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Prevalence of Human Cytomegalovirus IgM antibodies in Pregnant Women in Port Harcourt, Rivers State, Nigeria

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ABSTRACT: Notwithstanding, human cytomegalovirus (HCMV) infection is linked to a possible danger to the fetus, there is scanty data from Rivers State, Nigeria concerning the epidemiology and the impact of HCMV infections. This cross-sectional study was conducted between February 2019 and November 2019 among pregnant women attending an antenatal clinic at the University of Port Harcourt Teaching Hospital (UPTH), Port Harcourt, Nigeria, to investigate the magnitude and associated factors of HCMV infection. The specific HCMV IgM antibody was detected using an enzyme-linked immunosorbent assay (ELISA). A total of 90 pregnant women in the age range of 20-49 years were enrolled. The prevalence of HCMV IgM antibodies was 25.6%. Only occupation, family type, history of STDs, HIV status, and being married ($p < 0.05$) occurred to predict HCMV IgM seropositivity independently. Concerning the outcomes of preceding pregnancies, history of abortion/miscarriage independently predicted IgM seropositivity ($p=0.021$). Human cytomegalovirus (HCMV) prevalence amongst pregnant women in Port Harcourt, Nigeria is high. This is linked to deprived pregnancy outcomes. Infection with human cytomegalovirus (HCMV) during pregnancy may lead to congenital disease. Thus, emphasize the screening of HCMV routinely to establish the impact of HCMV during pregnancy.
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1. INTRODUCTION

Human cytomegalovirus (CMV) is a double-stranded DNA virus member of the Herpesviridae family (Zamora, 2011; Drebber et al., 2011; Alvarado-Esquivel et al., 2018). Primary infections with HCMV occurred worldwide (Davis et al., 2017), leading to lifelong latency with reactivation during relative immunosuppression in the host (Zamora, 2011; Drebber et al., 2011; Alvarado-Esquivel et al., 2018).

Principal sources of CMV infection during pregnancy are young children and intimate contacts (Johnson et al., 2012; Davis et al., 2017). Primary infection with CMV occurs in approximately 1-4% of pregnancies (Davis et al., 2017; Alvarado-Esquivel et al., 2018). Congenital HCMV transmission rates were as high as 50% in women who got infected during pregnancy and <2% in women with non-primary infection (Davis et al., 2017; Alvarado-Esquivel et al., 2018). Intrauterine HCMV transmission rates are about 30% and 0.2% for primary and non-primary infections, respectively (Saldan et al., 2017). HCMV can be spread from mother to child, during breastfeeding or intrapartum (Davis et al., 2017; Alvarado-Esquivel et al., 2018).

The seroprevalences of HCMV in pregnant women are high (Mocarski et al., 2007); with the highest proportions being reported in Qatar, Saudi Arabia, Turkey, Taiwan and sub-Saharan Africa (Abu-Madi et al., 2010; Mashuda et al., 2014; Yamamoto et al., 2013; Chibwe et al., 2017). In Rivers State, Nigeria, there is scanty information on the magnitude of HCMV amongst pregnant women, despite having high prevalences of congenital disabilities with unknown origins (Mashuda et al., 2014; Chibwe et al., 2017

Notwithstanding, human cytomegalovirus (HCMV) infection is linked to a possible danger to the fetus, there is scanty data from Rivers State, Nigeria concerning the epidemiology and the impact of HCMV infections. The study sought to determine the seroprevalence and correlates of HCMV infection amid pregnant women in Port Harcourt, Nigeria.

MATERIALS AND METHODS

2.1. Study Area

The study was carried out with pregnant women at the University of Port Harcourt Teaching Hospital (UPTH), Rivers State, Nigeria. UPTH is located at Alakahia, East-West Road of Port Harcourt, Rivers State, Nigeria. Port Harcourt lies 4.78^0N and 7.01^0E. The city is characterised by a moderate level of sanitation, improper waste management, moderate housing and potable water.

2.2 Ethical aspects

The Hospital Ethical Committees of UPTH and University of Port Harcourt approved this study. The purpose and procedures of this study were explained to pregnant women. Participation in this study was voluntary, and informed consents were gotten from all participants.

