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Seroprevalence of Cytomegalovirus among human immunodeficiency virus (HIV) positive patients attending HAART clinic in Kwara State, North-Central Nigeria

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ABSTRACT

Background: Cytomegalovirus (CMV) infection is usually acquired early in life resulting in an asymptomatic, subclinical, and mostly latent infection in immune-competent persons. Infections could be acquired during infancy with asymptomatic and undetectable infections in immune-competent persons. Cytomegalovirus infection has been documented to be prevalent among human immunodeficiency virus (HIV)-infected persons. This study was conducted to determine the seroprevalence of CMV among HIV positive patients as well as to determine the risk factors associated with the transmission of the disease. **Methods**: This study is a cross-sectional and epidemiological survey involving 400 HIV positive patients attending Highly Active Antiretroviral Therapy (HAART) clinic at Ilorin and Offa general hospital, Kwara State, North-Central Nigeria. 5 ml of serum was aseptically collected from the consented participants while Enzyme Linked Immunosorbent Assay (ELISA) kit (Biogenix, india) was used for the detection of CMV specific IgG antibodies. The test procedures were followed strictly according to the manufacturer instructions. **Result:** The results obtained showed that 368 samples out of the 400 samples screened for CMV were positive, which depicted a prevalence of 92% while 32(8.0%) were negative. Out of which, 270(73.4%) were females and 98(26.6%) were males. statistically, this research showed that there was a significant association between blood transfusion and CMV infection which may be suggestive that having blood transfusion could be a potential risk factor for CMV transmission (p=0.037), while there were no statistical relationship between the prevalence of CMV and some demographic characteristics such as age (p=0.71), gender (p = 0.748), educational Status (p = 0.104), occupation (p = 0.846), marital Status (p = 0.509), viral Loads (p =0.285), sexual activity (p = 0.314), and immunosuppression (p = 0.644). **Conclusion:** The high prevalence of CMV (92.0%) among the studied population is a clear indication that a vast majority of HIV seropositive individuals in Kwara State are at risk of contracting CMV infection and as such, more preventive measures should be employed to mitigate the menace of CMV infection among HIV positive patients.

Introduction

Cytomegalovirus (CMV) belongs to the virus family called human herpes viruses (HHV) [1]. It is a member of the subfamily *Betaherpesvirinae and family Herpesviridae.* Human herpes viruses

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types 6 and 7 are also members of this family *Betaherpesvirinae* which share some similar clinical symptoms [2]. Cytomegalovirus has similar characteristics such as viron structures, genome and virulence. Cytomegalovirus has a double stranded DNA with 162 hexagonal capsomeres surrounded by a lipid layer [2,3]. Human immunodefieciency virus (HIV)/AIDS has been documented as a principal threat to the public health most especially in Sub-Saharan Africa. Cytomegalovirus has been proven to increase HIV replication and hasten the advancement of HIV infection to AIDS. In immunocompetent individuals, symptomatic disease usually manifests as infectious mononucleosis. It is characterized by malaise, headache, sore throat and fatigue. Fever is also commonly present and may persist for weeks in 98% of cases of CMV mononucleosis [4]. Cytomegalovirus infection is more prevalent in populations risk for HIV infection; approximately 75% of injection drug users and >90% of homosexual men who are infected with HIV have detectable IgG antibodies to CMV [5]. Cytomegalovirus infection is nearly ubiquitous in HIV-infected subjects and may lead to CMV end-organ disease (EOD) and death as a consequence of the impaired immunity [6]. The detection of virus specific IgG and IgM antibodies makes the diagnosis of acute/primary virus infections possible even without any visible clinical symptoms[7]. Cytomegalovirus infection is more prevalent in developing countries [8] and the presence depends on some epidemiological factors [9]. Cytomegalovirus infection is endemic worldwide, with a 30–61% seroprevalence in developed countries and 60–100% seroprevalence in developing countries [10]. The immunological feature of CMV in immunocompromised patients are distinctive as the characteristics, the treatment measures and prognostic factors differs within HIV patients with CMV and patients infected with just CMV. Laboratory examination remains the mainstay of diagnosis for CMV infection because of the unspecific clinical symptoms. Cytomegalovirus viral replication and serum immunological methods which includes the detection of CMV IgG, CMV IgM and CMV antigen PP65 are the major clinical methods [11].

Therefore, this study intends to provide data on the prevalence of CMV among HIV seropositive patients and to determine the associated risk factors for transmission.

