

Study of *Toxoplasma gondii*, Rubella, CMV and HSV Antibodies among Pregnant Women in Pokhara, Nepal.

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Abstract:

Background: TORCH agents' causes ranges of infection in pregnant women and also in fetus, causing congenital anomalies and intrauterine fetal death. Detection of antibodies to these infections helps to establish primary, recurrent or past infections. Our aim of the study was to detect IgG and IgM antibodies against these infections agents among pregnant women by the CLIA Technique.

Methods: A total of 104 samples from pregnant women were studied. These samples were tested for IgG and IgM antibodies against *Toxoplasma gondii*, Rubella, CMV and HSV by LIAISON IMMUNOASSAY ANALYSER by CLIA Technique. Data were analysed using SPSS.

Result: The IgG/IgM seropositivity to *Toxoplasma gondii*, Rubella, CMV and HSV ½ were found to be 11.54%/1.92%, 95.19%/0.0%, 91.35%/0.96% and 81.73%/ 1.92% respectively. The primary infection by *Toxoplasma gondii* is higher among other TORCH infections

Conclusion: There is a prevalence of TORCH infections among the pregnant women. It is necessary to screen for TORCH agent among pregnant women since early diagnosis and treatment will help in proper management of the infected cases.

Keywords: TORCH, CLIA, IgG, IgM, Antibodies, Pregnant women

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I. Introduction

TORCH (Toxoplasmosis, "other", Rubella, CMV and HSV) was introduced decades ago to refer to the pathogens which causes infection in utero presenting with similar features (Ghazi et al 2002). The TORCH infections are group of viral, bacteria and protozoan infections which gain to the fetal bloodstream transplacentally via the chorionic Villi (Alsamarai & Aljumaili, 2013). Maternal infections transmissible in utero at various stages of gestation can lead to recurrent pregnancy wastage (Vyas & Acharya, 2018). The consequence of TORCH infection in fetus depend upon the type and virulent of the infecting agent and also the stage of pregnancy. Serious fetal complication can occur if these infections occur in first five month of pregnancy (Nabi et al, 2013).

Toxoplasma gondii is an obligate intracellular parasite, which can cause infection in all mammals. The serological studies show that *T.gondii* is the most predominant TORCH agent (Zeb et al. 2018). Infection may occur through ingestion of oocysts through close contact with infected cat or cat faeces or ingestion of water contaminated with oocyst or eating raw meat from infected animal that contain the tissue cyst or congenital infection (Ertug et al. 2005). Toxoplasmosis during pregnancy can lead to the infection in fetus which causes lesions normally involving in the brain and eye and may ultimately result in loss of the fetus (Song et al. 2005).

Rubella is a RNA virus belonging to paramyxovirus which is highly contagious (Zeb et al., 2018). Maternal infection early in the infection cause teratogenic effect in fetus and congenital rubella syndrome in infants. Maternal infection during first eight weeks after the last menstruation results all fetus become infected and mainly develop congenital defects (Uyar et al. 2008). The diagnosis of infection can be carried out using various techniques like isolation from nasopharyngeal secretion and detection of rubella specific IgM which can be estimated on 23rd week of pregnancy (Zeb et al., 2018).

Human CMV, HHV-5, belongs to the beta herpes family and it is the main cause of congenital viral infection associated with vision loss, permanent hearing loss and neurological defects (Neirukh et al., 2013). Since most women present nonspecific symptoms laboratory methods are required to diagnose acute CMV infection. IgG antigen avidity has been used to distinguish primary or non primary infections. The presence of CMV IgM may not be indicative of primary infection since it is also produced during reactivation and reinfection (Munro et al., 2005).

Herpes simplex virus (HSV) is an enveloped, doublestranded DNA virus, which belongs to the family of Herpesviridae and is transmitted across mucosal membranes and non intact skin that migrate to nerve tissues,

where they persist in a latent state. HSV-1 predominates in orofacial lesions, and it is typically found in the trigeminal ganglia, whereas HSV-2 is most commonly found in the lumbosacral ganglia. The acquisition of HSV infection during the pregnancy period has been associated with spontaneous abortion, intrauterine growth retardation, preterm labor and congenital and neonatal herpes infections (Dwyer & Cunningham, 1993). HSV-2 primary infections, especially during the last trimester of pregnancy are associated with high morbidity and mortality of the infant (Vesikari et al., 1999). The presence of HSV infection can be done one method by viral detection; a technique which is carried out by culture and HSV antigen detection by PCR and other method is the antibody detection either IgG or IgM specific to HSV1 and HSV2 in the laboratory (Dwyer & Cunningham, 1993).

