AFRICAN JOURNAL OF FETOMATERNAL MEDICINE

Original Article

Seroprevalence of Cytomegalovirus Infection Among Antenatal Women

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Abstract

Background: Primary cytomegalovirus (CMV) infection in pregnancy constitutes a major threat to the developing foetus due to trans-placental transmission which can affect the central nervous system and sensory function of the foetus. The severity of congenital damage is higher when infection occurs in the first trimester, thus CMV infection during pregnancy is a public health problem throughout the world. **Objective:** To determine the Seroprevalence of CMV infection among pregnant women attending the antenatal clinic and associated risk factors. **Materials and Methods:** This was a laboratory-based cross-sectional study. Antenatal attendees who consented to the study were recruited. Blood samples were collected and analysed using ELISA method. The data were analysed using SPSS software version 22. Chi-square test of independence was conducted to test for relationship between ELISA and the various characteristics of the cases considered. A p-value less than 0.05 was considered statistically significant at confidence interval of 95%. **Results:** The CMV seroprevalence was 30.2%. 165 participants (97.6%) were not aware of CMV infection. Higher level of education was associated with low seroprevalence (P-value 0.04). The relationship between age, parity, occupation, and marital status, stage of pregnancy, blood transfusion and CMV acquisition were not statistically significant. **Conclusion:** Seroprevalence of cytomegalovirus infection is high but awareness of the disease is low. Therefore, the need to create awareness of the disease cannot be overemphasized.

Keywords: Seroprevalence, Cytomegalovirus, Antenatal

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Introduction

Cytomegalovirus (CMV) belongs to the family herpesviridae of beta herpesvirinae subfamily[1]. It is a member of a family of 8 human herpes viruses (HHV) designated as HHV type 5. Other member of beta herpesvirinae includes HHV type 6 and 7 which share common clinical characteristics with CMV. It is a double stranded DNA virus with 162 hexagonal capsomeres surrounded by a lipid layer. It shares some common characteristics with other herpes viruses like genomic structure, virion structure and ability to cause latent and persistent infections[2].

Infections with CMV are common worldwide and primary infection may lead to lifelong latency in the host with reactivations during periods of relative immunosuppression[2,3]. The infection is of public health importance, in that it is transmitted in-utero to foetuses and could cause congenital damages to new-borns which manifest as sensorineural hearing loss, visual impairment, motor and cognitive deficits. Other transient symptoms in such new-borns may include thrombocytopenia, hepatosplenomegaly and jaundice.[1, 3]

The prevalence of congenital CMV infection ranges between 0.2 – 2.5% in different populations. The high-risk groups include those born to black and Asian women and those born prematurely. The vertical transmission of CMV is mainly due to primary infection in seronogative mothers which constitutes 30-40% of cases whereas 1-2% of cases would be due to reactivation or reinfection. This suggests that maternal preconception immunity may play a role in preventing intrauterine infections[3-5].

The routes of transmission of CMV are transplacental, contact with body fluids like saliva, urine, cervicovaginal secretions, semen, milk and blood. For pregnant women, important sources of infection are contact with urine and saliva of children, sexual activity and blood transfusion.[1, 6] CMV is also transmitted via organ transplantation and it is known to be a common opportunistic infection among HIV infected individuals.[2,6] Sexual contact is one of the most important mechanisms of transmission of CMV. This has been demonstrated between heterosexual men and women and between homosexual men. Penetrative vaginal intercourse including anal receptive intercourse is a risk for this transmission. CMV has been isolated in semen, the semen for donors for artificial insemination were randomised and the prevalence of CMV in such semen was found to be about 0.4%[8,9]. Similarly, it has been isolated from about 13-23% of women attending clinics for suspected venereal diseases[7].

