Section: Microbiology

Original Research Article

SEROPREVALENCE OF CYTOMEGALOVIRUS AND RUBELLA VIRUS IN NON-PREGNANT WOMEN OF CHILD BEARING AGE

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ABSTRACT

Cytomegalovirus infection (CMV) is a global infection, even if there are significant differences in seroepidemiologic between and within countries. Present study was carried out in SGT medical college in the department of microbiology. A total of 180 blood samples were collected from non- pregnant females of child bearing age belonging to 18-40 years at SGT Hospital. These samples were tested for IgG antibodies against Cytomegalovirus and Rubella virus by ELISA. Prevalence of CMV IgG antibodies among non-pregnant females was recorded as 88.9% (160/180) and prevalence of Rubella IgG antibody recorded as 75% (135/180). Those 180 females were divided into three age group 18-25, >25 – 32 and >32 - 40 years as well as according to their residential community. Out of 180 females 160 were positive for CMV IgG antibody and out of 180 females 135 were positive for Rubella IgG antibody.

Keywords: Cytomegalovirus infection (CMV), Rubella virus and Seroprevalence.

INTRODUCTION

Cytomegalovirus (CMV) is a member of the Herpesviridae virus family, which causes active and reactivated viral infection. The primary source of cytomegalovirus are humans.[1,2] Infection with CMV during pregnancy is much more complex than other infections because the virus can often be reactivated during childbearing years and can be transmitted to the foetus despite maternal immunity and can lead to congenital infection with CMV. After primary CMV infection, a lifelong latent infection occurs.[3] It is globally endemic, and is more prevalent in developing countries, affecting the majority of the population where the seroprevalence of CMV IgG antibodies differs with a variety of epidemiological factors such as age, geographical distribution and marital status.[7] India, together with ten other member countries of the WHO South East Asia Region, resolved to eradicate measles and monitor rubella / congenital rubella syndrome (CRS) by 2020.[11] And prevalence in a study of Amritsar (Punjab) established 76.9% of non-pregnant women in the childbearing age group.[9] A survey of unvaccinated girls aged 10-16 in Tamil Nadu (South India) recorded 86.5 percent prevalence.[12] In this direction, in a phased way across the country, the Ministry of Health and Family Welfare has initiated measles - rubella (MR) vaccination campaign in the age group of 9 months to under 15 years 6. The campaign aims to promptly creating immunity in the population for both measles and rubella diseases to eradicate the disease. Due to measles and Rubella / CRS in the world, the measles-rubella campaign is a part of global efforts to reduce disease and death.[13,14] Many countries have applied the rubella vaccine to their national immunization programs, even without a global recommendation.[5] Collection of maternal antibodies against infection or immunization protects the foetus from rubella.

MATERIALS AND METHODS

Study area

- Department of Microbiology, Shree Guru Gobind Singh Tricentenary (SGT) Hospital, Chandu Budhera, Gurugram

Duration of study

- Six months (October 2019 to March 2020)

Sample size

- One hundred eighty blood samples from non-pregnant women attending SGT Hospital

Inclusion criteria

- Non – pregnant females of reproductive or child bearing age group (18 to 40years)
- Non - immunosuppressed patient

Exclusion criteria: Any acute illness and Pregnant women

Study Design: Observational study

Sample processing: - Blood samples received in the laboratory were allowed to clot for 10-12 minutes. Then the sample were centrifuged at 3000rpm for 5 min. The serum sample were stored in aliquots at -20˚C until processed. The test was done as per the directions given in the manual supplied along with the ELISA kits (CALBIOTECH). The desired number of coated strips were placed into the microtitre holder and wells were marked as blank, negative control, positive control and as calibrator. (fig 2) Samples to be tested were diluted by adding 10 ul of the sample to 200 µl of sample dilute and mixed well in order to prepare 1:21 dilution of the serum sample. In the accurate wells, 100 µl of dilute sera, calibrator and controls were dispensed. For the reagent blank, 100ul sample dilute in 1A well position was dispensed. The wells with sera and reagents were incubated for 20 minutes at room temperature. The wells were washed 3 times with 300 µl of 1X wash buffer. The wells were blotted on an absorbent paper until completely dried. It was equipped by adding the wash buffer contents (25ml, 20X) to 475ml of distilled or deionized water. Then 100µl of enzyme conjugate was dispensed to each other well and incubated for 20 minutes at room temperature than enzyme conjugate was removed from each well and wells were again washed with three times with 300 µl of 1X wash buffer. Than 100ul of TNB substrate was dispensed in each well and incubated for 10 minutes at room temperature. In the last step, 100µl of stop solution was added to each well. Optical density (OD) was read at 450nm using ELISA reader within 15 minutes.