2.3 Study design and women studied

This cross-sectional study was evaluated from February 2019 to November 2019. Participants were enrolled consecutively in the study when attending their antenatal care consultations at UPTH. Inclusion criteria for enrollment in the study were: 1) pregnant women at their 1 - 9 months of pregnancy, 2) 20 - 49 years old, and 3) voluntarily accepted to participate. Pregnant women were invited to participate in the study regardless of their socio-economic status, occupation, or educational level. In total, 90 pregnant women (average age: 25 years; range 20 - 49 years) were included in the study.

2.4 Study Population

Ninety pregnant women of different ages and socio-economic statuses at UPTH were enrolled in this study. This study commenced by recruiting consecutive consenting pregnant women until 90 blood samples were obtained. Relevant information on all pregnant women was gotten from the hospital records. Table 1 summarises the characteristics of pregnant women in Port Harcourt used for this study.

2.5 Socio-demographic, clinical, behavioural and family characteristics of pregnant women

The pregnant's socio-demographic, clinical, behavioural, and family characteristics were obtained with a standardised questionnaire in face-to-face interviews.

2.6 Sample Collection and preparation/processing

The method of sample collection employed was the venipuncture technique (Okocha et al., 2005). The samples of blood were collected into an EDTA bottle. These samples were elated to the laboratory in a cold chain for further analysis and processed using standard laboratory procedures.

2.7 Detection of anti-CMV IgM antibodies

ELISA Kit (Cytomegalovirus IgM) manufactured by DIA.PRO Diagnostic Bioprobes Srl via Columella $n^0$31 20128 Milano- Italy was used to screen for CMV specific-IgM antibodies following manufacturer's instructions. A blood sample from each participant was collected. After centrifugation of blood samples, plasma samples were obtained and kept frozen until analysed. Plasmas were tested for anti-CMV IgM antibodies by the commercially available enzyme-linked immunosorbent assay (ELISA) "CMV IgM" (Dia. Pro, Milano, Italy). Results were interpreted as described by the manufacturer.

2.8. Data Analysis

Data were analysed using Microsoft Excel 2016 version to calculate the results. The statistical analysis was performed with the aid of SPSS version 16.0. Pearson's Chi-square test (when cell values were < 5) was used to compare the frequencies among groups. Bivariate analyses assessed the association of pregnant women characteristics and CMV seropositivity. P value < 0.05 was taken as statistically significant.

3. RESULTS

3.1 Characteristics of Study Population

The total number of pregnant women included was 90. The socio-demographic data for these samples were stratified and shown in Table 1.

3.2 Prevalence of HCMV IgM antibodies

Of the 90 specimens tested, only 23 (25.6%) were seropositive for HCMV IgM antibodies, while 67 (74.4%) tested seronegative. Individual values of anti-HCMV IgM antibodies obtained in the 90 pregnant women studied are shown in Table 1. The General socio-demographic characteristics of the pregnant women surveyed are shown in Table 1. By bivariate analysis, none of the socio-demographic characteristics of pregnant women was associated (P > 0.05) with seropositivity to HCMV IgM antibodies.

3.3 Prevalence of HCMV IgM antibodies concerning age

The prevalence of HCMV IgM antibodies concerning age is shown in Table 1. Concerning age, all three age groups were reactive. A higher prevalence of HCMV IgM antibodies occurred in the 20-29 years and 40-49 years (33.3%) than in the age group 30-39 years (21.7%). There was, however, no significant relationship between the age groups and HCMV IgM (χ^2= 2.169, df = 2, p>0.05) (Table 1).

3.4 Prevalence of HCMV IgM antibodies concerning marital status

Table 1 shows the seropositivity rate of HCMV IgM antibodies according to marital status. None of the single pregnant women was seropositive (0.0%) for HCMV IgM, while 23 (25.8%) married women tested seropositive. Statistically, marital status was not significantly associated with HCMV IgM (χ^2= 0.017, df = 1, p>0.05).

