# Seroprevalence of TORCH in Aborted versus non-aborted Women

## Abdul Haseeb<sup>1</sup>, Aamir Ali Khattak<sup>3</sup>, Azam Hayat<sup>2,</sup> Mujaddad ur Rehman<sup>2,</sup> Syeda Asma Bano<sup>4,</sup> Bilal Ahmed<sup>1</sup>, Ibrar Khan<sup>2,</sup> Aneela Rehman<sup>2,</sup> Rizwan Ullah<sup>2,</sup> Shafiq Barki<sup>2,</sup>

<sup>1</sup> Department of Medical Lab Technology, Abbottabad University of Science and Technology, Abbottabad Pakistan

<sup>2</sup>Department of Microbiology, Abbottabad University of Science and Technology, Abbottabad Pakistan

<sup>3</sup>Department of Medical Lab Technology, The University of Haripur, Haripur Pakistan

<sup>4</sup>Department of Microbiology, , The University of Haripur, Haripur Pakistan

Abstract- This study sought to determine the prevalence of TORCH (Toxoplasma gondii, Rubella virus, Cytomegalovirus, and Herpes simplex virus) infection in pregnant women's in Abbottabad, Pakistan. TORCH infections seriously complicate pregnancy in poor nations like Pakistan. These infections may spread from the mother to the foetus, increasing the risk of intrauterine foetal mortality and congenital abnormalities. The current study is to check for TORCH infection while pregnant. Compared to the preceding illness, the initial infection has a greater fatality rate. IgM and IgG antibodies can be used to prove primary infection since they signify primary or recurring infection.5000 samples that were brought to the Hamdard laboratory for routine testing were included in the current investigation. According to the company, the ELISA method was used to detect IgM and IgG. IgM and IgG antibodies can be found using this approach, which is sensitive and specific. According to the current study, pregnancy-related TORCH infections occur 16.28% of the time. Toxo is responsible for 16.1% of complete abortions, 23.3% of incomplete abortions, 21 % of missed abortions, and 10.8 threatened abortions. Rubella is also responsible for abortion in pregnant females. Rubella is responsible for 34.7% complete abortion, 38.5% incomplete abortion, 40.7% missed abortion and 45.9% threatened abortion. CMV is also a TORCH agent and is also responsible for abortion. CMV is responsible for 27.9% complete abortion, 25.2 % incomplete abortion, 14.4 % missed abortion and 35.1 % threaten abortion. HSV is playing a critical role in abortion. HSV is responsible for 21.1 % complete abortion, 12.8 % incomplete abortion, 23.6 % missed abortion and 8.1% threatened abortion. TORCH varies from sample to sample how frequently anti-Toxoplasma Rubella cytomegalovirus and anti-Herpes simplex virus IgM and IgG antibodies are present. This study shows that TORCH infections need careful attention since they can lead to major malformations and abnormalities in both children and pregnant women.

Keywords: TORCH, Toxo, Rubella, CMV, HSV, IgG, IgM.

#### I. INTRODUCTION

Toxoplasmosis, Rubella virus, Cytomegalovirus, and Herpes Simplex virus infection are collectively referred to as TORCH. These kinds of infections are the main reason for significant pregnancy problems. The majority of the time, the infection is so bad that it harms the foetus more than the mother [1]. Transplacental transmission allows these pathogens to reach the foetal bloodstream. Additionally, transmission may occur at any point throughout gestation or occasionally right before delivery [2]. Compared to recurring infections, the main infection has a higher mortality rate and can result in congenital abnormalities, abortion, sudden uterine death, growth retardation, preterm, and live-born newborns who have signs of disease [3]. These infectious diseases significantly increase mortality and morbidity in underdeveloped nations [4]. 10-15% of abortions are brought on by TORCH infections [5]. These illnesses are more common in some nations than others. Specific IgM antibodies can be used to identify these maternal illnesses. Multiple symptoms are brought on by the fetus contracting various illnesses intrauterinally, which causes the birth of the child. Inadequate vaccines, STDs, and animal exposures during pregnancy are all maternal risk factors. Due to the fact that fatal harm typically depends on gestational age, the timing of maternal infection is a critical epidemiologic element. The severe results from infections occur during the first trimester, with the exception of HSV. Rubella, Toxoplasma gondii, CMV, HIV, hepatitis B virus, HSV-1 and -2, and other pathogens like syphilis, parvovirus, and varicella are among the TORCH illnesses. [7,8]. The infections can spread throughout pregnancy via the transplacental pathway, during delivery via blood, or during labour and delivery via vaginal secretions. Infections after delivery usually have less of an effect. Others, including syphilis, hepatitis B, and HIV, can be spread through sex with a mother who is vulnerable. The correct immunisation of moms can prevent the spread of varicella and rubella. Perinatal infections are responsible for 2% to 3% of all congenital abnormalities. [9] Initial signs of infection may appear during pregnancy, at birth, in a baby's first year, or even years afterwards. Congenital infections might appear intrauterinally as aberrant growth or developmental patterns. The newborn infants with the infection may exhibit many clinical and laboratory problems, aberrant growth, or developmental defects.

