

# Seroprevalence of torch infections in bad obstetrics history in hiv and non-hiv women in solapur district of maharashtra india

## Abstract

TORCH infections are very important group of infection in day today clinical practice. Some of them are preventable and some are curable to some extent. The study was aimed at evaluating seroprevalence of TORCH infections in HIV negative and HIV positive females with bad obstetric history (BOH). The TORCH test was conducted on 605 HIV negative and 51 HIV positive women. A serological evaluation of TORCH infections was carried out by detecting IgG and IgM antibodies against these infections by ELISA method. Number of patients and percentage of HIV negative women were for *Toxoplasma* IgG 123 (20.66%) IgM 21 (3.47%), Rubella IgG 418 (69.09%) IgM 13 (2.15%), CMV IgG 481 (79.50%) IgM 12 (1.98%), HSV-1 IgG 302 (49.92%) IgM 1 (0.17%) and HSV-2 IgG 19 (3.14%) IgM 5 (0.83%). Number of patients and percentage of HIV positive women were for *Toxoplasma* IgG 21 (41.18%) IgM 5 (9.80%), Rubella IgG 50 (98.04%) IgM 4 (7.84%), CMV IgG 50 (98.04 %) IgM 0 (0 %), HSV-1 IgG 44 (86.27 %) IgM 3 (5.88 %) and HSV-2 IgG 14 (27.45 %) IgM 3 (5.88%).

In HIV negative women maximum seroprevalence was observed for CMV followed by Rubella and HSV-1 infection. In HIV positive women seroprevalence for Rubella, CMV and HSV-1 was found maximum and for *Toxoplasma gondii* and HSV-2 was found minimum. In HIV positive women almost all TORCH infections were found more than in HIV negative women. Studies suggest that early diagnosis and appropriate intervention of these infections may help in proper management of these cases.

**Keywords:** TORCH, Seroprevalence, Bad obstetric history, IgG, IgM, HIV positive women, HSV

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**Abbreviations:** TORCH, *Toxoplasma Gondii*, Rubella, Cytomegalovirus, Harpes Simplex Virus I and II; CMV, Cytomegalovirus; HSV, Herpes Simplex Virus; HIV, Human Immunodeficiency Virus; BOH, Bad Obstetrics History; ELISA, Enzyme Linked Immunosorbent assay; IgG, Immunoglobulin G; IgM, Immunoglobulin M

## Introduction

Bad obstetric history (BOH) implies previous unfavorable fetal outcome in terms of two or more consecutive spontaneous abortions, history of fetal death, intrauterine growth retardation, still birth, early neonatal death and/or congenital anomalies Kumari et al.<sup>1</sup> Viral infections in pregnancy are a major cause of morbidity and mortality for both mother and fetus. Infections can occur in the neonate transplacentally, perinatally or postnatally. Cause of BOH may be genetic, hormonal, abnormal maternal immune response and maternal infection Turbadkar et al.<sup>2</sup> TORCH infections in the mother are transmissible to fetus in the womb or during the birth process and cause a cluster of symptomatic birth defects. In mother they are inapparent or asymptomatic and hence difficult to diagnose clinically. Primary infections cause by TORCH infections- *Toxoplasma gondii*, rubella virus, CMV, HSV-1 and HSV-2 are the major cause of BOH (McCabe and Remington 1988). Recurrent pregnancy wastage due to maternal infections transmissible in-utero at various state of gestation can be caused by wide array of organisms; to name a few would be TORCH infections, *Listeria*, and HIV.