Table 1: The Prevalence of HCMV IgM antibodies concerning the socio-demographic characteristics of the pregnant women

Socio-Demographic Characteristics	Groups	No. Tested (%)	No. Positive for CMV IgM
Age groups	20-29	24(26.7)	8(33.3)
	30-39	60(66.7)	13(21.7)
	40-49	6(6.7)	2(33.3)
Marital Status	Married	89(98.9)	23(25.8)
	Single	1(1.1)	0(0.0)
Educational Status	None	1(1.1)	0(0.0)
	Secondary	12(13.3)	3(25.0)
	Tertiary	77(85.6)	20(26.0)
Occupational Status	Student	9(10.0)	3(33.3)
	Unemployed	12(13.3)	4(33.3)
	Civil servants	29(32.2)	5(17.2)
	Trading	31(34.4)	8(25.0)
	Artisans	4(4.4)	0(0.0)
	Business Executive	5(5.6)	3(60.0)
Religion	Christianity	81(90.0)	21(25.9)
	Islam	0(0.0)	0(0.0)
	None	9(10.0)	2(22.2)
Family Type	Monogamous	75(83.3)	17(22.7)
	Polygamous	15(16.7)	6(40.0)
Gestation Period	1st Trimester	8(8.9)	3(37.5)
	2nd Trimester	44(48.9)	10(22.7)
	3rd Trimester	38(42.2)	10(26.3)
Parity	0	34(37.8)	11(32.4)
	1-2	38(42.2)	10(26.3)
	3-4	18(20.0)	2(11.1)
History of Abortion	Yes	30(33.3)	7(23.3)
	No	60(66.7)	16(26.6)
History of STDs	Yes	6(6.7)	2(33.3)
	No	84(93.3)	21(25.0)
HIV Status	Seropositive	3(3.3)	0(0.0)
	Seronegative	87(96.7)	23(26.4)
Total		**90(100.0)**	**23(25.6)**

3.5 Prevalence of HCMV IgM antibodies concerning educational background

The educational background had no significant relationship with HCMV IgM (χ^2= 0.787, df = 2, p>0.05). Zero prevalence occurred with no formal education (0.0%). Tertiary and secondary education had 26.0% and 25.0% seroprevalence for CMV IgM, respectively (Table 1).

3.6 Prevalence of HCMV IgM antibodies concerning the occupational status

Table 1 shows the seropositivity rate of HCMV IgM antibodies according to occupation. The pregnant women that were unemployed and students (33.3%), artisans (0.0%), business executive (60.0%), civil servants (17.2%) and traders (25.0%) were all seropositive for hCMV IgM antibodies. Statistically, no significant relationship between occupation and prevalence of HCMV IgM antibodies (χ^2= 2.846, df = 5, p>0.05).

3.7 Prevalence of HCMV IgM antibodies concerning religion

A higher prevalence of HCMV IgM antibodies occurred among Christians (26.0%) than no religion (22.2%), while zero prevalence (0.0%) was recorded for Islam, as highlighted in Table 1. However, no significant association was found between religion and the prevalence of HCMV IgM (χ^2= 5.297, df = 1, p>0.05).

3.8 Prevalence of HCMV IgM antibodies concerning the family type

Polygamous family type (40.0%) had a higher prevalence of HCMV IgM antibodies than the monogamous family type (22.7%), as shown in Table 1. However, no significant association was found between the family type and HCMV IgM antibodies (χ^2= 0.941, df = 1, p>0.05).

3.9 Prevalence of HCMV IgM antibodies concerning gestation period

A higher prevalence (37.5%) occurred in first trimester than the second (22.7%) and third trimester (26.3%), as shown in Table 1. However, no significant association was found between the gestation period and the HCMV IgM antibodies (χ^2= 0.941, df = 2, p>0.05).

3.10 Prevalence of HCMV IgM antibodies concerning parity

Nulliparous pregnant women had the highest prevalence (32.4%). Those with 1-2 and 3-4 parity had a prevalence rate of 26.3% and 11.1%, respectively, as shown in Table 1. However, no significant association was found between the parity and HCMV IgM antibodies (χ^2= 0.941, df = 2, p>0.05).

3.11 Prevalence of HCMV IgM antibodies concerning the history of abortion

Higher prevalence of HCMV occurred with no history of abortion (26.6%) compared to those with such a history (23.3%) (Table 1). However, no significant association was found between having abortion history and HCMV IgM (χ^2= 0.941, df = 1, p>0.05).