Many clinical symptoms of viruses that appear in the first few days after birth coincide. They typically cause a rash that might be purpuric, petechial, or maculopapular (think blueberry muffin rash). Microcephaly, sensorineural hearing loss (particularly with CMV), and chorioretinitis are possible side effects. Hepatosplenomegaly and abnormalities of the heart are other frequent abnormalities. [10]. Toxoplasmosis and maternal CMV affect 2 to 10 neonates per 1000. [11] In countries where women are not immunized, rubella is common, but it only occurs in the US in cases of imported disease following a national immunisation programme. Because HSV-2 primarily causes genital infections, humans are the herpes virus' natural hosts. Babies frequently contract the virus. exposure to cats and eating food that has been improperly prepared, such as undercooked

meat or unpasteurized dairy products, are risk factors for toxoplasmosis. [12] 13]

## Pathophysiology

Transmission of Toxoplasma gondii oocysts happens when contaminated tissue is consumed or faecal particles are inhaled. Congenital toxoplasmosis is brought on through transplacental transfer. The third trimester of pregnancy is when this most frequently happens. However, congenital abnormalities will be more severe the earlier the infection occurs. [14] Syphilis can spread vertically within the womb or through the placenta. The rate of transmission among recently infected women is greater than 80%. [15] Aerosols can spread rubella to the mother, and the placenta can spread it to the foetus. [16]. Transfusions of blood, organ transplants, or most frequently contact with mucous membranes can all transmit CMV to the mother. The foetus or newborn then receives it via the birth canal, the placenta, or breast milk. Although it has long been assumed that CMV infection rates in primary infections are higher than those in secondary infections, some new research suggests that this may not be as true. [17] HSV is passed from the mother to the foetus through either an ascending infection or exposure during parturition [18]. The greatest proportion of newborn infections occurs during the third trimester of maternal primary infection. [19] Transmission to the neonate is 10 to 30 times less likely to occur with secondary reactivation of HSV. [20]. HIV can be passed from mothers to their unborn children during parturition, via transplacental transmission in utero, or through postnatal maternal exposures like breast milk. [21]. The most widespread sexually transmitted viral illness (STD) in the world is HSV. HSV1 is spread by non-sexual contact during childhood, but HSV2 is usually sexually transmitted and is the main cause of genital herpes [22, 23]. Herpes can take between 4 and 21 days to incubate. Primary genital HSV infection is asymptomatic in more than 75% of cases [24]. This infection continues to be a leading cause of morbidity and mortality in neonates [25-26]. Congenital, neonatal, and spontaneous abortion are all possible outcomes of genital herpes infection during pregnancy [27, 28].

Since there have been few investigations on the seroprevalence of TORCH agents in HRP in Abbottabad, Pakistan, the current study was designed to assess both IgG and IgM levels on a larger sample size in order to identify meaningful results.

#### **II. MATERIALS AND METHODS**

On samples that were sent to Hamdard Pathology Laboratory zarbat medical center Abbottabad between September 2021 and September 22 for analysis, the study was carried out. The simple random approach was used to randomly acquire these samples. 5000 samples from pregnant women who visited various clinics for routine check-ups were obtained for this investigation. Eight millilitres of blood were drawn in an aseptic manner from a simple tube without anticoagulant. Separated serum was kept at -20°C. These samples were examined using the ELISA technique in accordance with the manufacturer's instructions to determine whether IgM and IgG anti-Toxoplasma, Cytomegalovirus, Rubella, and Herpes simplex antibodies were present. Before testing samples, both positive and negative controls were used. For the purpose of this study's IgM and IgG antibody detection, the ELISA method was chosen because it is a sensitive method. A sample was considered to be positive if the test value was

greater than 1.2 and negative if it was less than 1.0. The range of values between 1.0 and 1.2 was seen as ambiguous. The test was redone after 1-2 weeks for people whose sample results were bordering on ambiguous. We included samples from expectant women who had a possible TORCH infection in the current investigation. Women who had a TORCH infection that was chronically present were excluded. The consent of each participant was obtained immediately. The Pakistani university's ethical committee gave the current study their clearance.

