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Original Article

Seroprevalence of IgM antibodies against cytomegalovirus (CMV) in HIV patients: single center study

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Abstract

Introduction: The presence of cytomegalovirus (CMV) immunoglobulin M (IgM) in human immunodeficiency virus (HIV)-positive individuals indicates an active infection or reactivation of the virus.

Methodology: This study investigated the seroprevalence of CMV IgM antibodies among HIV-positive individuals attending the Ahmadu Bello University Teaching Hospital in Nigeria.

Results: Fifty nine out of 92 participants who were tested with an enzyme-linked immunosorbent assay (ELISA) were positive for CMV IgM, resulting in a prevalence rate of 64.1%. Analysis of sociodemographic variables revealed a statistically significant association between CMV IgM seropositivity, and both gender and residence of the participants. Clinical variables also indicated a significant association between CMV IgM seropositivity and the duration of HIV infection. Multivariate analysis showed that participants in the 18–29 years age group, those with a secondary education level, the unemployed, and those who had been infected with HIV for 1–3 years were most likely to test positive for CMV infection. Joint and muscle pain were the most commonly reported symptoms among participants.

Conclusions: The high seroprevalence of CMV IgM antibodies found in this study suggests that CMV infection is widespread in the study area. Therefore, it is essential to regularly screen HIV-positive individuals for CMV during routine antiretroviral therapy visits to enable early detection and improve treatment outcomes.

Key words: seroprevalence; IgM; antibodies; infection; CMV; HIV.

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Introduction

Human immunodeficiency virus (HIV) infection and cytomegalovirus (CMV) infection are two significant viral infections that frequently coexist in individuals with compromised immune systems. The interaction between these viruses has profound implications for immune dysfunction and clinical outcomes. HIV remains a major global health concern affecting millions of people, and individuals living with HIV continue to be vulnerable to opportunistic infections [1]. CMV, a ubiquitous virus, causes significant morbidity and mortality in immunocompromised individuals, including those who are HIV-positive [2]. CMV is a member of the Herpesviridae family, and is characterized by double-stranded DNA. It causes infection that can lead to severe complications, including retinitis, encephalitis, and gastrointestinal disorders, posing substantial risks for people living with HIV/acquired immunodeficiency syndrome (AIDS) [2–4]. The virus primarily infects human cells and has a wide host range that helps establish lifelong latency following primary infection [5,6]. Immunosuppression is one of the factors that causes CMV reactivation, which leads to viral replication and shedding [5].

CMV exhibits broad cell tropism and can be transmitted through saliva, urine, breast milk, or sexual contact [7]. The reactivation of CMV in HIV-positive individuals presents significant challenges, as it can lead to severe clinical manifestations and accelerate HIV pathogenesis [8]. Despite the increased availability of antiretroviral therapy (ART), CMV infections remain a substantial concern due to their propensity for reactivation and the induction of severe symptoms, even in the presence of treatment [8]. The detection of CMV immunoglobulin M (IgM) antibodies in HIV-positive individuals indicates an active or recent CMV infection [9]. Detecting CMV IgM is especially important in developing countries with limited resources because it shows that an infection is present. This helps guide further testing and possible treatment, leading to better care and improved patient outcomes

[10]. Despite previous studies reporting a high burden of CMV in the region, CMV can cause serious complications, particularly in immunocompromised individuals, as it can remain dormant and reactivate unexpectedly [11–13]. Early detection of IgM antibodies is crucial to prevent the virus from rapidly progressing and leading to severe conditions like retinitis or organ damage [14]. Therefore, this study aimed to investigate the presence of CMV IgM antibodies in HIV-positive individuals attending the Ahmadu Bello University Teaching Hospital (ABUTH) in the northwest region of Nigeria.

Methodology

Study population

A hospital-based cross-sectional study was conducted on HIV-positive individuals attending antiretroviral clinics at the ABUTH in Shika, Nigeria. The sample size was determined to be 92 participants using Fischer's formula for cross-sectional studies and a prevalence rate of 2.5% as reported by Pathirana *et al*. [15].

The study included all consenting HIV-positive individuals who visited the antiretroviral clinic during the study duration, irrespective of whether they exhibited symptoms of CMV infection. HIV-positive individuals who did not give their consent and declined to participate were excluded from the study.