3.12 Prevalence of HCMV IgM antibodies concerning the history of STDs

Higher prevalence of HCMV occurred with history of STDs (33.3%) compared to no history (25.0%), as shown in Table 1. However, no significant association was found between the history of STDs and HCMV IgM prevalence (χ^2= 0.941, df = 1, p>0.05).

3.13 Prevalence of HCMV IgM antibodies concerning HIV Status

Women with HIV seronegative status had the highest prevalence (26.4%) compared to those with HIV seropositive status (0.0%), as shown in Table 1.

4. DISCUSSION

Human cytomegalovirus (HCMV) remains the leading cause of congenital viral infection. Very little is known about the seroepidemiology of HCMV infection in pregnant women in Rivers State, Nigeria.

In this study among pregnant women, relationship between age, marital status, occupation, educational status, family type, religion, gestation, parity, history of abortions, STDs and HIV status, in HCMV prevalence were evaluated.

The study found a 25.6% seroprevalence of HCMV IgM antibodies in pregnant women. This 25.6% is higher than the 0.4% reported by Chibwe et al. (2017) in Tanzania; the 8.1% reported in Kenya (Maingi & Nyamache, 2014); the 3.5% reported in Makurdi, Benue State, Nigeria (Umeh et al., 2015). It is also higher than the 3.0% reported for HCMV IgM recurrent infection and 2.0% reported for primary HCMV IgM infection in Benin, Nigeria (Ogbaini-Emovon et al., 2015). It is higher than the prevalence rate of 1–4% reported in Iran before 2007 (Ziapour et al., 2016); the 10.5% among women in Mashad (Iran). All these studies had values far lower than the 25.6% reported here. In Ghana, none had HCMV IgM, similar to India. There are different reports from developed countries. For example, in Canada, the rate was 0.9% (Ziapour et al., 2016).

The 25.6% reported here is inconsistent with reports from other regions of the world, including Nigeria (Akinbami et al., 2011; Umeh et al., 2015). A low anti-HCMV IgM antibody (indicating recent HCMV infection) in pregnant women was reported, and low anti-CMV antibodies deviated from this study. Primary CMV infection is critical because primary HCMV infection is a significant risk factor for vertical transmission of HCMV infection to newborn babies with its entire associated clinical conditions (Leila et al., 2012; Umeh et al., 2015).

The 25.6% reported here is lower than the 31.67% reported by Zalei et al. (2017) in Iran, the 33.8% in Kerman study in Iran reported by Zalei et al. (2017); and the 60.0% reported in Bangladesh (Jahan et al., 2017). The rate of HCMV IgM is also higher in other evolving countries, dissimilar to the present study. There are different reports from developed countries. In Spain, the rate was 58.4% in men and 66.7% in women aged 2–60 years (Zalei et al., 2017).

Based on our results of HCMV IgM antibodies, all seropositive women had latent infections. As portrayed in this study, age was not significantly associated with HCMV infection. However, the increase in seroprevalence with age could be credited to the weakening of the immune system as age increases, as suggested by Redwan et al. (2001). This study showed that all three age groups were reactive. However, a higher prevalence of HCMV IgM occurred in the 20-29 years and 40-49 years (33.3%) than in the age group 30-39 years (21.7%). This observation is dissimilar to previous findings. Safabakhsh et al. (2013) reported highest rate in age range 30–39, and the lowest rate was seen in those younger than 20 years. The infection rate did not follow any particular pattern with age, which was not detected in several other studies. Other studies reported that rates of CMV IgM increase with age, and the lowest rate is seen in those younger than 20 years.

In the current study, highest rate of infection was seen in the age range of 20-29 and 40-49 years, which is slightly similar to Zalei et al. (2017), who reported highest HCMV rate in age range 26–31 years in Iran and Jahan et al. (2017) in the age range 21–30 years in Bangladesh. The current study contrasts with the Mashhad study by Safabakhsh et al. (2013). In previous studies, no significant associations were seen between age, the number of children, gender, educational level and CMV infection (Motamedi et al., 2009). In Spain, the rate was 66.7% in women aged 2–60 years (Zalei et al., 2017).

