Sero-frequency of Cytomegalo virus among pregnant ladies attending Omdurman Maternity Hospital antenatal care

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Abstract:

**Background:** Human Cytomegalovirus (HCMV) is the most common cause of congenital infections. Virus infections during pregnancy may cause fetal or neonatal damage. Cytomegalovirus is associated with an 80% risk of congenital abnormalities if acquired in the first 12 weeks of pregnancy. The maternal immune status plays a major role in the likelihood of congenital infection. The aim of this study was to detect serofrequency (IgM and IgG) against HCMV and to detect relation between CMV seropositivity and other factors, age and trimester among Pregnant Ladies attending Omdurman Maternity Hospital Antenatal Care.

**Methods:** This is a cross-sectional study included (90) Pregnant Ladies aged between (10 – 50) years old conducted Omdurman Maternity hospital, Khartoum State, Sudan, during (December 2014 to May 2015). Serum specimen were collected and analysed for CMV antibodies (IgM and IgG), using enzyme-linked immunosorbent assay ELISA. Generated data were analyzed by using SPSS program.
Results: From a total number of (90) Pregnant Ladies out of them 25(27.8%), were positive for IgG antibody and 12(13.3%),were positive for IgM. This study shown statistically significant relationship between Sero-frequency of IgG-IgM of CMV and with risk factors Pregnant ladies (p=0.05).

Conclusions: As the immunity gap in the studied population was high, CMV vaccination should be provided for all women of child-bearing age and children.

Key words: Cytomegalovirus, IgG, IgM, ELISA, Omdurman Maternity Hospital Sudan, pregnant women

Introduction

CMV is DNA virus. Human are only known host like other members of cause latent infection and infect people on all age and socioeconomic group, Human cytomegalovirus (HCMV) is existing throughout all geographic locations and socioeconomic groups, and infects between 50% and 80% of adults in the United States\(^1\). HCMV is the virus most frequently transmitted to a developing child before birth. HCMV infection is more widespread in developing countries and in communities with lower socioeconomic status and represents the most significant viral cause of birth defects in industrialized countries\(^2\). CMV is the most common cause of congenital infection and its incidence has been estimated to be between 0.2- 2.2% of all live births in different parts of the world\(^3\). Most congenital infections are asymptomatic; only (10%) of infected fetuses will develop clinical signs of CMV infection. Transmission of CMV infection to the fetus has been identified in all trimesters of pregnancy. Abortion can result from ascending CMV endometritis and the virus has been isolated from post-abortion uterine discharge\(^4\). In a study

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conducted in Malaysia, 1688 infants with congenital abnormalities were screened for evidence of congenital Cytomegalovirus infection and it was detected in 11.4% of the infants\textsuperscript{(5)}. Cytomegalovirus specific IgM antibodies was detected by ELISA in 15.9% of the 1918 studied pregnant women. In Sudan very sparse work was conducted on cytomegalovirus\textsuperscript{(6)}, The first study of HCMV in Sudan was done in 2004 in blood donors and antenatal women\textsuperscript{(7)}. The second study was done in 2006, in candidate recipients kidney candidate donors and blood donors\textsuperscript{(8)}. The most recent study was done in 2007 in Renal transplant patients and Haemodialysis patients\textsuperscript{(9)}. This study aimed to detect Sero-frequency of CMV Among Pregnant Ladies in Khartoum Sudan.

Materials and Methods:

Study design
This is a cross sectional study included Pregnant Ladies aged between (10-50) years old who attended Omdurman Maternity hospital, Khartoum State, Sudan, during December 2014 to May 2015. The data was collected by structured questionnaire. Ethical approval was taken from Al Neelain University research ethical board and from patients verbally.

Methods:

Collection of specimens and processing:

Experimental work
Serum specimens were collected from Pregnant Ladies, and screened for CMV IgG and IgM antibodies using enzyme linked immune sorbent assay ELISA The Kits were obtained from Dignostic Bioprobes (DIA PRO) Milano-Italy, capture Enzyme Immuno( ELISA) for determination of IgM, IgG antibodies to cytomegalovirus in human plasma and. The test was performed according to the manufacturer instructions in research.
laboratory \AL Neelain University. Serum specimens was diluted (1:10), pipetted 0.1 ml from control positive and control negative and calibrator added in wells,o.1 ml diluted samples were added to each well incubated together with CMV antigen fixed in the wells of the micro plates, incubation for 1 hour at 37°C, washing 5 cycles, then o.1 ml from immunocomplex wass added to each well except blanking ,incubation for 1 hour at 37°C, washing 5 cycles , (TMB/H$_2$O$_2$ mix) was pipetted 0.1 and added to each well ,incubation for 20 min at room temperature (18-24 ºC), then Sulphric Acid was pipetted into all wells.