Ethical clearance was obtained from the Ethical and Human Research Committee of Ahmadu Bello University Teaching Hospital (ABUTHZ/HREC/F43/2023). Written informed consent was obtained from all participants in accordance with the standards of human experimentation and the Declaration of Helsinki, 1975, as revised in 2000.

Laboratory assays

Participants were initially tested for HIV status using rapid kit tests (Abbott Diagnostics, Chiba, Japan) to confirm their status. Subsequently, each participant's viral load was measured using polymerase chain reaction (PCR). The CD4 count was qualitatively determined to be either above or below 200 cells/μL using the VISITECT CD4 advanced disease test (AccuBio Ltd, Scotland, United Kingdom).

Following these preliminary tests, the sera of eligible participants were subjected to CMV IgM enzyme-linked immunosorbent assay (ELISA) using the Ratio Diagnostics kit (Frankfurt, Germany).

The CMV IgM assay operates on the principle of capturing IgM antibodies and identifying those specific to CMV. This is achieved by their ability to bind to an antigen conjugated to peroxidase. The capture process involves monoclonal antibodies bound to a solid phase (microtitration strips). The antigen used in this assay consisted of purified and inactivated CMV antigen.

The samples were pipetted into the wells, allowing for the capture of human IgM antibodies. CMV antigen conjugated to horseradish peroxidase (HRP) was added following a wash step to remove unbound substances. A substrate solution was introduced following another wash step to eliminate unbound reagents, and this led to color development. The absorbance of the solution was measured within 30 minutes of adding the stopping solution, using a microplate reader set to 450 nm. The mean absorbance values (optical density) for the cut-off controls and the samples were recorded. The ratio between the mean absorbance of the cut-off controls and that of the sample was calculated to determine the seropositive status of the samples. The samples were considered positive if the ratio was greater than 1.1 and negative if the ratio was less than 0.9.

Data analysis

The data obtained were imported into Microsoft Excel and analyzed using GraphPad Prism 8. The results were presented using tables and charts where appropriate. Group differences were assessed using the Chi square test or Fisher's exact test, depending on the context. Relative risks, attributable risks, and odds ratios, along with their 95% confidence intervals (CI), were estimated using the Koopman asymptotic score, Newcombe/Wilson with continuity correction, and Baptista-Pike models, respectively. p values less than 0.05 were considered statistically significant

Results

The study focused on HIV-positive individuals attending the antiretroviral clinics at ABUTH in Zaria. CMV IgM antibodies were detected with ELISA. The results revealed that 59 out of 92 participants tested seropositive for CMV IgM, indicating a prevalence rate of 64.1%.

Females exhibited a higher prevalence (72.1%) compared to males (48.4%) and this difference was statistically significant. The ages of the participants ranged from 18 to 65 years, with the highest prevalence observed in the 54–65 years age group (72.7%) and the lowest in the 30–41 years age group (60.5%). Urban residents had a prevalence rate of 71.6%, whereas rural residents had a prevalence of 44.0%; this difference was also statistically significant. In terms of education, individuals with no formal education had the highest

Table 1. Distribution of sociodemographic characteristics of participants according to cytomegalovirus (CMV) seropositivity.

Variables	No. tested (%)	Seropositive	Prevalence	Chi square	*p* value
Gender					
Male	31 (33.7%)	15	48.4%	5.038	0.0248
Female	61 (66.3%)	44	72.1%		
Total	92 (100%)	59			
Age (years)					
18–29	11 (12.0%)	7	63.6%	0.6812	0.8776
30–41	43 (46.7%)	26	60.5%		
42 –53	27 (29.3%)	18	66.7%		
54–65	11 (12.0%)	8	72.7%		
Total	92 (100%)	59			
Residence					
Urban	67 (72.8%)	48	71.6%	6.047	0.0139
Rural	25 (27.2%)	11	44.0%		
Total	92 (100%)	59			
Education					
None	13 (14.1%)	11	84.6%	3.099	0.3766
Primary	27 (29.4%)	17	63.0%		
Secondary	39 (42.4%)	24	61.5%		
Tertiary	13 (14.1%)	7	53.9%		
Total	92 (100%)	59			
Marital Status					
Single	29 (31.5%)	20	69.0%	0.4304	0.5118
Married	63 (68.5%)	39	61.9%		
Total	92 (100%)	59			
Occupation					
Employed	38 (41.3%)	23	60.5%	1.181	0.5541
Self-employed	48 (52.2%)	31	64.6%		
Unemployed	6 (6.5%)	5	83.3%		
Total	92 (100%)	59			

prevalence (84.6%), while those with tertiary education had the lowest (53.9%). Married participants had a lower prevalence (61.9%) compared to single participants (69.0%). The prevalence was higher among the unemployed (83.3%), than the employed (60.5%) (Table 1).