Statistical Analysis: In this present study, written consent has obtained from the patient before data collection. Data has recorded in the presented proforma. The data has coded and entered into Microsoft Excel Worksheet. The categorical data has expressed as categorical data and has expressed as mean ± standard deviation. Result of the categorical data has analyzed by using person's Chi-square test.

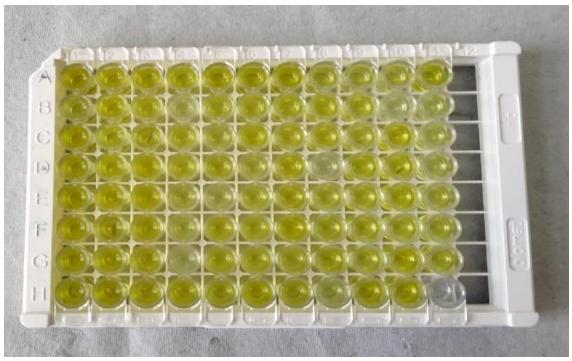

Figure 1: Microtiter pate showing reactivity to Cytomegalovirus

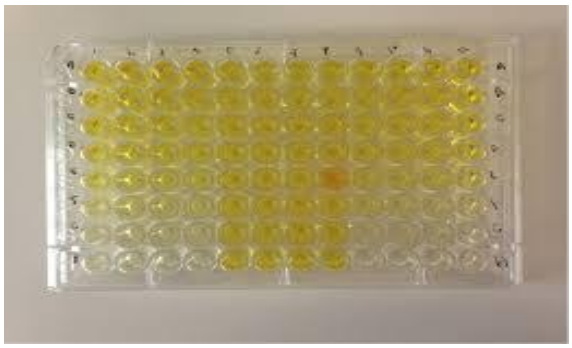

Figure 2: Microtiter plate showing reactivity to Rubella

RESULTS

The present study was conducted at SGT Medical College and Hospital, Gurugram, Haryana. In this prospective study, a total of 180 blood samples were collected from non-pregnant females at SGT hospital. These samples were tested for the presence of IgG antibody against CMV and Rubella virus by Enzyme linked Immune Sorbent Assay. The blood samples were specifically collected from non - immunized, non-pregnant women of child bearing age belonged to 18 – 40 years. Out of the total study participants, 85% (153/180) were from rural population and 15% (27/180) belonged to urban population.

Table 1: Distribution of study population according to various age groups and residential community

Age group	Frequency	Population	
	[n(%)]	Rural	Urban
18 – 25 years	39(21.6)*	21(53.8)**	18(46.6)**
>25 – 32 years	104(57.8)*	97(93.2)**	7(6.7)**
>32– 40 years	37(20.6)*	35(94.5)**	2(5.4)**
Total (n)	180(100)	153	27

* Total study participants in various age groups

**Calculated out of total CMV study participants in various age groupS

Table 2: Prevalence of CMV IgG antibodies among 180 study participants

S. No	Infectious Agent	Positive [n (%)]
1.	CMV	160 (88.9)

In this present study, we observed that the prevalence of CMV IgG antibodies among non-pregnant females in the study area was 88.9%.

Table 3: Distribution of IgG positivity among females of different age groups and different residential community (Rural and Urban)

Age Group	CMV IgG Positive [n(%)]	Population		*p* value (Rural vs Urban)
		Rural [n(%)]	Urban [n(%)]	
18- 25 years	**28 (71.7)***	**15 (53.5) ****	**13 (46.4)****	**(NS) >0.3**
>25 – 32 years	**97 (93.3)***	**93 (95.8)****	**04 (4.1)****	**< 0.001**
>32 – 40 years	**35 (94.5)***	**33 (94.3)****	**02 (6.5)****	**< 0.001**
TOTAL(n)	**160**	**141**	**19**	

In the present study, to determine the influence of age on CMV sero-positivity, non- pregnant females were divided into three age groups; 18 – 25 years, 26 – 32 years and 33 – 40 years respectively and according to their residential background i.e the location or area they belong.

Out of 180 patients, 39 non-pregnant females (21.6%) were from age group 18 – 25 years. Out of those 39 females, CMV IgG positives were observed among 28 of them. On the basis of population distribution, 15 (53.5%) females out of 28 sero-positives were from rural population while 13 (46.4%) females were from urban population respectively (Table 3). Among the age group 26 – 32 years, 104 females (57.7%) out of 180 were tested for CMV IgG antibody, out of which 97 females were positive. While classifying within rural and urban population, we observed that 93 (95.8%) females out of 97 were from rural population and only 04 (4.1%)" were from urban population (Table 3). In the age group 33 – 40 years, a total of 37 females (20.5%) out of 180 were tested out of which, 35 females were positive for CMV IgG antibody. Among those 35 positive females, 33 (94.3%) were from rural population and only a modest number of females 02 (6.5%) were from urban population (Table 3). Statistically significant difference is observed between IgG seropositivity of rural and urban females belonging to >25 – 32 years and > 32 – 40 years, whereas age group 18-25 years was found to be insignificant as significant p value was < 0.05 (Table 3).

