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ORIGINAL ARTICLE

Evaluation of Herpes, Cytomegalovirus, Rubella, and Toxoplasma Gondii Infections in Swabi, KPK, Pakistan

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ABSTRACT

Introduction: Toxoplasma gondii (TG), Rubella virus (RV), Cytomegalovirus (CMV), and Herpes simplex virus (HSV) 1 and 2, globally most neonatal and infant deaths are reported due to this group of infections.

Purpose: The objective of this research study is to estimate the Prevalence of TORCH infections.

Methods: This research study was carried out at the Hi-Tech clinical laboratory Shewa Adda Swabi KPK Pakistan for a duration of 4 months from April 2022 to July 2022. Data statistics looking at the clinical characteristics of Toxoplasma gondii, CMV, RV, and HSV were filled from n=371. All samples were qualitative and quantitative tested by using Immune chromatographic (ICT) and Chemiluminescence Microparticle Immunoassay (CMIA) techniques.

Results: Out of n=317 pregnant women, n=131 (35 %) women were found positive while n=240 (65 %) women were found negative. Prevalence of Toxoplasma gondi 32 %, RV 28 %, CMV 27 %, and HSV 13 % in Swabi, KPK, and Pakistan. Among them, a high-frequency fraction of co-infection was detected for CMV and Rubella 17 (58 %), followed by CMV, Herpes, Rubella virus, and Toxoplasma gondii combined 7(25 %). The highest number of infected individuals 37 % we found in the age-group 21-25 years, followed by the 15-20 years age group 23 %, 26-30 years age group 17 %, 31-35 years age group 23 % while the lowest number of positive individuals found in >36 age group.

Conclusion: In brief context, TORCH pathogens have potentially shocking clinical manifestations. Hence, screening before pregnancy, and timely diagnosis of TORCH can reduce disease and death in both kid, and the mother. Moreover, knowing the epidemiology survey is a significant aspect to develop strategies and implementation for the prevention of disease.

Keywords: Pregnancy, Prevalence, TORCH, Swabi, Infections.

INTRODUCTION

In 1971, TORCH terminology was proposed as a group of maternal infections caused by Toxoplasma gondii, Rubella virus, Cytomegalovirus, and herpes simplex Virus 1 and 2, globally most of the neonatal and infants deaths are reported due to this group of infection TORCH[1]. The most commonly recognized judgment about TORCH is that they usually cause very mild disease in immunocompetent adults, but when the TORCH infection is acquired during pregnancy, it can cause serious illnesses and complications in the fetus as well as in the newborn[2]. These complications range from multiple abortions, intrauterine growth restriction, sterility, stillbirths, and congenital malformations to fetal death. All these complications are closely linked to gestational age during infection[3]. Since all these parental infections are primarily asymptomatic and the clinical identities are unreliable[4]. The epidemiology of all these infectious agents varies between low-income and middle-income countries, wherever a load of disease is highest, TORCH infections are the main contributors to prenatal, and infant morbidity, and mortality[5].

Toxoplasma gondii was originally identified over 100 years earlier in the tissues of mammals and birds. Toxoplasmosis infection is found worldwide. The etiological agent of toxoplasmosis is Toxoplasma gondii, a strict intracellular organism, which is ever-present in the surroundings, and whose only conclusive hosts are members of the feline family[6]. Toxoplasmosis is commonly benign and frequently goes overlooked in immunocompetent people however, non-specific flu-like signs, lymphadenopathy, and some rare impediments might be related to the topmost infection[4]. Many warm-blooded animals can be infected by this zoonotic parasite and a substantial zoonotic and veterinary parasite[7]. In a human being, T. gondii infection is commonly infected by the oral ingestion of infected tissue which contains cysts having bradyzoites nevertheless, it can also be acquired by the ingestion of oocysts that are the product of a sexual round in cat guts[8,9]. Infrequently it can spread through transfusion of infected blood, and organ transplantation[10]. Expectant women and immunocompromised persons such as AIDS, cancer, and transplant patients are high-risk groups for toxoplasmosis infection[11]. Owed to the Toxoplasma gondii infection fondness to the eye, brain, and eye, poor prognosis, and complications such as chorioretinitis, glaucoma, retinal detachment, encephalitis, and brain abscess can occur during acute or recrudescent stage infection[12]. Furthermore, cerebral toxoplasmosis infection may have also an important correlation to neurodegenerative disorders like epilepsy, schizophrenia, and bipolar disorder[13].