It has been found that the virus does not readily spread among adults by ordinary nonsexual person to person contact even during prolonged exposure to individuals who are seropositive and known to be shedding the virus. For example, among health workers in close contact with actively infected patient, few, if any, can have seroconversions attributed to nursing functions.[7,8] Breast milk is by far the most common means of maternal to child transmission (MTCT) of CMV. By this means, majority of infants acquire CMV in the first year of life in cultures where mothers who are CMV seropositive breast feed their infants.[12] As infants acquire CMV via virolactia, other caregivers and their children in contact with such infants acquire the infection from them.

Maternal immunity does not prevent virolactia, neither does it prevent CMV disease from mothers' milk except where the new born is of low birth weight and premature.[1,6] Transmission from mother to foetus occurs in about 35% of pregnancies in which mothers have primary infection[9]. Transplacental transmission is about 20% with primary infection in the first trimester but as gestational age advances, the transmission rate increases such that in the third trimester, it is about 75%[9]. Recently, report has shown a positive correlation between post transfusion CMV infection and receipt of blood from CMV IgM positive donors[5]. Using such unit of blood to transfuse CMV seronegative women increases the incidence of CMV in the population-with the attendant consequence.[2, 5]

Serological surveys have shown increasing incidence of CMV infections in various populations worldwide and for the fact that during pregnancy in seropositive mothers the foetus is at risk, therefore, there is need to determine the seroprevalence of CMV antibodies in pregnant women. This will underscore the importance of CMV screening in blood units for use in pregnancy and labour. This is very necessary to avoid the transmission of CMV infected blood to women who are CMV seronegative.[2, 5]

CMV is the most common viral cause of congenital infection affecting 0.2-2.2% of live births and responsible for significant morbidity especially in infants who are symptomatic in the neonatal period.[3] It remains the leading non genetic cause of sensorineural hearing loss and a major cause of neurological disability.[10, 11] The severities of infection in neonates and infants are highly variable. About 10-15% of congenitally infected neonates are symptomatic at birth; 40-58% of them will experience permanent long-term sequelae.[12, 13] Moreover 13.5% of children with asymptomatic infection will develop late onset sequelae mainly consisting of hearing impairment and neurologic deficit.[11, 13] Foetal or neonatal death occurs in 10% of foetuses following intrauterine CMV infections[12].

The severity of congenital infection is higher if it occurs in the first trimester than in the third trimester. In pregnant women, the infection is often asymptomatic so it may go clinically unnoticed. When symptoms are present, they are nonspecific and resemble a flu like mononucleosis with fever, cervical lymphadenopathy, sore throat and myalgia.[14] For infants when symptoms occur at birth, they include rashes and feeding difficulties.[14]

Cytomegalovirus infection has been studied among pregnant women in different locations within Nigeria. A study in Murtala Mohamed Specialist Hospital, Kano showed a seroprevalence of 91.1%, while a multicentre study in Makurdi, Benue state showed a 93.3% seroprevalence whereas a study in Kaduna Nigeria showed a seroprevalence of 94.8%.[2,10,14]

There was a case reported in Osun state, Nigeria in 2004 where three siblings in a monogamous family presented with a history of visual impairment/blindness and it was confirmed to be due to CMV.[2]

CMV is a major public health concern due to its ability to cause congenital damage to foetuses. Therefore, this study was aimed at determining the seroprevalence of CMV in pregnant women so as to form basis or justification for routine screening in our facility and Nigeria at large.

Methodology

This was a cross sectional study of pregnant women that attended antenatal clinic in University of Abuja Teaching Hospital, Abuja who gave consent during the study period from August 2022 to October 2023. An informed consent was taken, each participant completed or was assisted to complete a structured questionnaire on demographic characteristics, environmental, personal and behavioural factors associated with CMV infection.

About 5ml of venous blood was collected from

each patient using a 5ml syringe and a 23-gauge needles. The blood sample collected was transferred into a test tube bearing patient's identification number. Centrifuging of blood samples was done at 4000 revolutions per minutes (rpm) for 5 minutes.

A Pasteur pipette was used to transfer serum into clean cryovials which were stored at - 23°C until the predetermined number of samples was obtained.