*Toxoplasma gondii* is a ubiquitous intracellular parasite causing serious infections in humans and domesticated animals. Rubella virus,

the only member of the Rubivirus genus, causes rubella (also known as German measles), an acute exanthematous infection of children and adults. The CMV is a member of Herpes viridae family and is classified as Human herpesvirus Type 5. The HSV-1 and HSV-2 are also known to cause abortion. The HSV-1 and HSV-2 are members of herpesvirus family, Herpesviridae, that infect humans. Changes in sexual behaviors of young adults may partly explain its higher incidence of HSV-2. Paz-Bailey et al.<sup>3</sup> Roberts et al.<sup>4</sup> Prevalence of these infections varies from one geographical area to another Kapil & Broor.<sup>5</sup> These maternal infections are initially unapparent or symptomatic and are, thus, difficult to diagnose on clinical grounds Daftary & Chakravarti,<sup>6</sup> Newton.<sup>7</sup> Therefore, diagnosis of acute TORCH infections in women is usually established by specific IgM antibodies Turbadkar et al.<sup>2</sup> Enzyme-linked Immunosorbent assay (ELISA) for IgM and IgG antibodies against these infections is highly sensitive and specific Thapliyal et al.<sup>8</sup>

A positive IgM result for *Toxoplasma*, Rubella and CMV, may not always indicate a primary acute infection, as IgM has a tendency to persist, even at high levels, after primary infection. False-positive IgM results may occur due to rheumatoid factor and antinuclear antibodies. Hence, IgG avidity testing is recommended to differentiate between primary infection, IgM persistence and reactivation. The measurement of IgG avidity for *Toxoplasma*, Rubella and CMV has proved to be a highly useful procedure, especially in combination with conventional serological assays. In case of *Toxoplasma gondii*, Rubella and CMV, a positive IgM accompanied by low-avidity IgG is suggestive of a primary infection, whereas a high-avidity IgG indicates either IgM persistence or reactivation. A low avidity index may also be seen in

a proportion of infected persons for months. Hence it is advised to perform IgM testing initially to point to the need for IgG avidity to avoid misinterpretation of results Hedman et al.<sup>9</sup>

Studies analyzing the role of maternal infection in the causation of BOH are less in number, probably due to lack of facilities in isolating etiological agents causing BOH and the prohibitive cost of commercial diagnostic kits. The area of investigation Barshi town of Solapur district of Maharashtra state, is a rural area lacking good medical, diagnostic and treatment facilities. Women having history of BOH are not tested for TORCH infections. There are no reports in the literature of TORCH infections of this area of investigation. Hence our aim of the present investigation is to find out the prevalence of TORCH infections in HIV positive and HIV negative women identified with BOH which help to prevent some of the TORCH infections and some can be cured to some extent.

## Materials and methods

This study at hospitals in Barshi, District Solapur, Maharashtra State, India was carried out during January 2010 to December 2014 over a period of five years. After obtaining institutional ethical committee's approval and informed consent from the patients, a total of 656 peripheral blood samples were collected of which 605 were HIV negative women and 51 HIV positive women with history of successive abortion fetus. Infections studied were *Toxoplasma*, Rubella, CMV, HSV-1 and HSV-2 called TORCH infections.

### Enzyme linked Immunosorbent assay (ELISA)

Both IgG and IgM antibodies were enumerated in the patient's serum by Enzyme Linked Immunosorbent Assay (ELISA). Both IgG and IgM antibodies were enumerated in the patient's serum by Sandwich ELISA kits commercial available in market, according to manufacturer's instructions and methods described in Sucilathangamet al.<sup>10,11</sup> Number of samples collected and analyzed for HIV negative women were 605 and HIV positive women were 51. Number of HIV negative women tested for TORCH infections were in the year 2010 (108), 2011 (141), 2012 (117), 2013(117) and 2014 (122). Number of HIV positive women tested for TORCH infections were 2010 (15), 2011 (9), 2012 (8), 2013 (13) and 2014 (6).

### Antibody avidity

The IgG antibody avidity for *Toxoplasma gondii*, Rubella and CMV was done as per method described by Bodeus & Goubau<sup>12</sup> and NML guide to services (2010) and Mubareka et al.<sup>13</sup>

## Results and discussion

This study showed seropositivity of the TORCH infections in the HIV positive and HIV negative women of Barshi town of the Solapur districts of Maharashtra, India. In India, there are few reports of seroprevalence of TORCH infections. Earlier studies reported that prevalence which is as low as 5% and as high as 80% in adult women Singh S,<sup>14</sup> Yashodhara et al.<sup>15</sup>

### TORCH infection in HIV negative women

**Toxoplasma gondii:** In the present study 605 patients HIV negative women were tested for TORCH infection and we found the *Toxoplasma gondii* specific IgG in 123 (20.66%) women while in IgM in 21 (3.47%) in HIV negative women.