II. Objectives

This study was aimed to determine the seroprevalence rate of IgG and IgM antibodies to common TORCH agents among pregnant women in Pokhara, Nepal. Most of the test performed in other few studies in Nepal was from ELISA technique. This was the first time that the investigation was done by fully automated immunoassay machine; LIAISON Immunoassay (Italy) using “Flash” chemiluminescence technology (CLIA).

III. Materials And Methods

The cross sectional study was done in from July to December 2018 on samples that came to Lifecare Diagnostics and Research Centre Pokhara Pvt Ltd. Lifecare Diagnostics is a grade "A" Laboratory as recognized by National Public Health Laboratory (NPHL), Ministry of Health, Nepal. In this study 104 sample from pregnant women were collected that came to various hospitals and polyclinics from Pokhara for routine checkup. Informed consent was taken from all the participants. 5 ml blood was collected in aseptic condition in plain Serum Separating Tube SST, without use of anticoagulant. Serum were separated and stored at -20 degree Celsius. These samples were tested in LIAISON Immunoassay Analyzer. LIAISON Analyzer adopts a “Flash” chemiluminescence technology (CLIA) with a paramagnetic microparticle solid phase (MP). These samples were tested for the presence of IgG and IgM antibodies against *Toxoplasma Gondii*, Rubella, CMV and HSV. Both Negative and Positive samples were processed along with their respective controls. Cutoff values were designed as per the manufacturer's instructions. The cutoff values for *Toxoplasma* IgG was <7.2IU/mL Negative, 7.2-8.8IU/mL; equivocal and ≥ 8.8 IU/mL; Positive. Similarly for *Toxoplasma* IgM <6AU/mL - Negative, 6-8AU/ml -Equivocal and ≥ 8 AU/ml- Positive. For Rubella IgG concentration <10IU/mL was considered Negative and ≥ 10 IU/mL as Positive. Similarly for Rubella IgM concentration <20 AU/mL was considered negative, 20-25 equivocal and ≥ 25 AU/mL as positive. Concentration of hCMV IgG <12U/mL was considered negative, 12-14 equivocal and ≥ 14 as positive. Similarly concentration of hCMV IgM <18U/mL was considered negative, 18-22 equivocal and ≥ 22 as positive. Index Value <0.9 Negative 0.9-1.1 Equivocal and ≥ 1.1 was considered for HSV1/2 IgG and IgM.

IV. Results

A total of 104 subjects were included in the study. All of the subjects were pregnant women with age ranging from 19 to 37 years with mean age of 26.6 years and median of 27 years. The age was categorized into 5 categories. Age less than or equal to 20 years, 21-25 yrs, 26-30 yrs, 31-35yrs and 35 and above. 49(47.11%) were ages between 26 to 30 yrs. The minimum legal age of marriage in Nepal is 20 years but girls are able to marry at 18 years with parental consent. Only 2 subjects were ≤ 20 yrs old.

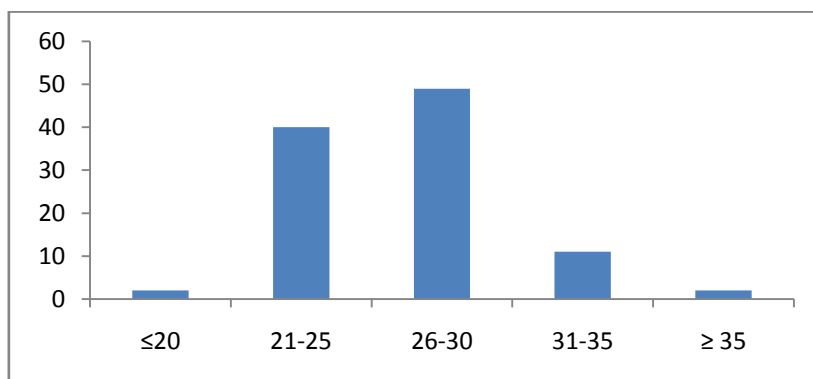


Fig. Study population by age.