Qualitative determination of CMV antibodies was carried out using Enzyme Linked Immunosorbent Assay (ELISA) technique with the use of CMV IgG kit (Dialab® Australia). Samples' reagents and calibrators were brought to room temperature before the test. Microwell plates were labelled for sample, control and blank.

Dilutions of samples were done at ratio 1:100 with samples' diluents. Afterwards, 100µl of each sample positive and negative control were dispensed into wells, leaving a well empty (the blank). The plate was covered with adhesive foil (VWR®, USA) and incubated at 37°C for 60 minutes using an incubator (NAPCO, Thermo Electron Corporation). Later the adhesive foil was detached gently to get the plate washed 7 times with 300µl diluted wash buffer 9 Biotek® plate washer. 100µl of enzyme conjugate was then introduced into the wells, covered again and incubated at body temperature (37°C) for another 60 minutes, to be washed again 7 times with 300µl of diluted buffer. Then 100µl of the substrate was dispensed into all the wells including the blank and each plate was covered with a new adhesive foil and this time it was incubated at room temperature for 20 minutes, protected from light after which 100µl of solution was introduced into all the wells.

The absorbance of each sample, control and blank was read off using soft max pro 5.4 software with molecular devices ®plate reader at wave length of 450nm. The cut off of IgG was set at 0.5 (WHO IU/ml). Only samples with concentrations >0.5 WHO IU/ml were considered positive for CMV IgG while samples with concentrations below the cut off were considered as negative results.

CMV IgG ELISA KIT is intended for detection of IgG antibody to CMV in human sera or plasma. Diagnostic sensitivity for IgG test kits has been calculated to be more than 98% on sera and plasma. There has not been any known cross reactivity.

Diluted patient's serum in each case was added to wells coated with purified antigen. IgG specific antibody, if present, binds to the antigen. All unbound materials were washed away and the enzyme conjugate was added to bind to the antibody-antigen complex. If present, excess enzyme conjugate was washed away and substrate was added. The plate was incubated to allow the hydrolysis of the substrate by the enzyme. The colour generated was proportional to the amount of IgG specific antibody present in the sample.

The kit and the reagents were regarded as bio hazardous materials and were handled with safety precautions by using surgical gloves and laboratory clothing. The reagents were handled at the biosafety level 2 as recommended by centre for disease control (CDC). All test protocols were strictly adhered to. The samples were stored at 2-8°C. All samples were equilibrated to room temperature (23-25°C) before use.

This was guided by the calibration factor (CF) on the calibrator bottle. Cut off value: calibrator optical density (OD) x calibrator factor. The validation of CMV ELISA kit was according to manufacturer's standard.

Statistical analysis of results was done using the statistical package for social science (SPSS Version 22). Chi-square test of independence was conducted to test for relationship between ELISA and the various characteristics of the cases considered. For chi-square test to be valid, at least 20% of observations must have expected count of ≥ 5. Where this was not the case, the Fisher's exact test was used for the analytic assessment and the differences were considered to be statistically significant if P-value obtained was ≤ 0.05.

Approval for the conduct of the study was sought and subsequently granted from the hospital's health research and ethics committee. Client's anonymity was maintained. Only code numbers were used to identify clients. No client was denied any form of services or promised facilitation of services based on decision to decline or give consent.

Statistical analysis of results was done using the statistical package for social science (SPSS Version 22). Chi-square test of independence was conducted to test for relationship between ELISA and the various characteristics of the cases considered. For chi-square test to be valid, at least 20% of observations must have expected count of ≥ 5.

Where this was not the case, the Fisher's exact test was used for the analytic assessment and the differences were considered to be statistically significant if P-value obtained was ≤ 0.05.

Results

One hundred and sixty-nine (169) pregnant women were enrolled and completed the study. Of these, 51 were seropositive for anti-CMV antibodies giving a seroprevalence of 30.2%. The participants were between the ages of 18 – 42 years with a mean age of 30.54 ± 4.72 years. The women between the ages of 30 – 39 years were 87 (51.5%) and formed the highest proportion of participants. The result shows that age as a risk factor was not statistically significant with a P-value of 0.99 (Table 1)

With respect to parity, seropositivity increased from 13.6% for the primigravidae to 14.2% for para 1-4. The relationship between parity and CMV-seropositivity was not statistically significant (P = 0.98) (Table 1).