Materials and Methods

The samples were collected from various HAART Clinics which were Civil Service Clinic, Sobi Specialist Hospital, Alagbado and General Hospital Offa. Ethical clearance was obtained from Kwara State Ministry of Health with approval number: MOH/KS/EU/777/560

Specimen collection

About 5 ml of blood samples were collected from each consenting HIV patients by venipuncture, and transferred into sterile anticoagulant-free bottles (Plain bottles), and allowed to clot. The clotted blood samples were centrifuged (3000 rpm, 5 min), and the serum was transferred into cryovials and stored at -20°C until required for analysis.

Serological screening

Serum samples were tested for the presence of IgG antibodies using commercially available Enzyme Linked Immunosorbent Assay (ELISA) kit (manufacturer - Biogenix) for detection of CMV specific IgG antibodies. The tests were performed and interpreted according to the manufacturer's instructions.

Results

Seroprevalence of cytomegalovirus among HIV positive patients

Table 1 showed that out of the 400 test samples of HIV positive patients analysed for seroprevalence of cytomegalovirus in North Central, Kwara state, 368 (92.0%) (table 1) were found to be positive for CMV, while 32(8.0%) were negative.

Age distribution of cytomegalovirus among HIV positive patients.

Table 2 showed the age groups of HIV Positive Patients, age group 36-45 (28.3%) had the highest prevalence of CMV infection, followed by age groups; 26-35 years (22.3 %), 46-55 years (20.1%), 56-65 years (12.5%), <25 years (10.3%), 66-75 years (4.9%) and the age group with lowest percentage was 76-85 years (1.6%). There was no statistical significance association of age groups (p=0.71) with seroprevalence of CMV among HIV positive patients at p<0.05.

Gender distribution of cytomegalovirus among HIV positive patients.

As shown in **table (3)**; 26.6% of HIV positive patients with CMV infection were male while the 73.4% of HIV positive patients with CMV infection were female. There was no statistical significance

association of Gender (p=0.748) with seroprevalence of CMV among HIV positive patients at p>0.05.

Educational level distribution of cytomegalovirus among HIV positive patients

Table 4 showed that 54.9% of the HIV positive patients with secondary level of education had the highest CMV infection. This is followed by those with tertiary level of education (28.8%), participants with primary level of education had (11.4%). 4.9% of them were not educated and had the least prevalence of CMV infection. There was no statistical significance association of educational level (p=0.104) with seroprevalence of CMV among HIV positive patients at p<0.05.

Occupation distribution of cytomegalovirus among HIV positive patients

Table 5 showed that HIV positive patients with CMV co-infection that are self-employed had the highest prevalence 14.4%. while House girls had the least prevalence of 0.5%. there was no statistical significance association of occupations (p=0.846) with seroprevalence of CMV among HIV positive patients at p>0.05.

Marital status distribution of cytomegalovirus among HIV positive patients

As shown in **table (6)** below; married HIV positive patients had the highest prevalence of CMV infection (57.1%), this was followed by those that were single (34.2%), divorced (6.0%) and then widowed had the lowest prevalence (2.7%). There was no statistical significance association of marital status (p=0.509) with seroprevalence of CMV among HIV positive patients at p<0.05.

Viral loads and immunosuppression distributions of cytomegalovirus among HIV positive patients

As shown in **table (7)** below, HIV positive patients with viral loads <20 had highest prevalence of CMV infection (59.2%), followed by those with 20-100 viral loads (17.9%), 100-1000 viral loads (13.6%), 1000-10000 and 10000-100000 viral loads had a prevalence of 4.4% and 100000-1000000 viral loads had lowest prevalence with 0.5%. There was no statistical significance association of viral loads (p=0.285) with seroprevalence of CMV and associated risk factors among HIV positive patients at p<0.05.

Immunosuppression distribution showed that 90.2% of HIV positive patients infected with cytomegalovirus were immunosuppressed while 9.8% of HIV positive patients infected with CMV were not immunosuppressed. There was no statistical significance association of immunosuppression (p=0.644) with seroprevalence of CMV among HIV positive patients at p<0.05.

History of blood transfusion in relation to cytomegalovirus prevalence among HIV positive patients

Based on history of blood transfusion, 21.7% of HIV positive for CMV infection had previous history of blood transfusion and 78.3% had no history of blood transfusion. There was a statistical significance association of history of blood transfusion (p=0.037) with seroprevalence of CMV among HIV positive patients at p>0.05.

Sexual activity of participants in relation to cytomegalovirus prevalence among HIV positive patients

Table 9 below showed that 94.0% of HIV positive patients infected with cytomegalovirus were sexually active and 6.0% of HIV patients infected with CMV were not sexually active. There was no statistical significance association of sexual activeness (p=0.314) with seroprevalence of CMV among HIV positive patients at p<0.05.

Table 1. Seroprevalence of cytomegalovirus among HIV positive patients.

