Original Article

Seroprevalence of TORCH Infections and Adverse Reproductive Outcome in Current Pregnancy with Bad Obstetric History

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ABSTRACT

Introduction: *Maternal infections caused by TORCH [Toxoplasma gondii, Rubella virus, Cytomegalovirus (CMV), Herpes simplex virus (HSV)] and others agents like Chlamydia trachomatis,Treponema pallidum, Neisseria gonorrhoeae, HIV, etc. are the major causes of bad obstetric history(BOH).*

Aim of the study*: To evaluate the incidence of TORCH infections in pregnancy wastage in women with BOH.*

Methodology: *The study included 87 pregnant women with bad obstetric history. Cases with hypertension, diabetes mellitus, eclampsia of pregnancy, and Rh incompatibility were excluded from the study. Serological evaluation for TORCH infections was carried out by IgM Enzyme Linked Immunosorbant Assay (ELISA) method. Antenatal follow-up and delivery outcome was recorded.*

Result*: The IgM/IgG sero positivity to T. gondii, Rubella, CMV and HSV-2 was 5.8/8.0%, 4.6/90.8%, 9.2/95.4%, and 2.3/5.8% respectively. Adverse outcome occurred in, 66.8% and 43.1% cases of IgM and IgG seropositives. Maximum number of IgM seropositives cases of abortion (33.4%) were associated with Toxoplasma, Rubella and CMV infection, congenital malformations (25%) were associated with rubella and CMV infection , intrauterine fetal death (8.4%) was associated with CMV infection and the IgG sero positives showed abortion(31.3%),intra uterine death(7%), congenital malformations (3.5%) and still birth(1.2%) .*

Conclusion: *A previous history of pregnancy wastage and the serological reaction for TORCH infections during current pregnancy must be considered while managing BOH cases so as to reduce the adverse fetal outcome.*

Key words*: Bad obstetric history, TORCH, ELISA*

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INTRODUCTION

Bad obstetric history (BOH) implies previous unfavorable fetal outcome in terms of two or more consecutive spontaneous abortion, history of intrauterine fetal death, intrauterine growth retardation, still births, early neonatal death and/or congenital anomalies.[1] Infections caused by TORCH [*Toxoplasma gondii*, Rubella

virus, Cytomegalovirus(CMV), Herpes simplex virus (HSV)] and others agents like *Chlamydia trachomatis*, *Treponema pallidum*, *Neisseria gonorrhoeae*, HIV, etc are the major causes of BOH.[2,3] These pathogens usually cause only asymptomatic or mild infection in mother, but can cause much more serious consequences in fetus.[4]

The prevalence of toxoplasmosis in BOH is known to be significantly higher than those without it. A recent study from Chandigarh reports rising seropositivity to *toxoplasma* in women with BOH.[5] Sero-epidemiological studies have shown that 10-20 percent of women in childbearing age in India are susceptible to rubella infection. Infection with Rubella during pregnancy may lead to congenital malformation in 10-54 percent of cases.[3]

Maternal CMV is the commonest viral infection in perinatal period and is the leading cause of congenital CMV infection. [6] The incidence of congenital CMV ranges from 0.5-3.0% in all live births.[7] Primary HSV infection during first half of pregnancy is associated with increased frequency of spontaneous abortion, still birth, and congenital malformation.[8]

The prevalence of these infections varies from one geographical area to another. [1] These maternal infections can be established by demonstration of specific IgM antibodies. Hence a prospective study was designed to find the infections due to TORCH agents in pregnant women with BOH and the clinical outcome in such cases.

MATERIALS AND METHODS

A total of 87 pregnant women with previous unfavorable fetal outcome in terms of two or more consecutive spontaneous abortion, history of intrauterine fetal death, intrauterine growth retardation, still births, early neonatal death and/or congenital anomalies, in the age group of 19-36 years, attending the antenatal clinic at a tertiary care centre between May 2010 to April 2011 were studied. The approval of the institute's ethical committee was obtained prior to the sample collection. Informed written consent was obtained from all the participants. Detailed clinical history, physical examination, and conventional laboratory investigations were conducted. A preformatted questionnaire, including the socio-economic status, was completed during the antenatal follow-up period from gestation to birth. Delivery outcome was recorded for cases with reference to the gestational age and mode of delivery. Cases with hypertension, diabetes mellitus, eclampsia of pregnancy, and Rh incompatibility were excluded from the study.