For the relationship between education and CMV seropositivity, the women with tertiary level of education formed the highest proportion of the participants making up 63.9%. Only 16% of them demonstrated anti – CMV antibodies and with a P-value of 0.04, the relationship between education and CMV infection was found to be statistically significant (Table 1).

Table 1: Socio-Demographic Data of the Participants

Variables	Seropositive F (%)	Seronegative F (%)	P-value
Age (years)			
< 19	1 (0.6)	2 (1.3)	
20 – 29	21 (12.4)	54 (32.0)	0.99
30 – 39	28 (16.6)	59(34.9)	
≥ 40	1 (0.6)	3 (1.8)	
Parity			
0	23 (13.6)	55 (32.5)	0.98
1 – 4	24 (14.2)	54 (32.0)	
> 4	4 (2.4)	9 (5.3)	
Education			
None	0 (0)	3 (1.8)	
Primary	4 (2.4)	2 (1.2)	0.04
Secondary	20 (11.8)	32 (18.9)	
Tertiary	27 (16.0)	81 (47.9)	
Occupation			
Civil servant	10 (5.9)	29 (17.2)	
Trader	6 (3.6)	13 (7.7)	0.49
Self employed	21 (12.4)	41 (24.3)	
Unemployed	8 (4.7)	28 (16.6)	
Student	6 (3.6)	7 (4.1)	
Marital Status			
Single	2 (1.2)	1 (0.6)	0.45
Married	49 (29.0)	116 (68.6)	
Separated	0 (0.0)	1 (0.6)	
Trimester			
1st	7 (4.1)	17 (10.1)	0.75
2nd	19 (11.2)	37 (21.9)	
3rd	25 (14.8)	64 (37.9)	

Table 2: Relationship Between Blood Transfusion and Anti-CMV IgG Seropositivity

PTH	Sero-positive (%)	Sero-negative (%)	Chi-square	P-value	95% C I
Yes	4 (2.4%)	13 (7.7%)	0.396	0.53	- 0.181 - 0.651
No	47 (27.8%)	105 (62.1%)			0.177 - 0.441

Self-employed women formed the highest proportion, 62 (36.7%); and of whom 12.4% were seropositive. The relationship between occupation and CMV seropositivity was not statistically significant (P = 0.49) (Table 1).

97.6% of the participants were married. Of the married women, 29.0% were seropositive and 1.2% of the single women were also seropositive. The statistical analysis shows that the relationship between marital status and CMV seropositivity was not statistically significant (P = 0.45) (Table 1).

In this study, 4.1% of the women were seropositive in the first trimester. The relationship between stage of pregnancy and CMV seropositivity is not statistically significant (P = 0.75) (Table 1).

Analysis based on past history of blood transfusion showed that 10.7% of the participants had positive history of past blood transfusion. Of those with positive history, 2.4% were seropositive. With a P-value of 0.53, there was no statistically significant relationship between blood transfusion and CMV – seropositivity in this study (Table 2).

None of the participants had previous history of still birth/miscarriages and none worked in day-care centres. CMV status of partners was not known and all the participants did not answer in the affirmative to the role of diapers and baby pacifiers.

Discussion

In this study, 169 pregnant women were enrolled and completed the study. Of the study population of 169 participants, 51 of them demonstrated anti-CMV antibodies. This gave a seroprevalence of 30.2% which is very significant. CMV infection is present worldwide and in pregnant women, the infection usually appears with mild clinical symptoms and signs, which makes it difficult to recognise, so if the serology testing is not done as a common analysis in pregnant women, the infection can easily be overlooked and lead to infection of the foetus, thereby causing congenital disease.[1, 2,3]