Rubella word is derived from the Latin word that means “little red”. This infection is also called the “third disease” it was formerly thought to be a form of scarlet fever or measles but from the year 1814, it was individualized as a distinct disease and named again as “German measles”[14]. Rubella virus (RV) is a member of the family Togaviridiae and the only member of the genus Rubivirus. It is a delicate virus, which can easily be destroyed by heat, detergents, and extremes of pH. RV structure contains RNA, a capsid, and a lipoprotein envelope. Protein (C) is the part of the capsid while the envelope comprises 2 glycoproteins E1 and E2. These proteins induce major immune responses[15]. Even if Rubella virus infection is usually a mild, occasionally asymptomatic disease in childhood, the magnitudes and consequences of Rubella virus infection in pregnancy can be shocking[16]. In 2010, the Pan American Health Organization proclaimed that the State of the Americas had eradicated the Rubella virus, and congenital rubella syndrome[17]. Approximately 80 % of cases of rubella infection is a is associated with a risk of congenital deformities if it is acquired in the first trimester of pregnancy[15]. In 2011, with this great progress toward the Rubella virus eradication goals, the 27th Pan American Sanitary Conference and resolution established in wave the process for the records as well verification of the stoppage of rubella transmission, and endemic measles[17].

Cytomegalovirus (CMV) is the main reason for intrauterine infection, happening in 0.2 % to 2.2 % of entirely live births, and is a common cause of mental retardation and sensorineural hearing loss[11]. Furthermost healthy individuals who obtain CMV after birth experience few or no signs and no lasting sequelae. Approximately experiencing a mononucleosis-like disease with indications including persistent fever, cervical lymphadenopathy, malaise,

myalgia, and, less commonly, hepatitis, and pneumonia[18]. Furthermore, when fetal infection prestige is unidentified, ultrasound deformities predict indicative congenital infection in one a third of cases[19]. Individual 10–15 % of congenitally diseased babies' current symptoms of contagion at birth and these newborns have a perinatal death rate of around 10 % with 70–80 % of surviving children presenting main neurological sequelae[20]. No nation does methodical screening for CMV infection amongst pregnant females as widespread screening for CMV main infection[18].

Herpes simplex virus type 1 (HSV-1) and type 2 (HSV-2) both are large double-stranded DNA viruses and belong to the Herpesviridae family[21]. Genital herpes is the chief cause of genital ulcers globally the frequency of herpes HSV type two infections in overall people range subsequently 10 to 60%. Maximum genital herpes infections are caused by HSV-2, even though HSV-1 accounts for around half of new cases in established countries[22]. The epidemiology of HSV-2 infections differs between females and boys, with a greater chance of spreading from male to female as compared from female to male[23]. The causes that affect a female's risk of HSV infection before pregnancy include, earlier onset of sexual activity poverty, the number of lifetime sexual partners, ethnicity, cocaine abuse, sexual behavior, and the presence of vaginosis especially vaginosis[24,25].

Nowadays many developed states have itemized TORCH analysis as a routine screening program for expectant women[26]. Similarly, the outbreak of intrauterine infection with the Zika virus in Brazil, which has run to microcephaly in babies, has over aroused global distress about TORCH infection[27]. Universally healthcare system has been moving with the service of information technology (I.T), which allows remote analysis by execution of non-invasive tests that even do not require venipuncture[28,29].Risk decline of TORCH infections mainly involve stoppage of infection in expectant women, which needs awareness about individual status during preconception and knowledge about the accurate way to stop each infection plus rules of vaccination/hygiene, and interaction with at-risk groups[30]. The main aim and objectives of this study are to aware the public and health care community regarding TORCH infection for early identification and efficient treatment that can decrease the infection burden on health care setup, motility, and morbidity of mother and child. A recommended reasonable methodology is the annual and serial monitoring of TORCH pathogens in the healthy adult female population for early finding, prevention, and follow-up in deficient individuals after supplementation, which is strongly encouraged.

MATERIALS AND METHODS

This research study was carried out at the Hi-Tech clinical laboratory Shewa Adda Swabi KPK Pakistan for a duration of 4 months from April 2022 to July 2022. Data statistics looking at the clinical characteristics of Toxoplasma gondii, CMV, RV, and HSV were filled from n=371 women after taking the consent form. All specimens n=371 were collected from pregnant women at the Hi-Tech clinical laboratory.

All those pregnant women included who were willing to participate in the study. A universal sampling technique was used to acquire direct information from the study participants. All those pregnant women who were suffering from any systemic illness, as well as congenital infections and family history, were excluded from the research.

A total of 371 blood samples were collected from pregnant. Samples were collected in a yellow top Gel, and Clot activator tube. The complete history of every individual was noted for example Age, gestational age, and specific history. All samples were aseptically taken and labeled properly with a unique Identity number and sample-specific barcodes. Samples were initially centrifuged at 6000 revolutions per minute (Rpm) to obtain serum from the samples.