#### III. RESULTS

5000 blood samples from pregnant women were taken for the current investigation. Using the ELISA technique, IgM and IgG antibodies were checked to see if they were present in these samples. The average age of the women was 24. Out of 5000 samples, 764 tested positive for anti-Toxoplasma, cytomegalovirus, rubella, and herpes simplex, while 4236 tested negatives for these diseases. Table & fig 1 contains information on IgG and IgM levels

Organism Name	IgG	IgM
Тохо	45	211
Rubella	173	59
CMV	91	131
HSV	37	17
Total	346	418

Table 1: The table show the concentration of IgG and IgM in a pregnant woman against TORCH.



Figure 1: The Ratio of IgG and IgM in a pregnant woman against TORCH.

Now we separate the infected women according to their age. We categorize them into three groups. Group A female patients are in between 17-29 years, whereas group B, is in-between 30-39 years and the last group is group C where individuals are above 40 years. Group A has Young infected females who have a

higher ratio of antibodies IgA and IgM whereas group patients' lowest level of IgM and IgG. These show that group, A infected females has higher ratio of infection whereas group C have lowest level of infection. The group A infected patient's active immunity which tend to secrets more antibodies to cope the infection.

	Age group	Toxo IgG	To xo Ig M	Rubel la IgG	Rubel la IgM	C M V Ig G	CMV IgM	HSV IgG	HSV IgM
Grou p A	17-29	26	11 4	97	33	52	90	25	09
Grou p B	30-39	13	68	60	17	31	23	12	05
Grou p C	>40	06	29	16	09	08	18	00	03
	Total	45	21 1	173	59	91	131	37	17

Table2. This table shows the infection ratio and antibody production in different ages of pregnant females



Figure2. The graph shows the infection ratio and antibody production in different ages of pregnant females.

TORCH agents are responsible for abortion in pregnant females. We categorized them as complete abortion, incomplete abortion, missed abortion, and threatened abortion. Toxo is responsible for 16.1% of complete abortions, 23.3% of incomplete abortions, 21 % of missed abortions, and 10.8 threatened abortions. Rubella is also responsible for abortion in pregnant females. Rubella is responsible for 34.7% complete abortion, 38.5% incomplete abortion, 40.7% missed abortion and 45.9% threatened abortion. CMV is also a TORCH agent and is also responsible for abortion. CMV is responsible for 27.9% complete abortion, 25.2 % incomplete abortion, 14.4 % missed abortion and 35.1 % threaten abortion. HSV is play a critical role in abortion. HSV is responsible for 21.1 % complete abortion, 12.8 % incomplete abortion, 23.6 % missed abortion and 8.1% threatened abortion. The result shows that TORCH agents are equally responsible for abortion in pregnant females. However, our results show that rubella is more common in the Abbottabad population.

Organis m/Agent	Control	Complete Abortion (n= 118)	Incomplete Abortion (n= 210)	Missed Abortion (n= 76)	Threatened Abortion (n= 37)
Toxo (%)	Normal Delivery	19 (16.1%)	49 (23.3%)	16 (21.0%)	4 (10.8%)
Rubella (%)	Normal Delivery	41 (34.7%)	81 (38.5%)	31 (40.7%)	17 (45.9%)
CMV (%)	Normal Delivery	33 (27.9%)	53 (25.2%)	11 (14.4%)	13 (35.1%)
HSV (%)	Normal Delivery	25 (21.1%)	27 (12.8%)	18 (23.6%)	3 (8.1%)

Table 3: The table shows the abortion, incomplete abortion, missed abortion, and threatened abortion percentages by different TORCH agents.