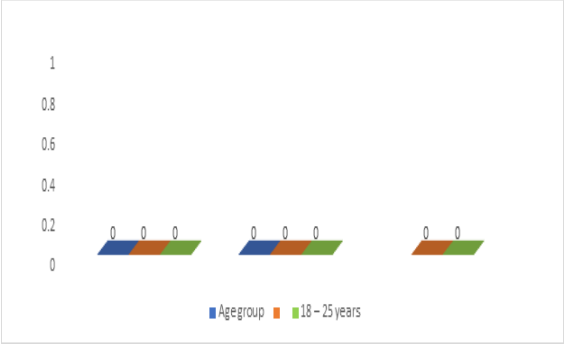

Figure 3: Distribution of IgG positivity among females of different age groups and different residential community (Rural and Urban)

Table 4: Prevalence of Rubella IgG antibodies among 180 study participants

S. No	Infectious Agent	Positive [n (%)]
2	Rubella	135 (75)

In this present study, we observed that the prevalence of Rubella IgG antibodies among non-pregnant females in the study area was 75%.

Table 5: Distribution of Rubella IgG positive females among different age groups and residential community

Age Group	Rubella IgG Positive	Population		*p* value (Rural vs Urban)
	[n(%)]	Rural [n(%)]	Urban [n(%)]	
18- 25 years	**21 (53.8)***	**12 (57.5)****	**09 (42.6)****	**(NS) > 0.4**
>25 – 32 years	**91 (87.5)***	**84 (92.3)****	**07 (7.7)****	**< 0.001**
>32– 40 years	**23 (62.1)***	**21 (91.3)****	**02 (8.7)****	**< 0.001**
Total (n)	**135**	**117**	**18**	

In the present study, to determine the influence of age on Rubella seropositivity, non-female was divided into three age groups; 18-25 years, 26 – 32 years and 33 – 40 years respectively and according to their residential background i.e the location or area they belong to rural as well as urban population (Table 1). Out of 180 patients, 39 non-pregnant females (21.6%) were from age group 18 – 25 years. Out of those 39 females, Rubella IgG positives were observed among 21 of them. On the basis of population distribution, 12 (57.5%) females out of 21 sero-positives were from rural population while 09 (42.6%) females were from urban population respectively (Table 5). Among the age group 26 – 32 years, 104 females (57.7%) out of 180 were tested for Rubella IgG antibody, out of which 91 females were positive. While classifying within rural and urban population, we observed that 84 (92.3%) females out

of 91 were from rural population and 07 (7.7%) were from urban population (Table 5). In the age group 33 – 40 years, a total of 37 females (20.5%) out of 180 were tested out of which, 23 females were positive for Rubella IgG antibody. Among those 23 positive females, 21 (91.3%) were from rural population and only a modest number of females 02 (8.7%) were from urban population (Table 5). Statistical significant difference is observed between IgG seropositivity of rural and urban females belonging to >25 – 32 years and > 32 – 40 years, whereas age group 18 -25 years was found to be insignificant as significant p value was < 0.05 (Table 5).

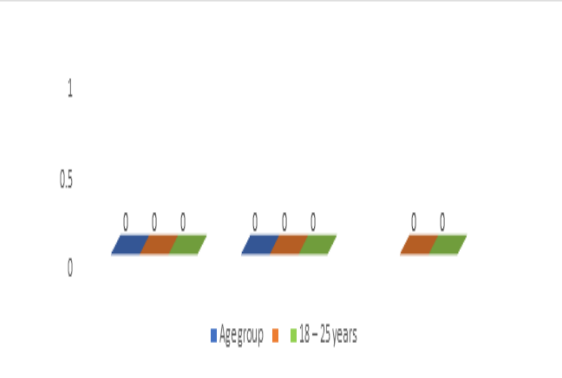

Figure 4: Distribution of Rubella IgG positive females among different age groups and residential community

Table 6: Combined CMV and Rubella IgG antibody positivity among females according to different age groups

Age Group	CMV IgG [n(%)]	Rubella IgG [n(%)]	Combined CMV and Rubella IgG Positivity	P value
18- 25 years n = 39	28 (20)	21 (15.56)	14	(NS)> 0.05
>25 – 32 years n = 104	97 (60.63)	91 (67.41)	85	(NS)> 0.6
>32 – 40 years n = 37	35 (19.37)	23 (17.03)	26	(NS)> 0.2
TOTAL	160 (100)	135 (100)	125	

In this present study, it was observed that out of 160 CMV IgG positive females and 135 Rubella IgG positive females, 69.44% of the females (125 out of 180) were positive for both CMV and Rubella IgG antibody. (Table 6). The distribution was also done according to the different age groups wherein we observed 14 females had both CMV and Rubella IgG from 1- 25 years of age, in the age group 26 - 32 years 85 females had IgG antibody of both CMV and Rubella whereas, 26 females from the age group 33-40 years had both of them.