**Measurement**
The absorbance of specimens were measured with spectrophotometer at 450nm. The results were calculated by relating each sample optical density (OD) value to the Cut off value of plate.

Calculation of Cut off (C.O) value.
* Cut off = NC + 0.250.
*NC: Negative Control.

**Calculation and interpretation of result:-**
*Negative results*: samples giving absorbance less than Cut-off value negative for this assay.
*Positive result*: sample giving absorbance equal to or greater than Cut-off considered initially reactive.
*Borderline*: sample with absorbance to Cut-off value are considered borderline and retesting of these samples in duplicate is recommended.

**Data analysis:-**
Data was analyzed by SPSS (Statistical Package of Social Science) software program version 21.
Result

A total of 90 pregnant women were enrolled in the study. Their age ranged from 10 to 50 years, with mean 28.00 years. Most of them were in third trimester of pregnancy (54.4%), had no chronic illnesses (78.9%) and had no history of past history of abortion (80.0%).

12 (13.3%) and 25 (27.8%) were positive for Cytomegalo IgM and IgG antibodies, respectively (fig 1,2). However 8(8.8%) were positive for both (fig 3), High frequency of positive IgM and IgG results was observed among 20-30 age range group (table 1), and whom had no history of abortion, in third trimester, and were had no history of chronic illnesses (as demonstrated in tables 2,3, and 4). Statistical analysis showed that there was significant association (P value less than 0.05) between result of CMV IgG and result of CMV IgM (demonstrated in table 5). But there is insignificant correlation (P value more than 0.05) between seropositive and age (as demonstrated in table 1), history of abortion (as demonstrated in table 2), Trimesters (as demonstrated in table 3) and history of chronic illnesses (as demonstrated in table 4)

Figure 1: Frequency of antiCytomegalo virus IgM among study population (n= 90)
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Figure 2: Frequency of anti Cytomegalovirus IgG among study population (n= 90)

Figure 3: Frequency of both anti Cytomegalovirus IgM & IgG among study population (n= 90)

Table 1: Serofrequency of CMV among study population according to age group.

<table>
<thead>
<tr>
<th>Age groups in year</th>
<th>IgM seropositive</th>
<th>IgG seropositive</th>
<th>IgM – IgG seropositive</th>
<th>Negative</th>
</tr>
</thead>
<tbody>
<tr>
<td>10-20</td>
<td>1(8.3 %)</td>
<td>3 (12.0%)</td>
<td>0(0%)</td>
<td>14 (20.0%)</td>
</tr>
<tr>
<td>20-30</td>
<td>8(66.7%)</td>
<td>16 (64.0%)</td>
<td>7(87.5%)</td>
<td>36 (51.4%)</td>
</tr>
<tr>
<td>30-40</td>
<td>3(25.0%)</td>
<td>5(20.0%)</td>
<td>1(12.5%)</td>
<td>19(27.1%)</td>
</tr>
<tr>
<td>40-50</td>
<td>0(0.0%)</td>
<td>1(4.0%)</td>
<td>0(0.0%)</td>
<td>1(1.4%)</td>
</tr>
<tr>
<td>Total</td>
<td>12(100%)</td>
<td>25(100%)</td>
<td>8(100%)</td>
<td>45(100%)</td>
</tr>
<tr>
<td>p-value</td>
<td>0.66**</td>
<td>0.421**</td>
<td>0.177*</td>
<td></td>
</tr>
</tbody>
</table>

**Not significant difference at the 0.05 level.

Table 2: Serofrequency of CMV among study population according to past history of abortion.