The principle of this test is the detection of infection antibodies and specific antigens. Specimen, kit, buffer, droppers, package insert, and timer. First of all, we brought the specimen and the test component was brought to room temperature. The device was placed in a clean aseptic place and plated place.2nd the step device was labeled with an ID number, then take two drops were transferred with a disposable dropper into the well. Then we waited for the results for up to 15 minutes for the determination of CMV, Rubella, and Toxoplasma gondii antigen. If no visible line appears on the T-band, it means a negative result, whereas if a line appears on the T-line, it indicates a positive result.

The principle of the CMIA technique is based on the emitting of light during a reaction. In this method, a chemiluminescent particle is used as an indicator label to detect or quantify reactions. In the first step centrifuged the samples, Then centrifuged samples were and processed on a CMIA machine for the determination of immunological reaction of CMV, Rubella, and Toxoplasma gondii. Through processing on immunoassay, an exact quantity of specimen was drawn by the analyzer and was conceded through the column where antigen and antibody reacted with each other, which as a whole sensed by the machine detector and were displayed and the result was noted. The tests were done as per directions provided on the manufacturer's kit Abbott aidd Sligo, Ireland.

Data were analyzed to Microsoft Excel and STATA MP 14.2.

RESULTS

In this research study, we analyzed n=387 blood samples which were collected from pregnant women at the Hi-Tech clinical laboratory. All the samples are processed aseptically by ICT and CMIA. Out of n=317 pregnant women, 131 (35%) women were found positive while n=240 (65 %) women were found negative as results shown in figure/Table: 1.

Table 1: Number of positive and negative samples and percentages

Name of specimen	Number of specimens	Percentage %
Positive	131	35%
Negative	240	65%
Total	371	100

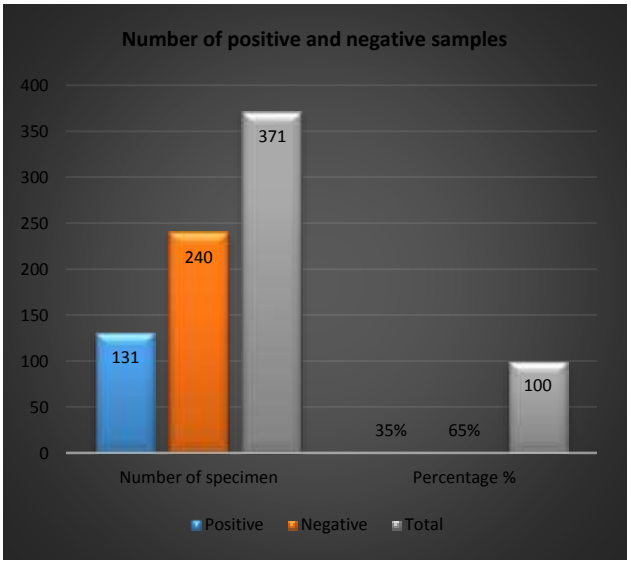

Figure 1: Number of positive and negative samples and percentages

In the current study n=131, pregnant women were positive TORCH profiles. Among TORCH infections frequency of different pathogens were that Toxoplasma gondi 32 %, RV 28 %,CMV 27 %, and HSV 13 %.The number and percentages are listed in Figure/Table No: 2.

Table 2: Distribution of different TORCH-positive pathogens n=131 out of n=371

Pathogen	Number of positive cases	Percentage %
Toxoplasma gondi	42	32 %
Rubella virus	36	28 %
Cytomegalovirus	35	27 %
Herpes Simplex Virus	18	13 %
Total	131	100

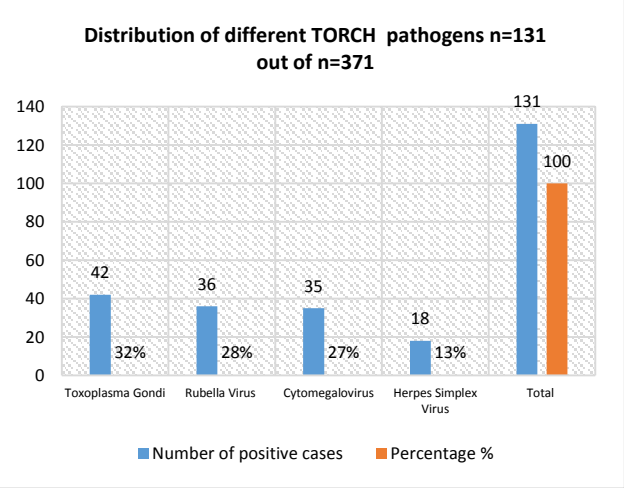

Figure 2: Distribution of different TORCH-positive pathogens n=131 out of n=371

In this study the highest number of infected individuals 37 % we found in the age-group 21-25 years and followed by the 15-20 years age group 23 %, the 26-30 years age group 17 %, 31-35 years age group 23 % while the lowest number of positive individuals found in **>36** age group only 8 %, detail description is listed below in Table/Figure: 3.