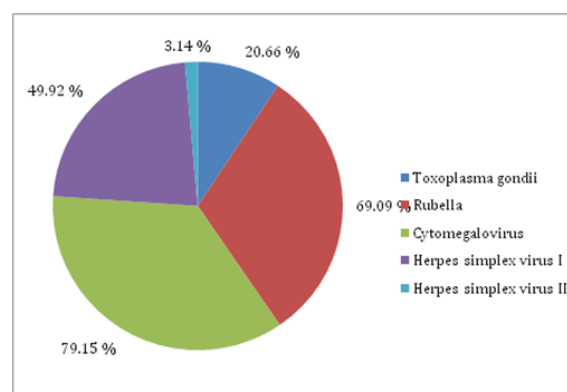
In the present investigation *Toxoplasma* specific IgG found was 20.66% and 3.47% IgM antibodies. The IgG and IgM antibodies were comparatively less in HIV negative women in literature where they

found 19.4% IgM antibodies in women. Similar more prevalence was observed (12%) for *Toxoplasma* IgM Yashodhara et al.<sup>15</sup> Results shown in Table 1 indicate number of patients those were found to exhibit existence of IgG and IgM antibodies within serum.

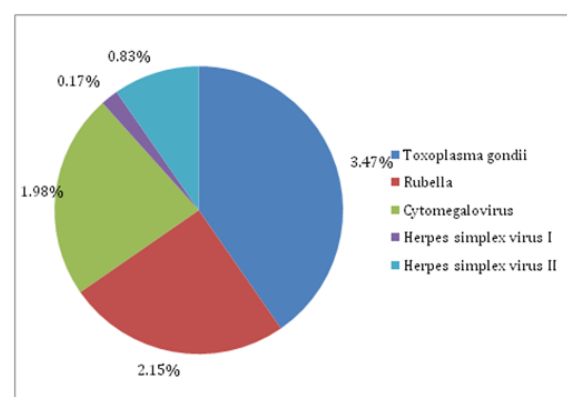
**Table 1** IgG and IgM antibodies for TORCH infections in HIV negative and HIV positive women

TORCH Infection	HIV Negative Women (n=605)		HIV Positive Women (n=51)	
	IgG	IgM	IgG	IgM
<i>Toxoplasma gondii</i>	125	21	21	5
Rubella	418	13	50	4
Cytomegalovirus	481	12	50	0
Herpes simplex virus I	302	1	44	3
Herpes simplex virus II	19	5	14	3

Results shown in Figure 1 denote percentage of IgG and IgM antibodies respectively, in HIV negative patients (n=605). The percentage of IgG was as high as 79.15% for Cytomegalovirus whereas lowest 3.14% for HSV-2 in HIV negative patients. The IgG 69.09% and 49.92% were for Rubella and HSV-1 in HIV negative patients while it was 20.66% for Toxoplasma (Figure 2). Existence of IgM antibodies in HIV negative patients was highest with Toxoplasma 3.47% followed by Rubella 2.15% CMV 1.98% HSV-2 0.83% and HSV-1 0.17% (Figure 2).