Based on the clinical data of the participants, the prevalence rates of CMV seropositivity were 76.7% for those on ART and 58.1% for those not on ART. A higher prevalence rate was observed among individuals with higher viral loads (70.6%) and higher CD4 counts (75.0%), although these differences were not statistically significant. The participants who had been infected with HIV for more than 3 years exhibited a 100% prevalence rate, compared to 85.7% among those infected for less than a year, and those infected between 1 and 3 years (Table 2).

Several independent factors were significantly associated with CMV seropositivity in the multivariable regression model, including gender, residency, and the duration of HIV infection. Participants aged 18–29 years [relative risk (RR) = 1.05, odds ratio (OR) = 1.14, attributable risk (AR) = 0.03] were more likely to test positive for CMV compared to the 30–41 years age group. Those with secondary education [RR = 1.14, OR = 1.37, AR = 0.08] were more likely to be CMV

Table 2. Distribution of clinical characteristics of participants according to cytomegalovirus (CMV) seropositivity.

Variables	No. tested (%)	Seropositive	Prevalence	Chi square	*p* value
Antiretroviral therapy (ART)					
Yes	30 (32.6%)	23	76.7%	3.041	0.0812
No	62 (67.4%)	36	58.1%		
Total	92 (100%)	59			
Viral Load					
< 30 copies	75 (81.5%)	47	62.7%	0.3781	0.5386
≥ 30 copies	17 (18.5%)	12	70.6%		
Total	92 (100%)	59			
Years since infection					
< 1yr	7 (7.6%)	6	85.7%	7.736	0.0209
1–3yr	76 (82.6%)	44	57.9%		
> 3yr	9 (9.8%)	9	100.0%		
Total	92 (100%)	59			
CD4 count (cells/µL)					
< 200	72 (78.3%)	44	61.1%	1.313	0.2519
≥200	20 (21.7%)	15	75.0%		
Total	92 (100%)	59			

Table 3. Multivariate analysis for cytomegalovirus (CMV) seropositivity according to sociodemographic characteristics.

Variables	% Seropositive	RR (95% CI)	OR (95% CI)	AR (95% CI)	*p* value
Gender					
Male	0.48	1.00	1.00	1.00	
Female	0.72	1.49 (1.00–2.21)	2.76 (1.12–6.79)	0.234 (0.03–0.46)	0.0248
Age (years)					
18–29	0.64	1.05 (0.63–1.75)	1.14 (0.29–4.51)	0.03 (-0.26–0.38)	0.8473
30–41	0.60	1.00	1.00	1.00	
42–53	0.67	1.10 (0.77–1.58)	1.31 (0.48–3.58)	0.06 (-0.17–0.31)	0.6012
54–65	0.73	1.20 (0.78–1.86)	1.74 (0.41–7.51)	0.12 (-0.13–0.49)	0.4523
Residence					
Urban	0.72	1.63 (1.02–2.60)	3.22 (1.24–8.33)	0.28 (0.06–0.52)	0.0139
Rural	0.44	1.00	1.00	1.00	
Education					
None	0.85	1.57 (0.90–2.74)	4.71 (0.734–30.28)	0.31 (0.01–0.71)	0.0892
Primary	0.63	1.17 (0.65–2.08)	1.46 (0.38–5.57)	0.09 (-0.23–0.42)	0.5815
Secondary	0.62	1.14 (0.65–2.00)	1.37 (0.39–4.87)	0.08 (-0.24–0.39)	0.6245
Tertiary	0.54	1.00	1.00	1.00	
Marital status					
Single	0.69	1.11 (0.82–1.52)	1.37 (0.54–3.49)	0.07 (-0.013–0.30)	0.5118
Married	0.62	1.00	1.00	1.00	
Occupation					
Employed	0.61	1.00	1.00	1.00	
Self-employed	0.65	1.07 (0.77–1.49)	1.19 (0.49–2.87)	0.04 (-0.17–0.25)	0.6991
Unemployed	0.83	1.38 (0.89–2.14)	3.26 (0.35–30.74)	0.23 (-0.01–0.72)	0.2805