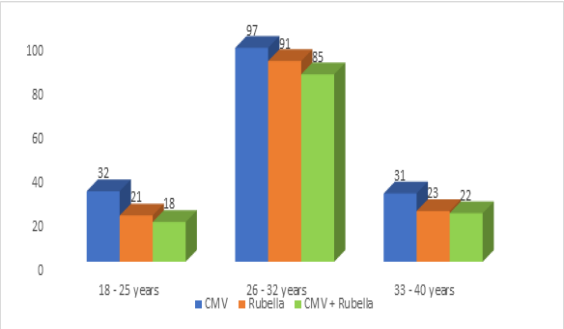

Figure 5: Combined CMV and Rubella IgG antibody positivity among females according to different age groups

DISCUSSION

Cytomegalovirus (CMV) effects both adult and children but primary infections in gestational months can lead to serious complications. CMV is an important pathogen in individuals whose immune system is immature or compromised, such as unborn child, So, it mostly causes congenital infection and can occur at any stage of pregnancy.[10] In this present study 180 non – pregnant females of child – bearing age attending SGT Hospital were included. These study participants were divided into three age groups 18 – 25, >25 – 32 and >32 – 40 years.[17] According to this study, the prevalence rate of CMV IgG antibody by was found to be 88.9% whereas, seroprevalence of Rubella antibody was observed to be 75%. Concordant to our study, a study conducted by Siennick et al. from Poland reported 81.9% prevalence of CMV IgG antibody among females of child bearing age. Whereas, another study of Jindal et al. from Amritsar, India documented the prevalence rate as 87.4% of CMV.[16,7]

In contrast to the present study some studies showed higher prevalence rate of CMV IgG such as 97.5 – 100%.[8,3,15] A study from America by Wang C, has reported 97.5% prevalence in non – pregnant women, another study from China shown 100% prevalence rate among college girls, whereas a study from Podlaska has recorded 98.3% prevalence rate.[18] Seroprevalence of Rubella, some studies have shown higher prevalence rate as compared to our study. 98.3% and 90.8% has reported from Poland and Nepal among non-pregnant women of child bearing age group.[49]

In our study there was an increasing trend in seropositivity with the increasing age. Seroprevalence of CMV among females of 18 - 25 years of age is 71.7%, 93.3% was seen among females of age between 26 - 32 years and highest among 33 - 40 years of age group that is 94.5%.[4] Similar study from Poland also showed the variability in seroprevalence of CMV according to different age groups. This influence of age group on seroprevalence was shown by the authors where they reported higher prevalence of CMV IgG antibody among females of 30- 40 years. In the present study 75% women were found to be seropositive which

implies that 25% of them are still susceptible and are at potential risk of acquiring Rubella infection. Concordant to our study, a study from Punjab have reported the same sepositivity,[19] According to our study, the influence of age assessed for rubella IgG positivity showed higher prevalence 87.5% among age group 26 - 32 years. A similar study from Punjab conducted by Singla et al have also shown the variation of seropositivity according to age, they reported higher prevalence rate in the age group 26 - 35 years.[20] Occupational exposure such as doctors, school teachers, nurses can be a factor that increases a chance of contracting Rubella infection in this age group. In our study most of the observations were similar to result from other studies conducted within India, the significant prevalence rate, comparative higher prevalence in rural population and low immunization against Rubella in the study area targets the necessities of women to be enrolled in education such as to maintain good hygiene, limited contact of pregnant women with infected children.

CONCLUSION

The present study demonstrates a high seroprevalence of CMV (88.9%) and Rubella (75%) antibodies among non-pregnant women of reproductive age, with a clear age-related increase in seropositivity. These results align with several national and international studies, though some report even higher prevalence rates. Notably, a considerable proportion of women remain vulnerable to Rubella infection, highlighting the urgent need to strengthen immunization coverage. The variation in seropositivity across age groups and the potential influence of occupational exposure further emphasize the necessity for focused public health strategies, including awareness campaigns, improved hygiene practices, and targeted interventions—particularly in rural areas and among women regularly exposed to children.

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