3-5ml blood sample was collected under aseptic precautions and was allowed to clot and centrifuged at 3000rpm for 5min. The serum samples were stored in small screw capped vials at 20^{o}C until processed. The Samples were tested for the presence of IgM and IgG antibodies against *T. gondii,* Rubella virus, Cytomegalovirus and Herpes simplex virus using ELISA kits [Diagnova Eliscan ^{TM} RFCL Limited. Selaqui, Dehradun, Uttarakhand]. The tests were done as per the directions given in the manual supplied along with the kits. The optical density (OD) was read at 450 nm on the ELISA reader [Mindray MR-96A, Microreader]. The sero-titers were interpreted as Non-reactive (<0.9), Equivocal (0.9-1.1) and Reactive (>1.1)

as per the literature supplied along with the kits. Syphilis was diagnosed using RPR test. Tests for presence of HIV antibodies using 3rd generation, qualitative, sandwich immunoassay [Retroscreen-HIV, Qualpro Diagnostics 88/89,Phase IIC,Verna Industrial Estate ,Goa,India] and HbsAg using ELISA kits [Qualisa HbsAg, Qualpro Diagnostics 88/89, Phase IIC, Verna Industrial Estate, Goa, India] were done and the results were recorded in the proforma.

RESULTS

The age specific distribution and clinical presentation of 87 BOH cases is shown in Table 1. Majority of BOH cases [45(51.9%)] were found in females aged 25-30 years followed by [30(34.4%)] 19-24yrs.

Of the 87 cases, serological evidence for combination of IgM and IgG with any one of the TORCH agents was detected in 12 (13.8%) and IgG alone in 74(85.1%) [Fig-1a]. Sero-negativity to IgM/IgG was observed in one case (1.1%). The IgM/IgG antibody positivity to *T. gondii*, Rubella, CMV and HSV-2 was 5.8/8.0%, 4.6/91.7%, 9.2/96.4%, and 2.3/5.8% respectively [Fig-1b]. Age group of 25-30 years had the maximum serological evidence of either IgM or IgG for any one of the *Toxoplasma,* Rubella virus, Cytomegalovirus and Herpes Simplex viruses(2.3/3.4%,3.4/45.3%,4.6/44.1% and 2.3/2.3%) followed by 19-24yrs age group (1.2/4.6%, 0/34.8%, 2.3/37.2% and 0/2.3%) as shown Table 2. The number of seropositivity for IgM antibodies against *T. gondii*, Rubella, CMV and HSV-2 for either a single organism or in combination were 12(13.8%). It was observed that, antibody positivity was highest for CMV (33.3%) followed by combination of Toxoplasma and Rubella (16.7%), and CMV and Rubella (16.7%) [Tab - 3].

All the pregnant women with BOH included in the study were followed till delivery. Post-delivery adverse outcomes were observed in both IgM and IgG seropositives. The obstetric outcome results showed maximum number of IgM seropositives cases of abortion (33.3%) were associated with *Toxoplasma*, Rubella and CMV infection either alone or in combination, congenital malformations (25%) were associated with co-infections with rubella and CMV and mixed infection with toxoplasma, CMV and HSV, intrauterine fetal death (8.3%) was associated with CMV infection [Tab - 4a]. The IgG sero positives showed abortion (31.4%), intra uterine death (7%), congenital malformations (3.5%) and still birth (1.2%) [Tab - 4b]. The congenital malformations encountered were cardiac anomaly, anencephaly and meningomyelocele.

DISCUSSION

Maternal infections play a critical role in pregnancy wastage and their occurrence in patients with BOH or complicated pregnancy is a significant risk factor. [9] These infections cause fetal and neonatal mortality and an important contributor to early and later childhood morbidity. [10] All viral pathogens usually cause a primary maternal viremia which may infect the placenta and thereby the fetus with the exception of HSV-I or II, which causes an ascending infection via the genital tract to fetal membranes and then to the fetus. [11]

Women affected with any of these diseases during pregnancy are at high risk for miscarriage, stillbirth, or for a child with serious birth defects and/or illness and also a hazard to attending staff nurses. [12] Thus, these tests are performed before or as soon as pregnancy is diagnosed to determine the mother's exposure to *Toxoplasma,* Rubella virus, Cytomegalovirus and Herpes Simplex virus and the necessary precautions be taken.

Primary infection with TORCH complex in pregnant women can lead to adverse outcome, which is initially inapparent or asymptomatic and thus difficult to diagnose on clinical ground.[13] In this prospective study on 87 pregnant women with BOH, it was observed that incidence of *Toxoplasma*, Rubella, CMV, and HSV infection was more common in 25-30 yrs age group.