AR: attributable risk; OR: odds ratio; RR: relative risk.

positive compared to those with tertiary education. Additionally, self-employed individuals [RR = 1.07, OR = 1.19, AR = 0.04] had a higher likelihood of CMV positivity compared to employed participants, while the unemployed group [RR = 1.38, OR = 3.26, AR = 0.23] had the highest likelihood (Table 3). Notably, individuals infected with HIV for less than a year [RR = 1.48, OR = 4.36, AR = 0.28] were more likely to be CMV positive compared to those who had been infected for 1–3 years (Table 4).

One of the limitations of the study was the potential for selection bias, as the study population was limited to a single teaching hospital, which may not have been representative of all HIV-positive individuals in the region

Among the participants, the majority reported symptoms related to CMV infection. These symptoms included fever, fatigue, joint and muscle pain, weight loss, skin rash, and low appetite. Joint and muscle pain were the most frequently reported symptoms, while skin rash and low appetite were the least recorded (Figure 1).

Discussion

To the best of our knowledge, this is the first recent study to investigate current CMV infection among HIV-positive individuals in the study area. We recorded a seroprevalence of 64.1% among the 92 participants. This finding is notably higher compared to other Nigerian studies on similar populations. Onoja *et al.* reported a CMV IgM prevalence of 21.3% among HIV-positive individuals, while Moses-Otutu *et al.* reported a prevalence of 32.7% [16,17]. However, Kamori *et al.* recorded CMV prevalence of 69.1% among the HIV-infected population in Tanzania [18]. Other studies conducted outside Nigeria generally reported lower prevalence rates [10,19–21]. The high prevalence observed in our study indicates a significant endemicity

Table 4. Multivariate analysis for cytomegalovirus (CMV) seropositivity according to clinical characteristics.

Variables	% Seropositive	RR (95% CI)	OR (95% CI)	AR (95% CI)	*p* value
Antiretroviral therapy (ART)					
Yes	0.77	1.32 (0.99–1.76)	2.37 (0.89–6.36)	0.19 (0.01–0.42)	0.0812
No	0.58	1.00	1.00	1.00	
Viral load					
< 30 copies	0.63	1.00	1.00	1.00	
> 30 copies	0.71	1.13 (0.79–1.60)	1.43 (0.46–4.49)	0.08 (– 0.14–0.37)	0.5386
Years since infection					
< 1yr	0.86	1.48 (1.04–2.11)	4.36 (0.50–38.05)	0.28 (0.10–0.73)	0.1501
1–3yr	0.58	1.00	1.00	1.00	
> 3yr	1.00	1.64 (1.30–2.09)	13.88 (0.78–248.00)	0.42 (0.30–0.81)	0.0137
CD4 (cells/µL)					
< 200	0.61	1.00	1.00	1.00	0.2519
> 200	0.75	1.23 (0.90–1.68)	1.91 (0.62–5.84)	0.14 (-0.06–0.41)	

AR: attributable risk; OR: odds ratio; RR: relative risk.