Out of 104 subjects 12(11.54%) were positive for *Toxoplasma gondii* IgG, while 92(88.46%) were negative for *Toxoplasma gondii* IgG similarly 2(1.92%) was positive for *Toxoplasma gondii* IgM, while

102(98.08%) were negative for *Toxoplasma gondii* IgM, 99(95.19%) were positive for Rubella virus IgG antibody while 5(4.81%) women were negative for Rubella virus IgG. Interestingly all subjects were negative for Rubella virus IgM antibody. Out of 104 subjects, 95(91.35%) were positive for hCMV IgG Antibody while 9(8.65%) were negative for hCMV IgG antibody, while 1(0.96%) were positive and 103(99.04%) were negative for hCMV IgM antibody. 85 (81.73%) were positive for HSV1/2 IgG, while 19(18.27%) were negative for HSV1/2 IgG. Similarly 2(1.92%) was positive for HSV ½ IgM antibody and 102(98.08%) were negative for HSV 1/2 IgM antibody.

Table: 4.1 Descriptive data

Serological Test(n=104)	No of Positive Cases (%)	No of Negative cases (%)
Toxoplasma IgG	12 (11.54%)	92(88.46 %)
Toxoplasma IgM	2(1.92%)	102(98.08%)
Rubella IgG	99(95.19%)	5(4.81%)
Rubella IgM	0	104(100%)
CMV IgG	95(91.35%)	9(8.65%)
CMV IgM	1(0.96%)	103(99.04%)
HSV ½ IgG	85(81.73%)	19(18.27%)
HSV ½ IgM	2(1.92%)	102(98.08%)

V. Discussion

There are many studies of TORCH infections done in different parts of the world especially among the pregnant women. The acute infections which are caused by TORCH agents during pregnancy are often associated with adverse foetal outcomes and reproductive failures. The global prevalence of toxoplasmosis is different among the regions. Toxoplasmosis is present in every country and the infection rates from one country to another or even within the same country has variations. Study by (JENUM et al., 1998) among the 35,940 pregnant women found the 10.2% incidence of *T gondii*. This incidence is 1.4% greater than our study. (Zhou et al., 2011) found a 12.3% *T. gondii* IgG positive rate among 3281 individuals from the northeast and the south of China. The incidence rate is similar to our study. In the USA a recent seroprevalence study by (Jones, Kruszon-Moran, & Wilson, 2003) found that toxoplasma is prevalent in 14.9% women of child bearing age group. Similarly study by (Ghazi et al., 2002) among 926 pregnant Saudi women found the incidence of 35.6% IgG positivity which is quite high as compared to our study. Another study by (Alanazi et al. 2017) among 340 pregnant Saudi women showed 13.5% Toxoplasma IgG positivity which is different from the study by Ghazi. The study by Alanazi showed toxoplasma IgM positivity of 0.6% which is quite low among other studies. This showed that the prevalence of toxoplasmosis has been linked to several factors such as different climates in different regions and rural or urban settings. Study by (Josheghani et al. 2015) found a 37.5% Toxoplasma IgG positivity and 3.8% IgM positivity in Kashan, Iran. The IgG positivity was higher than our study while the IgM positivity is similar to our study. (Nabi et al., 2013) among 111 pregnant women in Dhaka showed 23.42% toxoplasma IgG seropositivity. The study also showed 0.9% Toxoplasma IgM positivity which is similar to our studies. The seroprevalence is quite high in AlBaiji area of Baghdad, Iraq as the IgM seropositivity studied by (Ali, Mohammed, & Talib, 2017) was found to be 18.63% which may be due to the fact that the area is an agricultural area and there are many animals keeper which makes high possibility of transmission of parasites. Study by (Zeb et al., 2018) Pakistan showed a toxoplasma IgM positivity as 2.5% which is similar to our study. Many studies in india showed different seropositivity rates. (Singh & Pandit, 2004) found overall IgG seroprevalence rate of toxoplasmosis as 45% while 3.3% were found to have IgM antibodies. The high percentage of IgG might be due to the inclusion of most women reported to have more than one pregnancy losses. The seropositivity of torch infections among patients with BOH was performed to find out its significance. (Khan, 2014) found Toxoplasma IgM positivity among 300 women in Aligarh, India with BOH and 75 control groups as 5.28% and 2.0% respectively. Similarly study by (Nellimarla & Kumari, 2017) in Kakinada town India, showed toxoplasma IgG positivity among BOH and control group as 30% and 16.6% respectively and IgM positivity among BOH and control group as 20% and 10% respectively. The difference in the seropositivity among two groups was found statistically significant. Data on seropositivity in the local population were not available from any part of Nepal. Study by (Sv Pradhan, 2015) among pregnant women with unfavorable previous pregnancy from eastern Nepal showed the IgM/ IgG sero-positivity to *T. gondii* as 26.2/9.2 %. The toxoplasma IgG positivity is lower than the IgM which indicates the more recent infection might be due to the fact that all the cases had a history of unfavorable pregnancy outcome.