As far back as 1973, it was discovered that CMV antibodies were more prevalent in the developing countries due to low socioeconomic status. In the developed world, the seroprevalence is lower.[5] Among the French women, the seroprevalence was found to be 51.5%, whereas in Italy, Ireland, USA and Australia, the seroprevalences were 63.8%, 30.4%, 39.94%, and 56.8% respectively. [2, 5, 10, 14] In Kano, Kaduna, Makurdi and Lagos Nigeria, the reported seroprevalences were 91.1%, 94.8%, 93.3% and 97.2% respectively. [2, 5, 10, 14]

In this study, 169 pregnant women participated with a mean age of 30.54 ± 4.72 years and majority of them were within 20 – 29 and 30 – 39 years. This finding is similar to a study by Porobic-Jahic et al in which the mean age was 28 ± 4.97 years.[61] In this study, there was 30.2% seropositivity to CMV infection, this was significant when compared with the results from other parts of Nigeria but similar to the seropositivity rate in Ireland (30.4%).[5] Analysis of results by age showed no significant association between age and CMV acquisition. This is contrary to reports by Okwori et al and Sadler et al, who also used ELISA method but found a significant association between maternal age and CMV acquisition.[2, 5, 59, 60] In this study, the higher seroprevalence was found among the younger age group, these could be explained by increase in sexual activities in the younger age group since the virus can be sexually transmitted.[2, 7] In the same vein, younger maternal age may be associated with poor hygiene and increased risk of the CMV infection. [54, 55]

The Kenyan and Sudanese studies reported a significant association between higher parity and risk of CMV seropositivity with the explanation that the women with higher parity are more likely to have more contact with young children with a higher tendency to viral shedding. [56, 57]

A Tanzanian study reported a conflicting finding, indicating a significant association between low parity and acquisition of CMV with the explanation that

women with lower parity would be younger and less experienced, and therefore with poorer hygiene habits thus increasing their chances of CMV acquisition.[65] Okwori et al, noted there was no significant association between parity and CMV acquisition. [59]

We observed that mothers with more than 4 deliveries had a lower tendency for CMV seropositivity which is in conflict with the Kenyan and Sudanese studies and also not in tandem with the Tanzanian studies.[56, 57, 65] Therefore the role of parity in CMV acquisition remains contentious making it necessary for further research to be undertaken to determine the exact role parity plays in the maternal acquisition of CMV.

Some authors have suggested that such research could use CMV DNA and IgM seropositivity instead of the popular IgG seropositivity. In this study, a significantly higher number of IgG anti CMV antibodies among women with lower level of education and with a growth in the percentage of negative CMV antibodies as level of education increases ($p = 0.04$). This finding is similar to what was found in a study in Maiduguri where it was shown that the degree of seronegativity increases with level of education. [50]

In the study by Mamuye et al, there was no significant correlation between seropositivity to CMV and education, however, the influence of education can be explained by the direct relationship between education and income which is the main determinant of living conditions.[51, 52, 53] Those women with a higher level of education could afford better and healthy living conditions with an expected decrease in the exposure to the virus.[2]

In this study, with respect to occupation, seroprevalence of CMV infection by employment shows that seronegativity is higher among the self-employed (24.3%) and this increases according to occupational status. Similar findings were noted by Yero et al. Thus, it was explained that high socioeconomic status implies the ability to acquire education and therefore improved living conditions which decreases exposure to the virus. [2]

Concerning marital status of the participants, the study shows that 97.6% of the women were married. This shows almost identical data in a study conducted by Mamuye et al were 98.5% of the participants were married. This finding is also similar to study by Porobic-Jahic et al, in which 98.33% of the participants were married.[51, 61] It goes to show that a vast majority of pregnant women that book for antenatal care is married. The single mothers, may be because of stigma, are less likely to book for antenatal care and therefore unlikely to test for CMV [61].