The seroprevalence of *Toxoplasma gondii* infections ranges between 7.7 and 76.7% in different countries (United Kingdom, 7.7-9.1%; Norway, 10.9%; India, 45%; Brazil, 50-76% and Nigeria 75.4%).[14-16] The seroprevalence of *Toxoplasma* IgM/IgG among pregnant women with BOH in our study was 5.8%/8.0% respectively. Sadik MS, et al.[17] *and* Turbadkar D, *et al.* [2] have also reported an incidence of 18%/6% and 10.5%/42.1% respectively. Janak K, et al.[11] reported overall IgM antibody positivity of 8.3% in 60 cases of BOH. The Indian studies showed varied results ranging from 11-55%.[18] Studies have proved that persistence of encysted forms of *Toxoplasma* in chronically infected uteri, and their rupture during placentation, lead to infection of the baby in the first trimester and often to recurrent miscarriages. [2,16,17,19]

In our study, the seroprevalence of the BOH cases for Rubella IgM/IgG were 4.6% /91.7% respectively. Surpam RB, *et al*. [18] and Yasodhara, *et al.*[20] have also reported overall IgM antibody positivity of 4.66 and 6.5% in cases of BOH while other workers reported seropositivity ranging from 4-17.7%.[21,22] Rubella is a mild viral illness in children but can occasionally infect adults. WHO estimates that, worldwide, more than 1 lakh children are born with congenital rubella syndrome each year, most of them in developing countries. Nearly 50% of rubella infection is subclinical.[23] 10-20% of women in child bearing age are susceptible to Rubella. Primary virus infection during pregnancy may cause fetal damage.[2] A review of literature identified, 15 rubella serosurvey in women of child bearing age in India which showed that susceptibility ranged from 5-45% reflecting pattern of rubella virus circulation.[23]

Primary CMV infection in pregnancy has a higher incidence of symptomatic congenital infection and fetal loss. This infection, being asymptomatic in adults, is difficult to diagnose clinically. [2] Demonstration of IgM antibodies is indicative of primary infection. The present study shows seropositivity rate of 9.2%/96.4% for CMV IgM/IgG in women with BOH. Several studies have reported between 84.5-95% prevalence of CMV IgG among pregnant women in Turkey. [16] The transmission of CMV infection to fetus occurs in 40% of the cases with primary infection and results in the delivery of 10-15% symptomatic and 85-90% asymptomatic congenitally infected newborns.[7]

Table 1: Obstetric history of BOH cases in various age group

Clinical presentations	Age in years			Total
	19-24 yrs	25-30yrs	31-36yrs	No (%)
	No (%)	No (%)	No (%)	
Abortion	17(19.6)	23 (26.5)	07(8.0)	47(54.1)
Intrauterine death(IUD)	07(8.0)	09 (10.4)	04(4.6)	20(23.0)
Still birth	03(3.4)	04 (4.6)	00(-)	07(8.0)
Congenital malformations	03(3.4)	09 (10.4)	01 (1.1)	13 (14.9)
Total	**30(34.4)**	**45(51.9)**	**12(13.7)**	**87(100)**

Table 2: Positive Serological Evidence for TORCH Infections in Various Age Groups.

Age in years	IgM No. (%)	IgG No. (%)
Toxoplasma		
19-24	01 (1.2)	04(4.6)
25-30	02 (2.3)	03(3.4)
31-36	02 (2.3)	00(-)
Rubella		
19-24	00(-)	30(34.8)
25-30	03(3.4)	39(45.3)
31-36	01(1.2)	10(11.6)
CMV		
19-24	02(2.3)	32(37.2)
25-30	04(4.6)	38(44.1)
31-36	02 (2.3)	13(15.1)
HSV		
19-24	00 (-)	02 (2.3)
25-30	02 (2.3)	02 (2.3)
31-36	00 (-)	01(1.2)

Table 3: Prevalence of IgM Seropositivity of TORCH either alone or in combination.

IgM antibody present for a single/ combination of organisms	No. of positive cases (%)
Toxoplasma	01(8.3)
Cytomegalovirus(CMV)	04(33.3)
Herpes simplex virus(HSV)	01(8.3)
Toxoplasma and Rubella	02(16.7)
Toxoplasma and CMV	01(8.3)
CMV and Rubella	02 (16.7)
Toxoplasma, CMV and HSV	01(8.3)
Total	12(100)

Table: 4a Association between IgM antibody to TORCH agents in pregnant women with BOH and their clinical outcomes

Clinical outcome of pregnancy	No. Of IgM positive cases (%)	Type of infection
Abortion	04(33.3)	*Toxoplasma* , *Toxoplasma* and Rubella, and *Toxoplasma* and CMV
Congenital malformations	03(25.0)	Rubella and CMV, *Toxoplasma* ,CMV and HSV
Intrauterine death(IUD)	01(8.3)	CMV
Normal delivery	02(16.7)	CMV and HSV
LSCS	02(16.7)	CMV
Total	**12(100)**	-

Mini PS, et al.[7] observed a seropositivity rate of 7.8% in women with obstetric problems. A positivity of 8-24% in women with BOH and other obstetric problems has been reported previously.[2,24]