**Figure 1** IgG (%) of TORCH infection in HIV negative women (n = 605)



**Figure 2** IgM (%) in TORCH infection in HIV negative women (n=605).

In women with bad obstetric history (BOH), high rate 55.2% of Toxoplasma was reported in Nepal Rai et al.<sup>16</sup> and lowest one 19.44% was reported from Pune, India by Natu et al.<sup>17</sup> IgM in BOH high rate 42.5%, was reported by Shashi et al.<sup>18</sup> at Pune, India. The similar findings were reported by Sadik MS et al.<sup>19</sup> from Hyderabad, India with Toxoplasma (6.97% IgM). In India, reports

of seroprevalence studies were found for some cities. Prevalence of Maximum seroprevalence for *Toxoplasma* (10.52% IgM, 42.1% IgG) was observed in Mumbai Turbadkar et al.<sup>2</sup> In present studies prevalence was found (3.47% for IgM and 20.66% for IgG) next to Mumbai. The prevalence for *Toxoplasma* was 20.93% IgG, 6.97% IgM at Hyderabad Sadik MS et al.<sup>19</sup> Vellore 20.9% IgG, 11.9% IgM Ponutoti M et al.<sup>20</sup> Varanashi 19.7% IgM Sen et al.<sup>21</sup> Pune 19.44% IgG Natu et al.<sup>17</sup> and Pune 18.25% IgM, 09.73% IgG Lavan Singh et al.<sup>22</sup> Highest rate of prevalence for *Toxoplasma* IgG was (94%) was reported in Iraq by Mossa HAL<sup>23</sup> and in Bahrain 15.8% was reported by Tabbara & Saleh.<sup>24</sup> In case of BOH, IgG ranges between 77.1% in Iraq Aziz & Druish<sup>25</sup> and 6.84% from Iraq by Hadi NJ<sup>26</sup> While the range of *Toxoplasma* IgM was between 58% in Iraq Al-Khashab et al.<sup>27</sup> and 0.97% in Iraq by Razzak et al.<sup>28</sup>

Numerous reports stating seroprevalence of *Toxoplasma gondii* were observed of studies done in Iraq in a review articles by Abdulghani MA et al.<sup>29</sup> Studies were carried out in different cities and province of Iraq viz. Al Muthana, Al-Anbar, Al-Hila, Al-Tameem, Baghdad, Basrah, Divala, Diwana, Duhok, Kirkuk, Mosul, Najaf, Thi-Qar and Wasit province of Iraq. Of these reports seroprevalence IgM antibodies for *Toxoplasma* observed was between 4.84 to 81% and median at 41%. The highest IgG antibody seroprevalence of 81% was observed at Basrah and lowest at Kirkuk of 4.84% Abdulghani MA et al.<sup>29</sup> The seroprevalence of IgM observed was in the range 0.97% to 58.1% in various places of Iraq with median of 17%. The highest 58.1% IgM prevalence was observed at Baghdad and lowest at Duhok providence of Iraq. Seroprevalence in Egypt for *Toxoplasma* was 65% Hammouda et al.<sup>30</sup> and Nepal 55.20% Rai et al.<sup>16</sup>

**Rubella:** The present studies of Rubella antibodies were found IgG 69.09% and IgM (2.15%), in HIV negative women which is very high as compare to findings in Mumbai (26.00%) Turbadkar et al.<sup>2</sup> and IgM antibodies in 30.4% in Varanashi Sen et al.<sup>21</sup> The risk of the congenital Rubella infection following a maternal infection ranged from 5% to 50% in various studies, with an increasing severity when it was acquired in the first trimester of the pregnancy Miller et al.<sup>31</sup> But Rubella is such a mild disease, that not more than 1 in 10 cases are recorded.

Maximum seroprevalence for Rubella in India was observed in Bangalore India, 4.6% IgM, 90.8% IgG, Padmavathy M et al.<sup>32</sup> followed by Kirkuk, Iraq, 1.7% IgM, 89.1% IgG Aljumaili ZKM & Alsamarai AM,<sup>33</sup> Tamil Nadu, India 31.58% IgM, 78.9% IgG Ravindran V et al, 2012, Najaf, Iraq 4.66% IgM, 77% IgG Nama J et al.<sup>34</sup> Mumbai India 61.3% IgG, 26.8% IgM Turbadkar et al.<sup>12</sup> Hyderabad, India 29.06% IgG, 4.65% IgM Sadik MS et al.<sup>19</sup> and lowest prevalence at Kashmir, India for IgG 26.12%, IgM 8.96% Fomda et al.<sup>35</sup> was observed.