With respect to stage of pregnancy, this study shows 4.7% of the participants were seropositive in the first trimester. High seropositivities, 11.2% and 14.8%, were seen in 2nd and third trimesters respectively. This finding is corroborated by Hamid et al, where high seropositivity was found in the second and third trimesters.[10] and also similar to the findings by Gehrz et al.[58] The explanation is that, in the 2nd and 3rd trimesters, there is marked transient depression of CMV specific cellular immunity with corresponding increased susceptibility to infection. [10, 58]

Transmission of CMV can occur through transfusion of CMV infected blood. [61, 63]

In our environment currently, there is no routine screening of blood or blood products for CMV before transfusion is carried out; it is therefore possible that some of the participants with previous history of blood transfusion could have acquired CMV through blood transfusion. When CMV is acquired through blood transfusion it would be reactivated in settings of compromised immunity and pregnancy, thus increasing the risk of vertical transmission.

In this study, past history of blood transfusion has been shown not to be a significant factor ($P=0.53$) in CMV acquisition, however given the discrepancies in the seroprevalence as noted above, it may be inferred that pregnant women may be receiving CMV contaminated blood through transfusion. [6]

Conclusion

In our centre currently only, few people are aware of CMV disease and the dangerous consequences of the infection to the newborn baby. It is therefore necessary to create awareness among care givers as well as the public about the infection in pregnancy. When such awareness is created, control measures can be put in place by agencies of government as well as non-governmental organisations with the aim of preventing the infection. As the demographics of the population in Abuja evolve, the CMV seroprevalence in pregnancy may change and could lead to increased incidence of congenital CMV infection.

Recommendation: Intervention measures such as minimizing close contacts with possible sources of infection like careful handling of diapers, baby pacifiers, through hand-washing when handling young children should be emphasized during antenatal talk. Offering screening for CMV IgG antibodies in pregnancy, educating women about CMV and providing simple hygienic precautions should be a routine. Detailed anomaly scan in search of features of congenital CMV infection should be emphasized. Screening of blood for CMV prior to transfusion should be adopted in our health institutions. Placentas of seropositive mothers should be examined following delivery for evidence of CMV infection and babies born to such mothers should also be screened with necessary interventions for such babies put in place.

References

1. Sana AA. Cytomegalovirus incidence in women with recurrent abortion. J Bioanul Biomed., 2012; 4:6.101-103.
2. Yero M, Aminu M, Musa BOP. Seroprevalence of cytomegalovirus infection amongst pregnant women in Kaduna state, Nigeria. Afr J Clin Exper Microbio., 2015; 16(1): 37 – 44.