Table 3: Age-Wise distribution of TORCH infections

Age groups	Number of positive patients	Percentage of positive patients
15-20	30	23
21-25	49	37
26-30	22	17
31-35	20	15
>36	10	8
Total	131	100

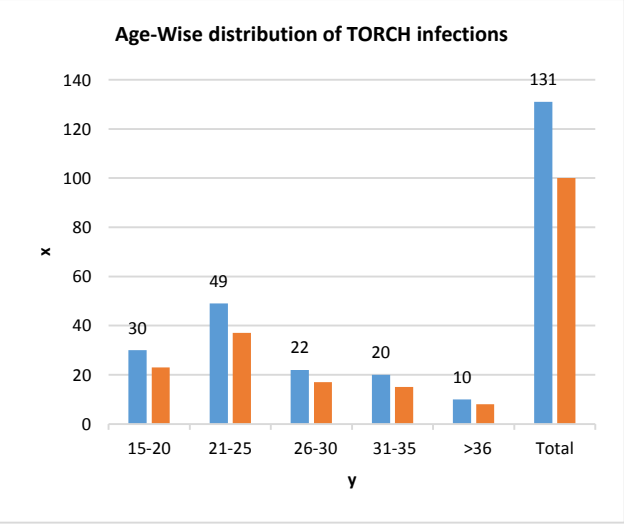

Figure 3: Age-Wise distribution of TORCH infections

Our research revealed the highest 58 % of co-infection found between CMV, and Rubella and followed by CMV, Rubella & Toxoplasma gondii, and Herpes, 25 %, and the lowest number of co-infection were found among CMV, Toxoplasma gondii, and Herpes, while no any co-infection was observed between Rubella & Toxoplasma gondii, detail description are listed in Table/Figure: 4.

Table 4: Co-infection of CMV, Rubella and Toxoplasma gondii, and Herpes

Co-infection of CMV, Rubella and Toxoplasma gondii, and Herpes		
Name of co-infection	Total reactive Samples	Total %
CMV, Rubella & Toxoplasma gondii, and Herpes	7	25
CMV & Rubella	17	58
CMV, Toxoplasma gondii, and Herpes	1	3
Rubella & Toxoplasma gondii	0	0
CMV, and Herpes	4	14
Total	29	100

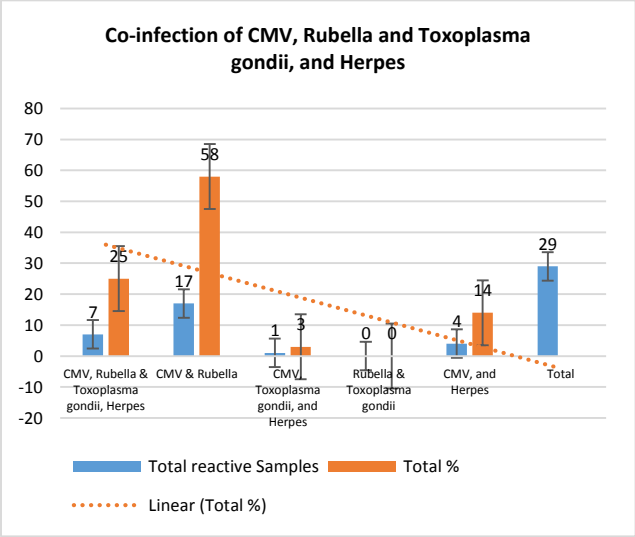

Figure 4: Co-infection of CMV, Rubella and Toxoplasma gondii, and Herpes

DISCUSSION

This current study was supported by a study conducted by Sadam et al., reported that 88.11 %, positive TORCH test while only and 11.88 % pregnant women were negative. Of 341 (88.11%) reactive samples[31]. A similar study conducted by Mocanu et al., in 2021, reported that 38 % of patients were positive for TORCH although the same diagnostic techniques for example ICT and CMIA were applied for the detection of Toxoplasma, CMV, and RV[32]. A study documented from the western area of Romania showed a very high prevalence of CMV 91.8 % among women in the reproductive period[33]. The current study was carried out in the urban area of the district Swabi and showed the highest prevalence of TORCH infections in the rural area as compared to an urban area, a similar study was reported [34]. The current study finding was in contrast to a study conducted by Zeb et al., from 2013-2015 2019 on pregnant women[35]. Another Like-wise study reported from china by Chen et al., in 2019,32 % prevalence of TORCH infections[36]. In India, a study was conducted on 4044 samples, and the seropositivity rate was observed at 33.46%[37].