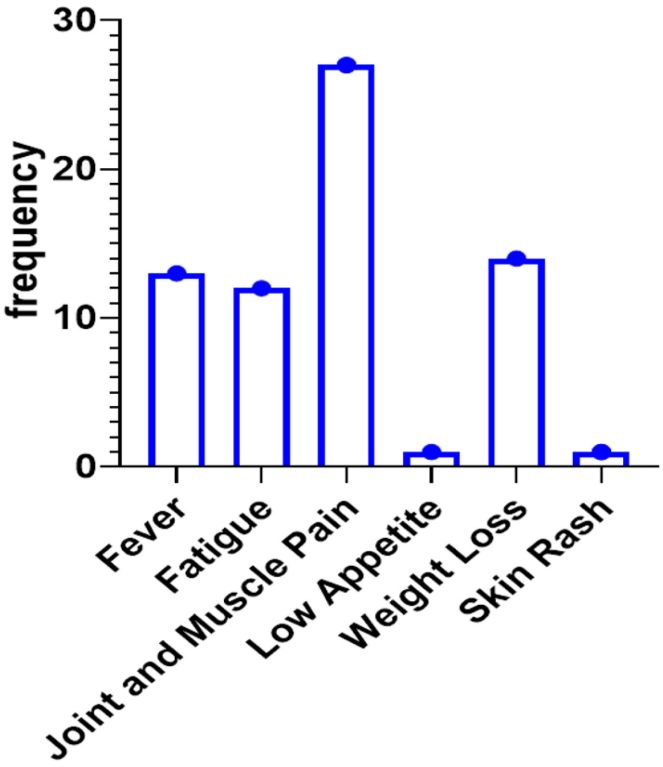
of CMV infection among the HIV-positive population in this region.

Our gender-based seroprevalence analysis revealed a higher prevalence of CMV among females compared to males, aligning with the findings reported by Zenebe *et al.*, who also observed higher prevalence in women [21]. However, contrasting results were reported by Moses-Otutu *et al*., who found a higher seroprevalence among males [17]. Despite these variations, the findings of this study were statistically significant. The highest seroprevalence was observed in the age group of 54–65 years, which contradicts the findings of Okonko *et al.*, who reported higher prevalence in the 21–40 years age group, and Ju *et al*., who found the highest prevalence in the 25–34 years age group [22,23]. Many CMV studies have focused on reproductive age groups, yet this study indicated a different age group as the most prevalent.

The highest seroprevalence was also observed among participants living in urban areas, consistent with Aljumaili *et al.*, who reported similar occurrences in urban populations [24]. Although a significant association between participants' residence and seropositivity was noted, it is possible that proper hygiene practices were not consistently followed in these urban settings. Regarding educational status, the highest prevalence was found among individuals with no formal education; which is different from Onoja *et al*., who reported the highest seroprevalence among those with a secondary level of education [14]. Furthermore, the highest prevalence was also seen among participants who were single and unemployed; contrasting with Mangare *et al*. who found higher seroprevalence among self-employed and married participants [25].

Analysis of the clinical characteristics of participants revealed that the highest prevalence of CMV infection was among those currently on ART and those with CD4 counts greater than 200 cells/mm³. This finding is consistent with Udeze *et al.* (2018), who reported similar results among HIV-infected individuals undergoing highly active antiretroviral therapy (HAART) [9]. Despite most participants being ART-naive and having CD4 counts less than 200 cells/mm³, the potential reactivation of CMV infection could explain the observed prevalence in this study. Furthermore, participants with viral loads exceeding 30 copies and who had been infected with HIV for more than 3 years also exhibited the highest prevalence of CMV. Similar findings were reported by Kiros *et al.* among treatment-naive HIV-1 infected patients, highlighting high HIV viral load as a significant risk factor for CMV disease progression, a trend that was also evident in our study [10].

Figure 1. Frequency of cytomegalovirus (CMV)-related symptoms.



Among the symptoms observed in the study participants, joint and muscle pain were the most common, aligning with studies by Xu *et al.* and Alanazi *et al*. which linked CMV infection to the incidence of rheumatoid arthritis characterized by muscle and joint pain. The inflammatory response induced by CMV infection could be responsible for these symptoms [26,27].

This study demonstrates the importance of routine screening HIV-positive individuals for CMV IgM, given the risk of reactivation and its potential to cause severe complications in those with weakened immune systems. Early detection of CMV infection in this vulnerable population can prompt timely interventions, reducing the likelihood of disease progression and improving clinical outcomes.

Conclusions

Despite the relatively small sample size, the high seroprevalence of CMV IgM antibodies observed in this study suggests that CMV infection was prevalent in the study area. This finding indicates that HIV-positive individuals, irrespective of their ART status, remain highly susceptible to CMV infection or its reactivation. Consequently, it is imperative to implement regular

CMV screening during routine ART visits. Early detection of CMV can enhance the effectiveness of ART and improve overall health outcomes for the HIV-positive population.

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Conflict of interests

No conflict of interests is declared.

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