<table>
<thead>
<tr>
<th>History of abortion</th>
<th>IgM seropositive</th>
<th>IgG seropositive</th>
<th>IgM – IgG seropositive</th>
<th>Negative</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes</td>
<td>3(25.0%)</td>
<td>6(24.0%)</td>
<td>1(12.5%)</td>
<td>11(15.7%)</td>
</tr>
<tr>
<td>No</td>
<td>9(75.0%)</td>
<td>19(76.0%)</td>
<td>7(87.3%)</td>
<td>59(84.3%)</td>
</tr>
<tr>
<td>Total</td>
<td>12(13.3%)</td>
<td>25(27.8%)</td>
<td>8(8.8%)</td>
<td>45(100%)</td>
</tr>
<tr>
<td>p-value</td>
<td>0.642**</td>
<td>0.566*</td>
<td>0.0791*</td>
<td></td>
</tr>
</tbody>
</table>

** Not significant difference at the 0.05 level
Table 3: Serofrequency of CMV among study population according to trimesters of pregnancy.

<table>
<thead>
<tr>
<th>Trimester of pregnancy</th>
<th>Result</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>IgM seropositive</td>
<td>IgG seropositive</td>
<td>IgM – IgG seropositive</td>
<td>Negative</td>
</tr>
<tr>
<td>First trimester</td>
<td>3(25.0%)</td>
<td>10(40.0)</td>
<td>1(12.5%)</td>
<td>22(31.4%)</td>
</tr>
<tr>
<td>second trimester</td>
<td>1(8.3%)</td>
<td>1(4.0%)</td>
<td>0(0.0%)</td>
<td>6(8.6%)</td>
</tr>
<tr>
<td>Third trimester</td>
<td>8(66.7%)</td>
<td>14(66.0%)</td>
<td>7(87.5%)</td>
<td>42(60.0%)</td>
</tr>
<tr>
<td>Total</td>
<td>12(100%)</td>
<td>25(100%)</td>
<td>8(100%)</td>
<td>45(100%)</td>
</tr>
<tr>
<td>p-value</td>
<td>0.638**</td>
<td>0.591**</td>
<td>0.230**</td>
<td></td>
</tr>
</tbody>
</table>

** Not significant difference at the 0.05 level.

Table 4: Serofrequency of CMV according to history of Chronic illnesses

<table>
<thead>
<tr>
<th>History of chronic illnesses</th>
<th>Result</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>IgM seropositive</td>
<td>IgG seropositive</td>
<td>IgM – IgG seropositive</td>
<td>Negative</td>
</tr>
<tr>
<td>Yes</td>
<td>5(1.7%)</td>
<td>6(24.0%)</td>
<td>3(37.5%)</td>
<td>13(18.6%)</td>
</tr>
<tr>
<td>No</td>
<td>7(7.7%)</td>
<td>19(21.2%)</td>
<td>5(62.5%)</td>
<td>57(81.4%)</td>
</tr>
<tr>
<td>Total</td>
<td>12(100.0%)</td>
<td>25(100%)</td>
<td>8(100%)</td>
<td>45(100%)</td>
</tr>
<tr>
<td>p-value</td>
<td>0.061**</td>
<td>0.677**</td>
<td>.144**</td>
<td></td>
</tr>
</tbody>
</table>

** Not significant difference at the 0.05 level.

Discussion:

The human cytomegalovirus (HCMV) is the most common viral infection worldwide and may be asymptomatic forms (90% of cases) to severe fetal damage and, in rare cases, death due to abortion.

The result of this study revealed cytomegalovirus IgM was detected in 13.3% (12 of 90) and IgG was detected in 27.8% (25 of 90) for both 8(8.8%) were positive in pregnant ladies studied in Khartoum State. This finding is a new conclusive finding, and alarming for the significant role of CMV in causing abortion and other problems. The high anti(CMV) antibody(91.1%) observed among pregnant women in MMSH Kano is due primary infection and or secondary infection with intermittent excretion of virus as result of reactivation of an endogenous virus or exposure to new virus strain from exogenous sources. Our result is lower to study among 5,959
pregnant women in Izmir Turkey \(^{(10)}\), in which 98.3% IgG seropositive was observed, also lower to study in Nigeria in which 179 pregnant in Lagos southwest Nigeria where pregnant women had (97.2%) anti(CMV) IgG antibodyies, also lower than the range of \((60 - 70\% )\) in urban U.S cities \(^{(11)}\). From the result above our study shows results lower than studies so we recommend to go in depth by using PCR.

Detailed molecular biology based study to characterize the HCMV circulating in Sudan is highly recommended.

**Acknowledgment:**

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**REFERENCES:**