Figure 3: The figure shows the abortion, incomplete abortion, missed abortion, and threatened abortion percentages by different TORCH agents

#### **IV. DISCUSSION**

This study shows that an average of 16.28% of pregnant women in Hamdard Pathology Laboratory zarbat medical center Abbottabad have TORCH infections. Studies carried out in India revealed a high incidence of TORCH infection of up to 80% and a low prevalence of 5% [29, 30]. In the current investigation, IgM antibodies (6.7%) were found in 764 samples, while IgG antibodies (58.250%) were present in samples with toxoplasma infection. In different countries, the prevalence of T. gondii diseases ranges between 7.7% and 76.7% (UK, 7.7%-9.1%, India 45%, Norway 10.9%, Nigeria 75.4%, and Brazil 50-76%) [31, 32]. Our findings come very close to the findings of an Indian study that found 3.47% IgM antibodies against T. gondii [33]. Sen et al[34] .'s report of 19.4% of IgM antibodies specific to Toxoplasma in India differs with the results of the current investigation. This distinction results from the fact that they included patients with a history of abortion. In our research, we discovered that 12 samples (1.5%) had IgM antibodies against rubella. In turkey, Tamer et al. [35] found that 0.2% of pregnant women had anti-rubella IgM sero-positivities. CMV, one of the herpes virus family members, is widespread around the world,

especially in places with low socioeconomic conditions. The majority of CMV infections are still asymptomatic and challenging to diagnose clinically. IgM and IgG antibodies against CMV were found in varying amounts in the current investigation. Between 17.5 and 52.3% of women who have had unplanned pregnancies or abortions are toxoplasma seropositive [36]. Age, nutritional condition, sociocultural practises, regional climate variations, the mode of transmission, and toxoplasmosis seropositivity all have an impact [37]. The infection this parasite causes affects one-third of the world's population [38, 39]. In the current study, 28% of HRP women had just IgG positive results, indicating remote infection, while 6% had both IgG and IgM positive results, indicating recent infection. The prevalence of toxoplasma seropositivity was 14.6% according to a study by Yasodhara et al. (2001) in the same region that focused solely on IgM seropositivity and used a smaller sample size [40]. With a higher sample size, the current study is the first from the South Zone of India to assess toxoplasmosis IgG and IgM seropositivity in high-risk pregnancies. In contrast, research from the West Zone by Mathur et al. (2002), Turbadkar et al. (2003), and Sood et al. (2009) revealed that the seropositivity was 28.5, 42.10, and 46.7% for IgG and 9.6, 10.52, and 41.3% for IgM, respectively [41, 42]. Women with H/o preterm labour had the highest seropositivity for toxoplasma in relation to the type of poor obstetric outcome, which was followed by H/o intrauterine foetal death, H/o repeated abortions, foetal congenital malformation in the current pregnancy, H/o congenital malformation in the previous pregnancy, and the least in relation to past H/o neonatal death. But a study by Shashi et al. (2004) from the North Zone revealed that among pregnant women with BOH, the highest seropositivity was for abortions (71.8%), followed by preterm delivery, stillbirths (22.2%), congenital abnormalities (4.8%), and neonatal death (1.2%) [43]This outcome is comparable to the Karad study [44], where 2.8% of participants had CMV IgM antibodies.

## V. CONCLUSION

In conclusion section, Rubella infections are common among TORCH illnesses and are associated with higher rates of abortion than other infections. It's likely that the prevalence of these illnesses has increased in Abbottabad Pakistan, but this is mostly owing to a lack of understanding, cultural restrictions, and a general reluctance among people to seek medical attention when pregnant. We also came to the conclusion that TORCH has a negative impact on pregnancy-related childbirth. Therefore, in order to lower the risk of morbidity and death in our region, we make an effort to learn about the prevalence and significance of these illnesses during pregnancy.

# VI. REFERENCES

[1] Boyer SG, Boyer KM. Update on TORCH infections in the newborn infant. Newborn Infant Nurs Rev, 2004, 4: 70-80.

[2] Kumar V, Abbas Ak, Fausto N, Aster JC. Robbins and cotran pathologic basis of disease. Saunders Elsevier, Philadelphia, 2009, 480.

[3] Maruyama K, Asai J, Ii M, Thorne T, Losordo DW, et al. Decreased macrophage number and activation lead to reduced lymphatic vessel formation and contribute to impaired diabetic wound healing. Am J Pathol, 2007, 170: 1178-1191.

[4] Das S, Ramachandran VG, Arora R. Cytomegalovirus and rubella infection in children and pregnant mothers - A hospitalbased study. J Commun Dis, 2007, 39(2): 113-117. Available from: http://www.ncbi.nlm.nih.gov/ pubmed/18338691

[5]. Singh L, Mishra S, Prasanna S, Cariappa MP. Seroprevalence of TORCH infections in antenatal and HIV positive patient populations. Med J Armed Forces India. 2015 Apr;71(2):135-8. [PMC free article] [PubMed].