The seroprevalence of Rubella IgG and IgM are different among the regions. Study by (Uyar et al., 2008) among pregnant women in Northern Turkey found Rubella IgG and IgM seropositivity as 94.3% and 1.7%. The Rubella IgG positivity of our study was 95.19% which is similar to their finding. Our study showed no recent rubella infection as the IgM positivity was 0% in our findings. (Tamer et al. 2009) showed IgG and

IgM positivity as 96.1% and 0.2% respectively. The epidemiology of rubella infection has been modified ever since the introduction of the rubella vaccination. Study by Ghazi et al among Saudi pregnant women shows rubella IgG antibodies in 92.1%. A study by Khan et al (2014) among women with BOH 4.6% has Rubella IgM while control group has 2.0% IgM positivity. A study by Nabi et al (2012) among pregnant women in Dhaka, Bangladesh showed IgG/IgM positivity as 81.08%/6.30%. The similar study by (Jubaida & Mondal, 2011) in Bangladesh found 84.33% IgG and 0.75% IgM Antibody positivity among the pregnant women. Similar study in Iran by (Josheghani et al., 2015) shows IgG and IgM positivity as 92.5% and 0%. None of the pregnant women enrolled in the study had Rubella IgM. This result is same as our result where our study also showed all negative Rubella IgM. CRS typically occurs in infants whose mother comes from countries without rubella immunization programmes. Study (Acharya et al., 2014) among women with spontaneous abortion in Dhulikhel hospital showed IgG and IgM seropositivity as 88.3% and 1.32% while the control group has IgG and IgM seropositivity as 88.3% and 0% respectively. These data shows higher IgG rates. This may be due to the inclusion of rubella vaccination in Nepalese routine immunization schedule has yield the high rate of IgG seropositivity which is suggestive for infection resistance. However (Subedi & Ghimire, 2016) reported a rubella IgG and IgM positivity as 7.76% and 43.68% among women with spontaneous abortion in first trimester. The rate is high since all the cases under study were the women with adverse pregnancy outcomes.

Maternal Cytomegalovirus (CMV) is the commonest viral infection in perinatal period and it is the leading cause for congenital CMV infection with a permanent hearing, vision loss and neurological impairment. The seroprevalence of hCMV is different among the regions. A study by (Hamdan et al. 2011) at El-Rahad hospital, Sudan to investigate seroprevalence of CMV among 231 pregnant women showed prevalence of 72.2% CMV IgG and 2.5% CMV IgM. (Uyar et al., 2008) in Northern Turkey found anti-CMV IgG antibody as 97.3%, while 1.0% was positive for the anti-CMV IgM antibody which is similar to our findings. Study by (Ali et al., 2017) in pregnant women in Baghdad showed 13.04% CMV IgM which is quite high as compared to our study. (Özdemir et al. 2011) found 0% positivity of CMV IgM among Turkish pregnant women. On the same study 98.8% were CMV IgG positive. (Zeb et al., 2018) found CMV IgM positivity among 800 pregnant women in Peshawar, Pakistan as 1.8% which is also similar to our findings. (Nellimarla & Kumari, 2017) studied TORCH infections in pregnant women with BOH around Kakinada Town, India which showed IgG and IgM percentage as 93% and 8% respectively among BOH group while against control group found IgG and IgM as 66.66% and 0% respectively indicating the seropositivity between two groups to be statistically significant. There are few studies done in Nepal in which a study by (Subedi & Ghimire, 2016) showed IgG and IgM seropositive for CMV as 41.74% and 19.41% among the women with spontaneous miscarriage. Only 41.74% were immune according to the study but in our study more pregnant were immune as the CMV IgG is quite high in our study. In a study conducted by (Sv Pradhan, 2015), 72.4% had IgG positivity while 4.5% had a recent infection. The prevalence is quite high as compared to our study.