3. Kalil A, Heath P. Soe A, Ville YG. Congenital cytomegalovirus infection. Update on treatment. BJOG, 2018;125:1 – 11
4. Dollad SC, Girsse SD, Ross DS. New estimates of the prevalence of neurological and sensory sequaele associated with congenital cytomegalovirus infection. Rev Med Virol., 2007; 17(5); 355 – 363.
5. Akinsegun AA, Kabiru AR, Adeniyi AA, Kikelomo OW Adedoyin OD, Titilope AA, et al. Seroprevalence of cytomegalovirus in Nigeria, Int J Womens Health, 2011; 3: 423 – 428
6. Pass RF Arav-Boger R. Maternal and foetal cytomegalovirus infection: diagnosis, management and prevention, 2018; 7: 255
7. Long DJ, Rummer JF, Hartly DP. Cytomegalovirus in semen: persistence and demonstration in intercellular fluids. N Eng J Med., 1974; 291; 121 – 123.
8. Robert FP. Brenna A. Mother-to-child transmission of cytomegalovirus and prevention of congenital infection. J Pediatric Infects Dis Soc., 2014; 1 2-6.
9. Lawrence DW, Becker Y. Cytomegalovirus as a sexually transmitted disease In: Molecular Aspects of Human Cytomegalovirus Disease, 1993; 92-93
10. Stuart P A. Screening for cytomegalovirus during pregnancy. Infectious diseases in: Obstetrics and Gynaecology, 2011; 1-9
11. Hamid KM, Onoja AB, Garba KN. Seroprevalence of cytomegalovirus among pregnant women attending Murtala Mohamed Specialist Hospital, Kano, Nigeria. Afr Health Sci., 2014; 14(1): 125 – 130.
12. Nigro G, Adlaer SP, La Tirre R, Best AM. Congenital cytomegalovirus collaborating Group. Passive Immunization during pregnancy for congenital infection N Eng J Med., 2005; 353; 1350 – 1362.
13. Saldan A, Forner G, Mengoli C, Gusseti N, Palu G, Abate D. Testing for cytomegalovirus in pregnancy. J ClinMicrobiol., 2017; 55:693-702
14. Umeh EU, Onoja TO, Aguoru CU, Umeh JC. Seroprevalence of cytomegalovirus antibodies in pregnant women, Benue State, Nigeria. J Infect Dis Ther., 2015; 3:242.
15. Porobic-Jahic H, Skokic F, Ahmetagic S, Petrovic J. Cytomegalovirus Infection in pregnancy-our experiences. MED ARCH, 2019; 73 (3); 149-53.
16. Okwori A, Olabode A, Emumwen E, et al. Sero-epedemiological Survey of Cytomegalovirus infection among expectant Mothers in Bida, Nigeria. The Internet J. of Infect. Dis., 2008; Vol.6 Number 2.
17. Stadler LP, Bernstein DI, Callaham ST, et al. Seroprevalence of Cytomegalovirus (CMV) and Risk Factors for Infection in Adolescent Males. Oxf. J. of Clin. Infect. Dis., 2012; 51 (10): 76-81.
18. Fowler KB, Stagno S, Pass RF. Maternal age and congenital cytomegalovirus infection: screening of two diverse newborn populations. 1980-1990. J Infect Dis., 1993; 168: 552-556.
19. Sherrif A, Golding J, Team TAS Factors associated with different hygiene practices in the homes of 15 month old infants. Arch Dis Child, 2002; 87: 30-35.
20. Maingi Z, Nyamache AK Seroprevalence of Cytomegalo Virus (CMV) among pregnant women in Thika, Kenya. BMC Res Notes, 2014; 7: 794.
21. Hamdan HZ, Abdelbagi IE, Nasser NM, Adam I Seroprevalence of Cytomegalovirus and rubella among pregnant women in western Sudan. Virol J., 2011; 8: 217.
22. Chibwe E, Mirambo MM, Kihunrwa A, Mshana SE Magnitude of the Cytomegalovirus infection among pregnant women attending antenatal clinic in the city of Mwanza Tanzania. BMC Res Notes, 2017; 10:489.
23. Nasir IA, Usman Z, Babayo A. Evaluation of Pregnant Women Susceptible to cytomegalovirus infection in Maiduguri, Nigeria Research Journal of Microbiology, 2015;10(7): 336-342.
24. Mamuye Z, Nigatu B, Bekele D, Challa F, Desale A and Solomon S. Seroprevalence and Absence of Cytomegalovirus infection Risk Factors among Pregnant Women in St. Paul's Hospital Millennium Medical College. Gynecol Obstet., 2015; 5: 299.
25. Hoes J, Boef AGC, Knol MJ, de Melker HE, Mollema L, van der Klis FRM, Rots NY, van Baarle D Socioeconomic status is associated with antibody levels against vaccine preventable diseases in the Netherlands. Front Public Health, 2018; 6: 209.
26. Meier HCS, Haan MN, Mendes de Leon CF, Simanek AM, Dowd JB, Aiello AE Early life socioeconomic position and immune response to persistent infection among elderly Latinos. Soc Sci. Med., 2016; 166:77-85.
27. Gehrz RC, Christanson WR, Linner KM, Conroy MM, McCue SA, Balfour HH. Cytomegalovirus specific humoral and cellular immune response in human pregnancy. J Infect Dis., 1981:143:391-395.
28. Akende O, Akanbi OA, Oluremi AS, OKonko IO, Opaleye OO Prevalence of cytomegalovirus lgG antibodies among pregnant women antenatal clinic, LAUTECH State, Nigeria. J Immunoass Say Immunochem., 2006; 37: 289-2