[6. Stegmann BJ, Carey JC. TORCH Infections. Toxoplasmosis, Other (syphilis, varicella-zoster, parvovirus B19), Rubella, Cytomegalovirus (CMV), and Herpes infections. Curr Womens Health Rep. 2002 Aug;2(4):253-8. [PubMed]

[7]. Palma S, Roversi MF, Bettini M, Mazzoni S, Pietrosemoli P, Lucaccioni L, Berardi A, Genovese E. Hearing loss in children with congenital cytomegalovirus infection: an 11-year retrospective study based on laboratory database of a tertiary paediatric hospital. Acta Otorhinolaryngol Ital. 2019 Feb;39(1):40-45. [PMC free article] [PubMed]

[8]. Newton ER. Diagnosis of perinatal TORCH infections. Clin Obstet Gynecol. 1999 Mar;42(1):59-70; quiz 174-5. [PubMed]

[9]. McLeod R, Boyer K, Roizen N, Stein L, Swisher C, Holfels E, Hopkins J, Mack D, Karrison T, Patel D, Pfiffner L, Remington J, Withers S, Meyers S, Aitchison V, Mets M, Rabiah P, Meier P. The child with congenital toxoplasmosis. Curr Clin Top Infect Dis. 2000;20:189-208. [PubMed]

[10]. Rehman F, Shah M, Ali A, Ahmad I, Sarwar MT, Rapisarda AMC, Cianci A. Unpasteurised milk consumption as a potential risk factor for toxoplasmosis in females with recurrent pregnancy loss. J Obstet Gynaecol. 2020 Nov;40(8):1106-1110. [PubMed]

[11]. Cabral Monica T, Pinto-Ferreira F, Martins FDC, de Matos RLN, de Matos AMRN, Santos AC, Nino BSL, Pereira L, Narciso SG, Garcia JL, Freire RL, Navarro IT, Mitsuka-Bregano R. Epidemiology of a toxoplasmosis outbreak in a research institution in northern Paraná, Brazil. Zoonoses Public Health. 2020 Nov;67(7):760-764. [PubMed]

Anzivino Elena, Fioriti Daniela, Mischitelli Monica, et al. Herpes simplex virus infection in pregnancy and in neonate: status of art of epidemiology, diagnosis, therapy and prevention. Virol J. 2009;6:40. doi: 10.1186/1743-422X-6-40. [PMC free article] [PubMed] [CrossRef] [Google Scholar]

[12]. Cusini Marco, Ghislanzoni Massimo. The importance of diagnosing genital herpes. J Antimicrob Chemother. 2001;47:9–16. doi: 10.1093/jac/47.suppl\_1.9. [PubMed] [CrossRef] [Google Scholar]

[13]. Ural HSerdar genital herpes in pregnancy. [http://emedicine.medscape.com/article/274874-overview],

Accessed 18 November 2011.

[14]. O'Riordan DP, Golden WC, Aucott SW. Herpes simplex virus infections in preterm infants. Pediatrics. 2006;118(6):e1612–e1620.

doi: 10.1542/peds.2005-1228. [PubMed] [CrossRef] [Google Scholar]

[15]. Brown ZA, Benedetti J, Ashley R, et al. Neonatal herpes simplex virus infection in relation to asymptomatic maternal infection at the time of labor. New Engl J Med. 1991;324(18):1247–1252.

doi: 10.1056/NEJM199105023241804. [PubMed]

[CrossRef] [Google Scholar]

[16]. Brown ZA, Selke S, Zeh J, et al. The acquisition of herpes simplex virus during pregnancy. New Engl J Med. 1997;337(8):509–515.

doi: 10.1056/NEJM199708213370801. [PubMed]

[CrossRef] [Google Scholar]

[17]. Biswas D, Borkakoty B, Mahanta J, et al. Seroprevalence and risk factors of herpes simplex virus type-2 infection among pregnant women in Northeast India. BMC Infect Dis. 2011;11:325. doi: 10.1186/1471-2334-11-325. [PMC free article] [PubMed] [CrossRef] [Google Scholar]

[18]. Saso A, Bamford A, Grewal K, Noori M, Hatcher J, D'Arco F, Guy E, Lyall H. Fifteen-minute consultation: Management of the infant born to a mother with toxoplasmosis in pregnancy. Arch Dis Child Educ Pract Ed. 2020 Oct;105(5):262-269. [PubMed]