There is considerable variation in the prevalence of the Rubella IgG antibodies among the women of the child bearing ages, with studies suggesting a prevalence of a 71.3% Rubella immunity thus leaving 1/3rd of women susceptible to the Rubella infection Singla et al.<sup>36</sup> This history of the Rubella vaccination could not be gathered from the study population, but on the basis of socioeconomic status and the educational background in this rural area, it could be presumed that most of them had not been previously vaccinated. Hence we recommend proper monitoring, counseling and management and consideration for prior routine vaccination (Table 1) (Figure 1).

**Cytomegalovirus:** CMV is a member of the herpes virus's family and it is found universally throughout all the geographical locations and in the areas of low socioeconomic rural conditions. A majority of these infections are asymptomatic as others and they are difficult to diagnose

clinically. However, the rate of the primary infection is significantly higher in HIV negative women 79.15% IgG and 1.98% IgM. IgG avidity in most of the IgG positive patients for CMV suggests very high occurrence of CMV infection in HIV negative women in this area of investigation found 37.70 % seropositivity for IgM (Table 2).

**Table 2** No. of HIV negative women IgG avidity for *Toxoplasma gondii*, Rubella and Cytomegalovirus infections

Avidity of IgG	40-60%	60-80%	80-100%	Total
<i>Toxoplasma gondii</i>	10	16	24	50
Rubella	10	46	101	157
Cytomegalovirus	10	65	121	196

Maximum seroprevalence for Cytomegalovirus observed was at Kirkuk, Iraq 98.3% IgG, 8.3% IgM Zainab Khalid Moh et al.<sup>37</sup> Bangalore, India 9.2% IgM, 95.4% IgG Singhal P et al,<sup>38</sup> Mumbai India 91.05% IgG, 08.4% IgM Turbadkar et al.<sup>12</sup> Seroprevalence for Cytomegalovirus in present studies was found minimum than studied in India and abroad. It was 1.98% for IgM and 79.15% for IgG. But high IgG antibody avidity for Cytomegalovirus is suggestive of possible role in BOH. Therefore, it is necessary that behavioral and educational interventions for prevention are recommended and routine testing should be done in this rural area.

**Herpes simplex virus 1 :** Neonatal Herpes which can be acquired in utero from maternal infection is quite severe and it is associated with a high morbidity and mortality Sebastian et al.<sup>39</sup> In this study, the prevalence rate of the anti HSV-1 IgM antibodies was 0.83% and for IgG 49.92% (Table 3). The seroprevalence for Herpes simplex virus 1 was maximum 61% at Hyderabad, India Rebekah KP et al.<sup>40</sup> The seroprevalence for HSV-1 in the present investigation was 0.17 % IgM, 49.92 % IgG. The prevalence was followed by Mumbai India 33.58% IgG, 3.6% IgM, Kirkuk, Iraq (24.2% IgG), and the lowest Pune, India 07.40% IgM, 10.49% IgG Lavan Singh et al.<sup>2</sup>

**Table 3** No. of HIV positive women showing IgG avidity for *Toxoplasma gondii*, Rubella and Cytomegalovirus infections

Avidity of IgG	40-60%	60-80%	80-100%	Total
<i>Toxoplasma gondii</i>	1	2	8	11
Rubella	2	8	12	22
Cytomegalovirus	1	8	12	21

**Herpes simplex virus 2 :** In the present investigation minimum seroprevalence for HSV-2 was observed 0.83 % for IgM, 3.14 % for IgG as compared to other reports. Highest seroprevalence for HSV-2 was observed at New Delhi, India Shweta et al.<sup>41</sup> followed by Kirkik, Iraq 34.9% IgG, 2.7% IgM Hala Mohamed MH et al.<sup>37</sup> Jammu, India 07.5 IgG Rathore et al.<sup>42</sup> and the lowest prevalence was observed at Bangalore, India, 2.3% IgM, 5.8% IgG.