The prevalence of herpes virus is variable with various factors may associated with the country and the region of residence, sex, sexual behavior and age. In our study HSV1 and HSV2 were detected in a single test. Most of the studies are similar to our studies while some study them separately. Study by (Zeb et al., 2018) in Peshwar, Pakistan found the incidence of HSV IgM as 1.12%. Similar study by (Ali et al., 2017) in pregnant women in Iraq showed 9.31% HSV IgM positivity. Study by (Prasoon et al., 2015) among 1158 high risk Indian women found 61% HSV IgG and 3% HSV IgM. Similarly (Josheghani et al., 2015) studied HSV antibodies among first trimester pregnancy in Kashan, Iran which showed 91.3% IgG and 7.5% IgM positivity. Separate study of HSV1 and HSV2 by (Nabi et al., 2013) in Bangladesh showed HSV1 IgG, HSV1, IgM, HSV2 IgG & HSV2 IgM seropositivity as 87.39%, 2.70%, 9.91% and 1.80% respectively. This shows that most acquire immunity against HSV1 than HSV2 and incidence of HSV 1 is more than HSV2. A study by (Nellimarla & Kumari, 2017) among BOH in India shows no HSV2 IgM while HSV2 IgG was 6.6%. In that study all the controls were also IgM negative. In another study in Northern India by (Khan, 2014) only 0.66% women with BOH were IgM positive while none of the control was IgM positive. However among 926 pregnant women in Saudi Arabia (Ghazi et al., 2002) found HSV1 IgG and HSV2 IgG as 90.9% and 27.1%. Seroprevalence of HSV2 among women with BOH shows higher positivity. A study by (Vyas & Acharya, 2018) in Rajastani women with BOH shows 13.3% IgM positive cases while only 2.38% were IgG positive. The HSV2 IgM in control was 4%. Study by (Tiwari, Arora, & Diwan, 2016) among aborted women in New Delhi India found 30.1% IgM positivity while only 6.6% of Controls were IgM positive indicating a significantly higher seropositivity in women with adverse pregnancy event.

VI. Conclusion

TORCH infections are the serious threats of congenital infections during pregnancy which may ultimately cause fetal damages or other anomalies. Therefore we make an effort to study the seroprevalence in Pokhara, Nepal. Most of the pregnant women had IgG antibodies against Rubella followed by CMV and then HSV and the last one Toxoplasma gondii while few had IgM antibodies against Toxoplasma gondii, CMV &

HSV. Absence of Rubella IgM shows the effectiveness of immunization programme. The prevalence of Toxoplasma gondii infection is high among other TORCH infections which may be related to the sanitations, lack of education and socioeconomic conditions. It is clear that there is a prevalence of TORCH infection in our part of our country. The vaccination approach and a public health policy could be effective especially in a developing country like Nepal to avoid undesirable fetal outcomes. Early diagnosis and treatment will help in proper management of the infected cases so it is recommended to all the pregnant women to be screened for TORCH agents. Furthermore the women with previous bad obstetric history are strongly advised to screen for TORCH agents.