[19].Slutsker JS, Hennessy RR, Schillinger JA. Factors Contributing to Congenital Syphilis Cases - New York City, 2010-2016. MMWR Morb Mortal Wkly Rep. 2018 Oct 05;67(39):1088-1093. [PMC free article] [PubMed]

[20]. Pereira L. Congenital Viral Infection: Traversing the Uterine-Placental Interface. Annu Rev Virol. 2018 Sep 29;5(1):273-299. [PubMed]

[21]. Maltezou PG, Kourlaba G, Kourkouni E, Luck S, Blázquez-Gamero D, Ville Y, Lilleri D, Dimopoulou D, Karalexi M, Papaevangelou V. Maternal type of CMV infection and sequelae in infants with congenital CMV: Systematic review and meta-analysis. J Clin Virol. 2020 Aug;129:104518. [PubMed]

[22]. Pass RF, Arav-Boger R. Maternal and fetal cytomegalovirus infection: diagnosis, management, and prevention. F1000Res. 2018;7:255. [PMC free article] [PubMed] [23]. Belanger BG, Lui F. StatPearls [Internet]. StatPearls Publishing; Treasure Island (FL): Jul 26, 2021. Embryology, Teratology TORCH. [PubMed]

[24]. Kimberlin DW, Baley J., Committee on infectious diseases. Committee on fetus and newborn. Guidance on management of asymptomatic neonates born to women with active genital herpes lesions. Pediatrics. 2013 Feb;131(2):e635-46. [PMC free article] [PubMed]

[25]. Yah CS, Tambo E. Why is mother to child transmission (MTCT) of HIV a continual threat to new-borns in sub-Saharan Africa (SSA). J Infect Public Health. 2019 Mar - Apr;12(2):213-223. [PubMed]

[26]. Grant GB, Desai S, Dumolard L, Kretsinger K, Reef SE. Progress Toward Rubella and Congenital Rubella Syndrome Control and Elimination - Worldwide, 2000-2018. MMWR Morb Mortal Wkly Rep. 2019 Oct 04;68(39):855-859. [PMC free article]

[27]. Pomares C, Montoya JG. Laboratory Diagnosis of Congenital Toxoplasmosis. J Clin Microbiol. 2016 Oct;54(10):2448-54.

[28]. Yasodhara P, Ramalakshmi BA, Naidu AN, Raman L. Prevalence of specifc IGM due to toxoplasma, rubella,CMV and C. trachomatis infections during pregnancy. Indian J Med Microbiol, 2001, 19: 52-56.

[29] Singh SS. Mother-to-child transmission and diagnosis of Toxoplasma gondii infection during pregnancy. Indian J Med Microbiol, 2003, 21: 69-76.

[30] Nash JQ, Chissel S, Jones J, Warburton F, Verlander NQ. Risk factors for toxoplasmosis in pregnant women in Kent, United Kingdom. Epidemiol Infect, 2005, 133: 475-483.

[31] Allain JP, Palmer CR, Pearson G. Epidemiological study of latent and recent infection by Toxoplasma gondii in pregnant women from a regional population in the UK. J Infect, 1998, 36: 189-196.

[32] Tamer GS, Dundar D, Caliskan E. Seroprevalence of Toxoplasma gondii, rubella and cytomegalovirus among.pregnant women in western region of Turkey. Clin Invest Med, 2009, 32: E43-47.

[33] Karad D, Kharat A. Seroprevalence of torch infections in bad obstetrics history in HIV and non-HIV women in Solapur district of Maharashtra, India. J Hum Virol Retrovirol, 2015.

[34]. Gandhoke I, Aggarwal R, Lal S, et al. Seroprevalence and incidence of rubella in and around Delhi (1988-2002) Indian J Med Microbiol. 2005;23(3):164–167. doi: 10.4103/0255-0857.16587. [PubMed] [CrossRef] [Google Scholar]

[35]. la de L Galvan Ramirez M, Mancilla S, Castrejon OV, et al. Incidence of anti-toxoplasma antibodies in women with high risk pregnancy and habitual abortion. Rev Soc Bras Med Trop. 1995;28(4):333–337. doi: 10.1590/S0037-