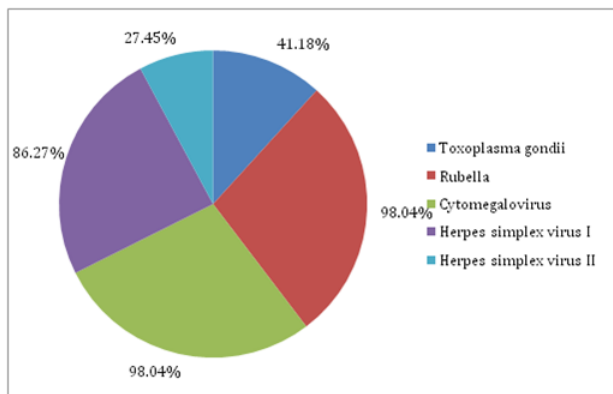
## TORCH infection in HIV positive women

In the present study 51 HIV positive women with BOH were tested for TORCH IgG and IgM antibodies. Number and percentage of HIV positive women were for *Toxoplasma* IgG 21 (41.18%) IgM 5 (9.80%) (Table 1) (Figure 3 & 4).

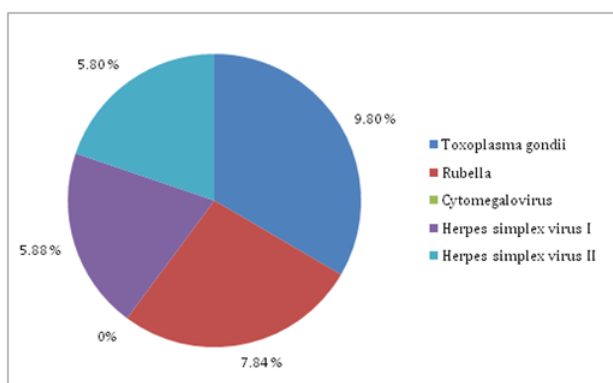
Studies conducted on 51 HIV positive women for detection of IgG and IgM antibodies indicated that high prevalence was noticed for Rubella, Cytomegalovirus and HSV-1 (Table 1). The antibody percent for IgG and IgM shown in pie diagram Figure 3 & 4 showed that as high as 98.04% IgG were detected for Rubella and cytomegalovirus, following to these two 86.27% IgG antibody were seen with HSV-1 infection while the IgG antibodies against *Toxoplasma* were 41.18% and the lowest was 27.45% for HSV-2 infection (Figure 3). The analysis



carried on serum for existence of IgM on (n=51) HIV positive women with BOH indicated highest prevalence for *Toxoplasma* antibodies 9.8% followed by Rubella with 7.84% and for HSV-1 and HSV-2 it was 5.88% and 5.8% respectively. Curiously, the IgM antibodies against Cytomegalovirus were not detected in either of HIV positive BOH patients (Figure 4).



**Figure 3** IgG (%) of TORCH infection in HIV positive women (n=51).



**Figure 4** IgM (%) of TORCH infection in HIV positive women (n = 51).

HIV infected patients are always at risk of developing illness due to infection with opportunistic microorganisms such as CMV and HSV-1 and HSV-2 and often have recurrent attacks of illness and complications. Opportunistic infections are signs of a declining immune system. Most life threatening infections occur when CD4 count is below 200 cells/mm<sup>3</sup>. Opportunistic infections are known to be the most common cause of death for people with HIV/AIDS. In the HIV/AIDS cases, *Toxoplasma* IgM and IgG antibodies were found 21.94% and 11.11% respectively at Pune India Lavan Singh et al, 2015), which is similar to the seroprevalence in the seroprevalence in a study in Nigeria (38.7%) (Ogoina et al, 2013). In the present study the seroprevalence for *Toxoplasma* was found more i.e. IgG 21 (41.18%) IgM 5 (9.80%).

Toxoplasmosis is an environmental disease as transmission of the infection has been shown to be promoted by poor environmental sanitation, overcrowding, eating habits, poverty and poor hygiene and many other factors. Similar condition exists in this area of the investigation. In the present studies most of HIV negative cases of acute and chronic toxoplasmosis present asymptotically but we found three cases developed *Toxoplasma* encephalitis of HIV positive women. In present investigation seroprevalence for Rubella was IgG 50 (98.04%) IgM 4 (7.84%). Seroprevalence for Rubella was found maximum as compared to published literature (Lavan Sing et al 2015).