References

- [1]. Acharya, D., Shrestha, A., Bogati, B., Khanal, K., Shrestha, S., Gyawali, P., & Gyawali Serological, P. (2014). Serological Screening Of Torch Agents As An Etiology Of Spontaneous Abortion In Dhulikhel Hospital, Nepal To Cite This Article, 2, 34–39. <https://doi.org/10.11648/J.Ajbls.20140202.11>
- [2]. Alanazi, F. B., Hassan, T. M., & Alanazi, W. F. (2017). Seroprevalence Of Toxoplasma Gondii Among Pregnant Saudi Woman In Arar, Northern Borders Province, Saudi Arabia. *Kasr Al Ainy Medical Journal*, 23(2), 104. https://doi.org/10.4103/Kamj.Kamj_13_17
- [3]. Ali, L. F., Mohammed, S., & Talib, M. (2017). Seroprevalence Of Anti-Toxoplasma Gondii, Anti-Rubella, Anti-Cytomegalovirus And Anti-Herpes Simplex Igm Antibodies In Pregnant Women In Baghdad. *Pakistan Journal Of Biotechnology*, 14, 785–790.
- [4]. Alsamarai, A. M., & Aljumaili, Z. K. M. (2013). Seroepidemiology Of Toxoplasma, Rubella, Cytomegalovirus And Herpes Simplex Virus -2 In Women With Bad Obstetric History. Part Ii. *Cytomegalovirus And Herpes Simplex Virus Infections*. *Our Dermatology Online*, 4(4), 536–544. <https://doi.org/10.7241/Ourd.20134.136>
- [5]. Dwyer, D. E., & Cunningham, A. L. (1993). 4 Herpes Simplex Virus Infection In Pregnancy. *Bailliere's Clinical Obstetrics And Gynaecology*, 7(1), 75–105. [https://doi.org/10.1016/S0950-3552\(05\)80148-8](https://doi.org/10.1016/S0950-3552(05)80148-8)
- [6]. Ertug, S., Okay, P., Turkmen, M., & Yuksel, H. (2005). Seroprevalence And Risk Factors For Toxoplasma Infection Among Pregnant Women In Aydin Province, Turkey. *Bmc Public Health*, 5(1), 66. <https://doi.org/10.1186/1471-2458-5-66>
- [7]. Ghazi, H. O., Telmesani, A. M., & Mahomed, M. F. (2002). Torch Agents In Pregnant Saudi Women. *Medical Principles And Practice: International Journal Of The Kuwait University, Health Science Centre*, 11(4), 180–182. <https://doi.org/10.1159/000065813>
- [8]. Hamdan, H. Z., Abdelbagi, I. E., Nasser, N. M., & Adam, I. (2011). Seroprevalence Of Cytomegalovirus And Rubella Among Pregnant Women In Western Sudan. *Virology Journal*, 8(October 2009), 2009–2012. <https://doi.org/10.1186/1743-422x-8-217>
- [9]. Jenum, P. A., Kapperud, G., Stray-Pedersen, B., Melby, K. K., Eskild, A., & Eng, J. (1998). Prevalence Of Toxoplasma Gondii Specific Immunoglobulin G Antibodies Among Pregnant Women In Norway. *Epidemiology And Infection*, 120(1), 87–92. <https://doi.org/10.1017/S0950268897008480>
- [10]. Jones, J. L., Kruszon-Moran, D., & Wilson, M. (2003). Toxoplasma Gondii Infection In The United States, 1999–2000. *Emerging Infectious Diseases*, 9(11), 1371–1374. <https://doi.org/10.3201/Eid0911.030098>
- [11]. Josheghani, S. B., Moniri, R., Taheri, F. B., Sadat, S., & Heidarzadeh, Z. (2015). The Prevalence Of Serum Antibodies In Torch Infections During The First Trimester Of Pregnancy In Kashan, Iran. *Iranian Journal Of Neonatology*, 6(1).
- [12]. Jubaida, N., & Mondal, M. (2011). Seroprevalence Of Rubella Antibodies In Pregnant Women. *Journal Of Armed Forces*, 7(1), 20–24. Retrieved From <http://Banglajol.Ubiquity.Press/Index.Php/Jafmc/Article/View/8621>
- [13]. Khan, H. M. (2014). Seroprevalence Of Torch Infection In Patients With Bad Obstetric History In And Around Aligarh , Northern India Nazish Fatima * Hiba Sami Nabeela Parvez Anwar Khan, 432–434.
- [14]. Munro, S. C., Hall, B., Whybin, L. R., Leader, L., Robertson, P., Maine, G. T., & Rawlinson, W. D. (2005). Diagnosis Of And Screening For Cytomegalovirus Infection In Pregnant Women. *Journal Of Clinical Microbiology*, 43(9), 4713–4718. <https://doi.org/10.1128/Jem.43.9.4713-4718.2005>