Primary infection with HSV-2 acquired by women during pregnancy accounts for half of the morbidity and mortality from HSV-2 among neonates while the other half results from reactivation of an old infection.[8] The Seropositivity rate for HSV IgM/IgG among BOH patients in our study was 2.3%/5.8%. Turbakar, et al.[2] and Janak k, et al.[11] reported a seropositivity rate of HSV IgM as 3.6 and 3.3%. HSV in asymptomatic women with recurrent infection during pregnancy was found to be

Table: 4b Association between IgG antibody to TORCH agents in pregnant women with BOH and their clinical outcomes

Clinical outcome of pregnancy	No.of IgG positive cases (%)	Type of infection
Abortion	27(31.4)	Rubella,and CMV, *Toxoplasma* ,Rubella and CMV, Rubella, CMVand HSV
Intrauterine death(IUD)	06(7.0)	Rubella and CMV , *Toxoplasma* ,Rubella and CMV
Congenital malformations	03(3.5)	Rubella and CMV, Rubella.
Still birth	01(1.2)	Rubella.
Normal delivery	42(48.8)	Rubella and CMV, *Toxoplasma* ,Rubella and CMV , Rubella, CMV and HSV, CMV
LSCS	07(8.1)	Rubella, CMV and Rubella, and CMV
Total	**86(100)**	**--**

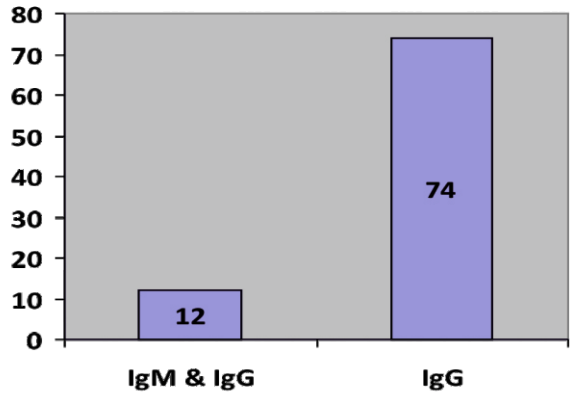

Figure 1a: Positive serological results of the combination of IgM & IgG and IgG alone with any one of the TORCH agents (No of cases-86)

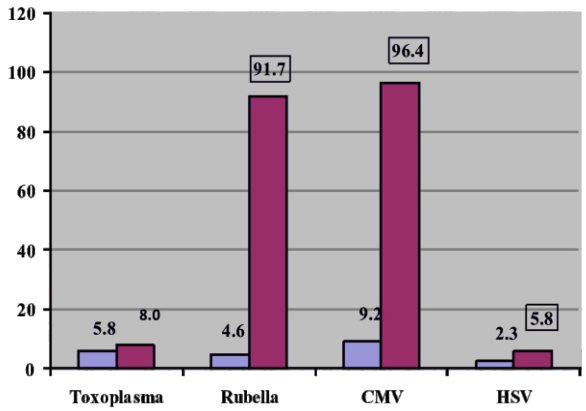

Figure 1b : Comparative Positive Serological Results of IgM and IgG TORCH Agents (No = 86cases)

0.6- 3% previously.[2] Studies had suggested that primary infection occurring in the first or second trimester caused an increase in spontaneous abortion and/or prematurity and fetal growth restriction. [25] It was also observed that in our study, mixed infections were noted in six cases of IgM and 76 cases of IgG seropositives cases. Similar observation of mixed infection has been made earlier. [2,26]

Congenital infections caused by *Toxoplasma gondii*, Rubella, and CMV are a significant cause of neonatal mortality and childhood morbidity worldwide.[10]In the present study, a very high percentage (66.8 and 43.1%) of adverse effects was observed in IgM and IgG seropositives. The study revealed adverse effects of abortion (33.3%), congenital malformations (25.0%) and IUD (8.3%) in 8 out of the 12 cases of IgM seropositives. The IgG seropositives also showed adverse effects of abortion (31.4%), IUD (7.0%) congenital malformations (3.5%) and still birth (1.2%) in 37 out of the 86 cases. It was noted that abortions were associated with co-infection with *Toxoplamsa* and Rubella in IgM seropositives and co-infection with Rubella and CMV in IgG seropositives respectively. The Congenital malformations detected in our study revealed co-infection with Rubella and CMV and mixed infection with *Toxoplamsa,* CMV and HSV-2 in IgM seropositives and one case of IUD had single infection with CMV.

CONCLUSION

In conclusion, this study has established that TORCH infections play a role on adverse fetal outcome in current pregnancy. We recommend that all antenatal cases with BOH, even if asymptomatic should be routinely screened for TORCH agents as early diagnosis and appropriate intervention will help to manage these cases and reduce adverse fetal outcome, diminishing the morbidity and mortality.

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