The CMV is the most common cause of BOH in HIV positive women. Moreover, congenital CMV is the most frequently identified viral cause of mental retardation and is the leading nongenetic cause of neurosensory hearing loss. In the present investigation seroprevalence of CMV was IgG 50 (98.04 %), IgM 0 (0 %). This is similar to the seroprevalence in Mumbai Turbadkar et al.<sup>12</sup> Occurrence of Rubella and CMV at the same time in a patient was observed frequently which is suggestive of immunosuppression due to HIV infection.

The seroprevalence of HSV-1 found was for IgG 44 (86.27 %) and for IgM 3 (5.88 %). Similarly the seroprevalence for HSV-2 infection was IgG 14 (27.45 %) and IgM 3 (5.88%) (Table 1) (Figure 3 & 4). Herpes simplex virus having a sexual mode of transmission, in high risk group like HIV positive women it is agreeable to secondary prevention. Baccard-Longere et al.<sup>43</sup> found comparatively less prevalence of IgG 25.51% as compared to our studies where we found 86.27% HSV-1. Bhatia et al.<sup>44</sup> observed maximum seroprevalence for rubella (38.88% IgG) followed by CMV (22.22% IgG) and HSV-1 (10.49% IgG) infection in HIV positive patients. HIV positive women had more TORCH antibodies than HIV negative women which may be responsible for increased abortions in HIV positive women.

In case of *Toxoplasma*, Rubella and CMV, IgG antibody avidity was observed above 50% in most of the patients. Approximately 50% of pregnant women with primary CMV infection transmit CMV to their infants. Measuring CMV IgG avidity can reliably distinguish primary infection from non-primary infection during pregnancy and in case of BOH. The presence of high avidity antibodies at the 12 to 16 weeks of gestation indicates a past infection, likely prior to conception Gupta R et al.<sup>45</sup>

In a nutshell, this study showed that the seroprevalence of the TORCH antibodies was notable amongst the HIV positive women than HIV negative women who resided in Barshi taluka of the Solapur district of Maharashtra, India. It has already been emphasized that knowing the epidemiology of the TORCH infections is an important aspect in the development of strategies for the prevention of congenital infection Khalil KM et al.<sup>46</sup> Li Z et al.<sup>47</sup> Hence it should be recommended that all the BOH cases should be routinely screened for the TORCH infections to avoid adverse foetal outcomes. Ogoina D et al.<sup>48</sup> Moreover, similar studies as this one, which documents the seroprevalence of the TORCH infections, should be done at district or block level to create a baseline data in the country. In such a context the development of vaccine strategy against these infections, especially in the rural areas of developing countries, should be considered.

## Conclusion

BOH is a mental shock for a women and hence caring for pregnant women challenges family affecting society. Maternal testing with TORCH test helps to diagnose possible threat to the foetal that causes BOH. Most of the TORCH infections are symptomless and serodiagnosis can help to treat these infections, otherwise the outcomes are generally poor.

TORCH infections are very important infections for treatment of bad obstetric history. Some of them are preventable and some curable. Vinod R et al.<sup>49</sup> Zainab Khalil et al.<sup>50</sup> In HIV positive women TORCH infections were found more than in HIV negative women. We recommend that all antenatal cases with BOH be routinely screened for TORCH infections as early diagnosis and appropriate intervention of these infections will help in proper management of these cases in rural areas of India. TORCH test should be made mandatory for HIV positive and HIV negative women. Immune profile of TORCH in HIV negative and HIV positive women has good prognostic significance.

High level of seroprevalence for TORCH indicates the need for prevention with counseling and education of the patients. Our data demonstrating high frequency of TORCH infections in BOH support the conclusion that routine prenatal TORCH screening is justified.

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## Conflicts of interest

None.

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