86821995000400005. [PubMed] [CrossRef] [Google Scholar] [36]. Coelho RA, Kobayashi M, Carvalho LB. Prevalence of IgG antibodies specific to Toxoplasma gondii among blood donors in Recife, northeast Brazil. Rev Inst Med Trop Sao Paulo. 2003;45:229–231. [PubMed] [Google Scholar]

[37]. Dubey JP. The history of Toxoplasma gondii – the first100 years. JEukaryotBukaryotMicrobiol. 2008;55:467–475.doi: 10.1111/j.1550-7408.2008.00345.x. [PubMed]

[CrossRef] [Google Scholar]

[38]. DubeyJP, Jones JL. Toxoplasma gondii infection in humans and animals in the United States. Int J Parasitol. 2008;38:1257– 1278. doi: 10.1016/j.ijpara.2008.03.007. [PubMed] [CrossRef] [Google Scholar]

[39]. Yasodhara P, Ramalakshmi BA, Naidu AN, et al. Prevalence of specific IgM due to Toxoplasma, Rubella, CMV and C Trachomatis infections during pregnancy. Indian J Med Microbiol. 2001;19(2):79–82. [PubMed] [Google Scholar]

[40]. Mathur MS, Rele MC, Turbadkar D. Seroprevalence of HIV infection in bad obstetrical history and its correlation with TORCH and VDRL. Int Conf AIDS. 2002;14:7–12. [Google Scholar]

[41]. Turbadkar D, Mathur M, Rele M. Seroprevalence of torch infection in bad obstetric history. Indian J Med Microbiol. 2003;21(2):108–110. [PubMed] [Google Scholar]

[42]. Sood N, Soni S, Vegad M, et al. Seroprevalence of toxoplasma gondii in women with bad obstetric history in Ahmedabad. Gujarat Med J. 2009;64:2. [Google Scholar]

[43]. Chopra Shashi, Arora Usha, Aggarwal Aruna. Prevalence of IgM antibodies to toxoplasma, rubella and cytomegalovirus infections during pregnancy. JK Sci. 2004;6(4):190– 192. [Google Scholar]

[44]. Letscher-Bru Valerie, Pfaff AW, Abou-Bacar Ahmed, et al. Vaccination with Toxoplasma gondii SAG-1 protein is protective against congenital toxoplasmosis in BALB/c mice but Not in CBA/J Mice. Infect Immun. 2003;71(11):6615–6619. doi: 10.1128/IAI.71.11.6615-6619.2003. [PMC free article] [PubMed] [CrossRef] [Google Scholar]

#### AUTHORS.

**Correspondence Author** – Abdul Haseeb, Email: <u>abdul.haseeb576.ah@gmail.com</u>, alternate email address: <u>abdulhaseeb@aust.edu.pk</u> (if any), Contact number: +923155900089.

**First Author** – Abdul Haseeb Department of Medical Lab Technology, Abbottabad University of science and technology, Abbottabad Pakistan ORCID ID: 0000-0002-5231-2423 abdul.haseeb576.ah@gmail.com

**Second Author** – Aamer Ali Khattak Department of Medical Lab Technology, the University of Haripur, Haripur Pakistan amir.khattak@hotmail.com

Third Author – Azam Hayat Department of Microbiology, Abbottabad University of science and technology, Abbottabad Pakistan azamhayyat@yahoo.com

**Fourth Author** – Mujaddad ur Rehman Department of Microbiology, Abbottabad University of science and technology, Abbottabad Pakistan mujaddad@aust.edu.pk

**Fifth Author** – Dr Syeda Asma Bano Department of Microbiology, the University of Haripur, Haripur Pakistan Syeda.asma@uoh.edu.pk

Sixth Author – Bilal Ahmed Department of Medical Lab Technology, Abbottabad University of science and technology, Abbottabad Pakistan mlsuoh1992@gmail.com

Seventh Author – Ibrar Khan Department of Microbiology, Abbottabad University of science and technology, Abbottabad Pakistan Me\_abrarkhan@yahoo.com

**Eighth Author** – Aneela Rehman Department of Microbiology, Abbottabad University of science and technology, Abbottabad Pakistan Aneelarehman88@yahoo.com

Ninth Author – Rizwan Ullah Department of Medical Lab Technology, Abbottabad University of science and technology, Abbottabad Pakistan rizwanmicrobiologist@yahoo.com

**Tenth Author** – Shafiq Barki Department of Microbiology, Abbottabad University of science and technology, Abbottabad Pakistan Shafiqbarki9@gmail.com