- [15]. Nabi, S. N., Wasey, A., Haider, K., Khan, A. A., & Hoque, M. M. (2013). Seroprevalence Of Torch Antibody In Pregnant Women. *Journal Of Armed Forces Medical College, Bangladesh*, 8(1 Se-Original Papers). <https://doi.org/10.3329/Jafmc.V8i1.13537>
- [16]. Neirukh, T., Qaisi, A., Saleh, N., Abu Rmaileh, A., Zahriyeh, E., Qurei, L., ... Azzeh, M. (2013). Seroprevalence Of Cytomegalovirus Among Pregnant Women And Hospitalized Children In Palestine. *Bmc Infectious Diseases*, 13, 528. <https://doi.org/10.1186/1471-2334-13-528>
- [17]. Nellimarla, K., & Kumari, R. L. (2017). Seroprevalence Of Torch Infections In Pregnant Women With Bad Obstetric History In And Around Kakinada Town, India, 6(4), 1899–1906. <https://doi.org/10.20546/Ijcmas.2017.604.226>
- [18]. Özdemir, M., Kalem, F., Feyzioğlu, B., & Baysal, B. (2011). Investigation Of Viral Pathogens During Pregnancy In A City Region In Turkey. *Anatolian Journal Of Clinical Investigation*, 5(2), 78–81.
- [19]. Prasoon, K. R., Srinadh, B., Sunitha, T., Sujatha, M., Deepika, M. L. N., Vijaya Lakshmi, B., ... Jyothy, A. (2015). Seroprevalence And Influence Of Torch Infections In High Risk Pregnant Women: A Large Study From South India. *Journal Of Obstetrics And Gynecology Of India*, 65(5), 301–309. <https://doi.org/10.1007/S13224-014-0615-3>
- [20]. Singh, S., & Pandit, A. J. (2004). Incidence And Prevalence Of Toxoplasmosis In Indian Pregnant Women: A Prospective Study. *American Journal Of Reproductive Immunology*, 52(4), 276–283. <https://doi.org/10.1111/J.1600-0897.2004.00222.X>
- [21]. Song, K.-J., Shin, J.-C., Shin, H.-J., & Nam, H.-W. (2005). Seroprevalence Of Toxoplasmosis In Korean Pregnant Women. *The Korean Journal Of Parasitology*, 43(2), 69–71. <https://doi.org/10.3347/Kjp.2005.43.2.69>
- [22]. Subedi, S., & Ghimire, S. P. (2016). Journal Of Nobel Medical College Relationship Of Torch Profile In First Trimester Spontaneous Miscarriage. *Journal Of Nobel Medical College*, 5(2), 17–21.
- [23]. Sv, P. (2015). Epidemiological And Serological Profiles Of Torch Infection In Pregnancy, 5, 705–708. <https://doi.org/10.3126/Jpn.V5i9.13690>
- [24]. Tamer, G. S., Dunder, D., & Caliskan, E. (2009). Seroprevalence Of Toxoplasma Gondii, Rubella And Cytomegalovirus Among Pregnant Women In Western Region Of Turkey. *Clinical And Investigative Medicine*, 32(1), 43–47.
- [25]. Tiwari, S., Arora, B., & Diwan, R. (2016). Torch Igm Seroprevalence In Women With Abortions As Adverse Reproductive Outcome In Current Pregnancy. *International Journal Of Research In Medical Sciences*, 4(3), 784–788. <https://doi.org/10.18203/2320-6012.Ijrms20160518>
- [26]. Uyar, Y., Balci, A., Akcali, A., & Cabar, C. (2008). Prevalence Of Rubella And Cytomegalovirus Antibodies Among Pregnant Women In Northern Turkey. *New Microbiologica*, 31(4), 451–455.
- [27]. Vesikari, T. (1999). Serological Evaluation Of Herpes Simplex Virus Type 1 And Type 2 Infections In Pregnancy. *Sexually*

- Transmitted Infections, 75(3), 168–171. <https://doi.org/10.1136/sti.75.3.168>
- [28]. Vyas, M., & Acharya, R. (N.D.). Seroprevalence Of Torch Infections In Women With Bad Obstetric History In North West Region Of Rajasthan, 3389, 355–357. <https://doi.org/10.21276/sjm.2018.3.7.3>
- [29]. Zeb, M. A., Jamal, S. F., Mir, A., Khan, A. A., & Ullah, A. (2018). Frequency Of Torch Infections During Pregnancy In Peshawar , Pakistan, 9(1), 22–26. <https://doi.org/10.1016/j.resuscitation.2012.12.002>
- [30]. Zhou, P., Chen, Z., Li, H.-L., Zheng, H., He, S., Lin, R.-Q., & Zhu, X.-Q. (2011). Toxoplasma Gondii Infection In Humans In China. Parasites & Vectors, 4, 165. <https://doi.org/10.1186/1756-3305-4-165>

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