.9% and 23.6% respectively as in table6.

**Contact with pet animals:** IgM seropositivity in persons with a history of contact with pet animals and without contact is 11.1%, 12.3% and IgG is 22.2%, 20.2% as in table7.

**4. DISCUSSION:**

Maternal infections play a critical role in pregnancy wastage and their occurrence in patients with BOH is a significant factor. Toxoplasma infection during pregnancy may lead to transmission to the fetus and results in abortion, intrauterine death, preterm deliveries, Intra uterine growth retardation and congenital anomalies. Diagnosis of toxoplasmosis during pregnancy is based on maternal serology due to asymptomatic nature of the disease.

In the present study, the seroprevalence of IgM & IgG Toxoplasma antibodies in healthy pregnant women were 5% and Nil respectively, where as in women with BOH was 11.9% and 20.9% respectively .This suggests that a relationship exists between BOH and maternal toxoplasmosis. Earlier studies also showed higher prevalence in women with BOH than in normal pregnant women.
Our study showed 5% incidence of Toxoplasma antibodies in normal pregnant women. This is in correlation with the studies done by Sandhu et al and Kumar et al (4%) [8].

In BOH cases, an incidence of 11.9% Toxoplasma IgM antibodies was noted in our study which is similar to studies done by Turbadkar et al (10.52%) [9] and Yasodhara et al (13.1%) [10]. In our study the IgG seropositivity for Toxoplasma was 20.9% this is correlated with the study done by Paschale et al (21.5%) [11] and slightly more than the Khurana et al (15.3%) [12].

Out of 134 cases, 90 cases (67.16%) were seronegative for both IgG and IgM Toxoplasma antibodies which shows that the BOH was due to causes other than Toxoplasma infections.

With reference to clinical presentation, the prevalence of Toxoplasma antibodies in our study shows 14.4% in recurrent abortions. Similar observation was made by Bhatia et al (12%) [13]. In IUD cases 13.3% which is correlated with Rajendra et al (17%) [14], congenital anomalies cases showed 13.3% which is similar to the study done by Swarna Kanta et al (15%) [15].

In our study, the seroprevalence with respect to age showed high prevalence of IgM antibodies in the age group of 30-36 compared to 24-29yrs. This agrees with the observations done by Bhatia [13] Chakrabharty et al [16]. This high incidence may be due to repeated infections over the years in these women or low immune response making this group less resistant to infections by Toxoplasma gondii.

Seropositivity of IgM antibodies in our study showed 8.3% in primi parous women and 13.3% in multi parous women. There is no significant difference in these groups. Similar observations made by Yasodhara et al [10].

In our study the IgM seroprevalence in urban and rural population is 11.4%, 12.7% respectively. There is no significant difference in these populations. It correlates well with study done by Khurana et al (16% and 22%) [12].

Toxoplasma infections are widespread among pregnant women and these infections can cause fetal abnormalities, but are treatable in comparison with other infections.

Ideally every women of reproductive age should know her Toxoplasma serologic status before conception [17]. If the patient has specific IgG antibodies, she is protected and there is no fetal risk unless she is immunocompromised. Women with negative serological tests are at risk of acquiring primary infection during pregnancy and educating regarding preventive measures should be provided. This shows that there is a regular need for screening of pregnant women for these infections and timely treatment to provide morbidity and mortality of infants born to such mothers [18].

The presence of IgM antibodies suggests, but does not prove an acute or recent infection because specific IgM antibodies remaining for 1 year and therefore it is important to distinguish IgM of primary acute / reinfection & reactivation using IgG avidity ELISA test developed recently to distinguish between them[7].

These data have a profound impact on the management of women with previous history of pregnancy wastages and positive serological reactions during the current pregnancy, highlighting the need for clinical
as well as laboratory studies to reduce adverse fetal outcome.

5. CONCLUSION:

The present study concluded that the seropositivity for IgM and IgG toxoplasma antibodies in BOH cases were higher than in normal pregnant women in our population and also there was significant increase in seroprevalence of toxoplasma IgM antibodies with an increase in age, parity and gestational period.

6. ACKNOWLEDGMENT:

The authors express sincere thanks to Dr.T.Kasturi Professor and HOD, Department of microbiology, Narayana medical college Nellore, Dr.P.Sreenivasulu reddy professor, Dr.Vasuundhara professor for your constant guidance and support. I also thank to my colleagues Dr. Jitendra assistant professor, Dr. Madhurima and PG’s.

7. REFERENCES