Known factors for HCMV infection, counting age, socio-economic status, sexual promiscuity, education, and blood transfusion (Maingi & Nyamache, 2014; Shigemi et al., 2015), were not found associated with HCMV infection in our study. None of the single pregnant women was seropositive (0.0%) for CMV IgM, while 23 (25.8%) married women tested seropositive. Statistically, marital status was not significantly associated with HCMV IgM prevalence. On the contrary, Umeh et al. (2015) reported that anti-HCMV IgM prevalence was significantly linked to marital status.

Educational background might be accountable for the high prevalence of CMV (Jahan et al., 2017), as all the pregnant women with no formal education had zero seroprevalence (0.0%). Tertiary and secondary education had 26.0% and 25.0%, respectively for HCMV IgM. Jahan et al. (2017) in Bangladesh reported a higher prevalence in residents of urban areas (81.0%), and 68.0% of them had more than five years of schooling.

Unemployed and students (33.3%), artisans (0.0%), business executive (60.0%), civil servants (17.2%) and traders (25.0%) were all seropositive for HCMV IgM antibodies. Jahan et al. (2017) in Bangladesh reported a higher prevalence of homemakers (88.3%) over other occupations. A higher prevalence of HCMV IgM antibodies occurred among Christians (26.0%) than no religion (22.2%), while zero prevalence (0.0%) was recorded for Islam. This observation is dissimilar to Jahan et al. (2017), who reported the highest rate in Islam (94.3%) over other religions in Bangladesh.

Pregnant women who were seronegative for HCMV in the first trimester had an approximately 14% chance of acquiring the virus during their pregnancy (Mussi-Pinhata et al., 2018; Plotkin, 2018). In this study, pregnant women in their first trimester had the highest HCMV prevalence (37.5%). This aligned with Jahan et al. (2017) in Bangladesh. It also agrees with Umeh et al. (2015), who reported that gestational age was not significantly linked with HCMV seroprevalence, even though prevalence was least in those who were in the first trimester of pregnancy.

The primary infection by HCMV occurs in 15–20% of pregnancies and transmits to the fetus in up to 40% of cases (Yinon et al., 2010; Zalei et al., 2017). Having children might also affect HCMV prevalence in pregnant women (Wujcicka et al., 2014; Jahan et al., 2017). However, nulliparous had the highest prevalence (32.4%). Those with 1-2 and 3-4 parity had a prevalence rate of 26.3% and 11.1%, respectively. This result disagrees with the study by Umeh et al. (2015), who reported higher HCMV seroprevalence in multiparous pregnant women. Jahan et al. (2017) in Bangladesh reported no association between parity/gravida and birth prevalence of CMV (p=0.95). Amongst the pregnant women in their study, 15.67% were of parity ≥ 3, 60.66% of parity 1-2 and 23.67% were primigravidae (Jahan et al., 2017).

A higher HCMV prevalence occurred in no history of abortion (26.6%) compared to those with a history (23.3%), nonetheless, this difference was insignificant. This result agrees perfectly with the finding of a previous study in Benue State, Nigeria, which reported that neither history of miscarriage nor the number of times miscarriage occurred showed any relationship with seroprevalence of anti-HCMV IgM antibodies (Umeh et al., 2015).

In this study, the rate of primary infection detected using CMV IgM was higher (25.6%) than in other studies. This figure was 8.2% lower than the study of Arabzadeh et al. in Kerman, which reported a prevalence rate of 33.8%. This difference can be ascribed to factors such as the study population and better laboratory techniques in detecting CMV infection in recent years. In addition, factors such as age, gender, residential place, health condition, and socio-economic conditions may be contributing factors. This study found a significant relationship between the prior history of abortion and primary HCMV infection, but other factors were not significantly associated.

As per preceding remarks in other studies (Drew et al., 2015; Chibwe et al., 2017), acute HCMV infection, as indicated by HCMV IgM antibodies, was high in the current study.

5. CONCLUSION

The study found a high HCMV IgM in pregnant women in Port Harcourt, Nigeria. The high prevalence of HCMV amongst pregnant women cannot be over-emphasised. As shown in most of the cited articles, pregnant women are highly susceptible to this infection. Therefore, they are advised to undergo antenatal check-ups since the infection could be asymptomatic.

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