This current finding contrast with a study conducted by Sadam et al., in 2021, the highest ratio of positive pathogens was noted in order of CMV 64.70 %, Rubella 67.74 %, and Toxoplasma gondii 72.7%[31]. An alike study reported by was shown in 2009 to determine the seroprevalence of different TORCH infections in pregnant women admitted at Omdurman Maternity Hospital, Sudan, the highest number of the pathogen was CMV 97.5 %[38]. The frequency rate of HSV infection in the current study was HSV

13 %, very closer to a study reported from India reported HSV is 17.43%[37].

The current finding regarding TORCH infections pathogens in pregnancy, similar to a study conducted in Peshawar Pakistan on pregnant women such as Toxoplasma IgM 2.5 %, rubella 1.5 %, Cytomegalovirus 1.8 % and Herpes simplex virus 1.1 % [35]. The frequency of T. gondii infection 7.7 %, and 76.7 % in several countries, UK, 7.7 %-9.1 %, India 45 %, Norway 10.9 %, Nigeria 75.4 % and Brazil 50-76 %[39].Another study reported the seropositivity to T. gondii, RV, CMV, and HSV was 5.8/8.0 %, 4.6/90.8 %, 9.2/95.4 %, and 2.3/5.8 % respectively [40]. A study reported from the region of In Croatia, reported that 29.1 % of T. gondii, 94.6 % of RV, 75.3 % of CMV, 78.7 % of HSV-1, and 6.8 % of HSV-2[41].

In epidemiological studies conducted among European expecting women, the prevalence of Toxoplasma gondii was 21.5 % in Italy, 24.6 % in Ireland, 33 % in Serbia, up to 46 % in Greece, and 48.6 % in Albania[42]. Roundabout studies have stated the middle of 84.5-95 % prevalence of CMV among pregnant women in Turkey[43]. A study reported on the prevalence TORCH infection from Khyber Pakhtunkhwa Peshawar, stated that the highest number TORCH infections were found in the age group 21-25. A study reported a high prevalence of HSV 38 % in the 17-24 age group[44].

Our study revealed the highest number of individuals were found in the 21-25 age group, while the same study was reported by[45]. Another study documented from KPK, Pakistan reported the highest number of infected individuals were found in the age group 18-24, 90.16 %[31]. A comparable study was documented in 2009 to determine the seroprevalence of TORCH infections, 25-32 years old individuals had the highest number of infections, while the lowest number of positive women was found in the 40-44 years old group. The maximum number of pregnant women was positive 77.2 % in the age group of 26—35 years[46]. The rate of simultaneous seropositivity to, CMV, Toxoplasma gondii, and Rubella virus among Romanian pregnant women reduced significantly between 2008–--2010, and 2015–2018 while the susceptibility to these infections increased[32].

Similar a study reported, the co-infection of CMV and Rubella, was 22.28 % and followed by for Cytomegalovirus, Rubella and Toxoplasma gondii[31]. In the northern region of India, the highest Coinfection was HSV with CMV observed[37]. Luckily, the current study revealed low co-infection as compared to a study conducted by[17].

CONCLUSION

The current study stated a high prevalence of Toxoplasma gondi 32 %, RV 28 %, CMV 27 %, and HSV 13 % in Swabi, KPK, and Pakistan. Among them, a high-frequency fraction of co-infection was detected for CMV and Rubella 17 (58 %), followed by CMV, Herpes, Rubella virus, and Toxoplasma gondii combined 7(25 %), and vice versa. The highest number of infected individuals 37 % we found in the age-group 21-25 years and followed by the 15-20 years age group 23 %, 26-30 years age group 17 %, 31-35 years age group 23 % while the lowest number of positive individuals found in >36 age group. In brief context, TORCH pathogens have potentially shocking clinical manifestations. Hence, screening before pregnancy, and early diagnosis of TORCH can reduce disease and death in both kid, and the mother. Moreover, knowing the epidemiology survey is a significant aspect to develop strategies and implementation for the prevention of disease. It is suggested to the aware community of the significance of TORCH infections to avoid complications. Good and fresh surroundings, a lifestyle, and clean drainage can help to prevent the TORCH infection.

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