Group	Total number screened	Cytomegalovirus Positive		Cytomegalovirus Negative	
		Freq.	Per.	Freq.	Per. (%)
HIV positive patients	400	368	92.0%	32	8.0%

Table 2. Age distribution of cytomegalovirus among HIV positive patients.

Variables	Number of cytomegalovirus among HIV Positive patients		X^2	*p*-value	Remarks
	Frequency N=368	Positive			
Age (years)			3.789	0.71	**NS**
<25	38	10.3%			
26-35	82	22.3%			
36-45	104	28.3%			
46-55	74	20.1%			
56-65	46	12.5%			
66-75	18	4.9%			
76-85	6	1.6%			
Total	**368**	**100%**			

Key. NS=Not Significant

Table 3. Gender Distribution of cytomegalovirus among HIV positive patients.

Variables	Number of cytomegalovirus among HIV positive patients		X^2	*p*-value	Remarks
	Frequency N=368	Positive			
Gender			0.5	0.748	**NS**
Male	98	26.6%			
Female	270	73.4%			
Total	**368**	**100%**			

Key. NS = Not significant

Table 4. Educational level distribution of cytomegalovirus Among HIV positive patients.

Variables	Number of cytomegalovirus among HIV Positive patients		X^2	*p*-value
	Frequency N=368	Positive		
Educational level			6.155	0.104
Uneducated	18	4.9%		
Primary	42	11.4%		
Secondary	202	54.9%		
Tertiary	106	28.8%		
Total	**368**	**100%**		

Key. NS= Not Significant

Table 5. Occupation distribution of cytomegalovirus among HIV positive patients.

Variables	**Number of cytomegalovirus among HIV positive patients**		**X^2**	***p*-value**	**Remarks**
	Frequency N=368	**Positive**			
Occupation			17.057	0.846	**NS**
Traders	46	12.5%			
Drivers	6	1.6%			
Students	24	6.5%			
House girl	2	0.5%			
Civil servant	36	10.0%			
Business men	12	3.3%			
Self employed	52	14.1%			
Shop sales	24	11.4%			
Public servant	2	1.1%			
Barbers	2	0.5%			
Farmers	2	0.5%			
Market sales woman	8	2.2%			
Sales girl	4	1.1%			
Retirees	14	3.8%			
House wives	16	4.3%			
Chemists	2	0.5%			
Contract workers	12	3.3%			
Tailors	30	8.1%			
Unemployed	6	1.6%			
Cashiers	12	3.3%			
Vendors	4	0.5%			
Teachers	4	1.1%			
Sales women	22	6.0%			
Shop owners	8	2.2%			
TOTAL	**368**	**100%**			

Key. NS = Not significant

Table 6. Marital status distribution of cytomegalovirus among HIV positive patients

Variables	**Number of cytomegalovirus among HIV positive patient**		**X^2**	***p*-value**	**Remarks**
	Frequency N=184	**Positive**			
Marital status			2.316	0.509	**NS**
Single	126	34.2%			
Married	210	57.1%			
Divorced	22	6.0%			
Widow	10	2.7%			

Key: NS=Not Significant

Table 7. Viral loads and immunosuppression distribution of cytomegalovirus among HIV positive patients.

Variables	Number of Cytomegalovirus among HIV positive patients		X^2	*p*-value	Remarks
	Frequency N=368	Positive			
Viral loads (Copies/ml)			6.229	0.285	**NS**
<20	218	59.2%			
20-100	66	17.9%			
100-1000	50	13.6%			
1000-10000	16	4.4%			
10000-100000	16	4.4%			
100000-1000000	2	0.5%			
Total	**368**	**100%**			
Immunosuppression			0.214	0.644	**NS**
Suppressed	332	90.2%			
Unsuppressed	72	9.8%			
Total	**368**	**100%**			

Key:
NS = Not significant
<20-1000 Copies/ml (Suppressed)
>1000 Copies/ml (Unsuppressed)

Table 8. History of blood transfusion distribution of cytomegalovirus among HIV positive patients.

Variables	Number of cytomegalovirus among HIV positive patients		X^2	*p*-value	Remarks
	Frequency N=368	Positive			
History of blood transfusion			4.348	0.037*	**S**
Yes	80	21.7%			
No	288	78.3%			
Total	**368**	**100%**			

Key. S = significant

Table 9. Sexual activity of participants in relation to cytomegalovirus prevalence among HIV positive patients.

Variables	Number of cytomegalovirus among HIV positive patients		X^2	*p*-value	Remarks
	Frequency N=368	Positive			
Are they Sexually Active?			1.012	0.314	NS
Yes	346	94.0%			
No	22	6.0%			
Total	**368**	**100%**			

Key. NS = Not Significant

Discussion

Cytomegalovirus is a very rampant infection complicating AIDS. Sexual transmission appears to be the prevalent route of infection in adults, though CMV can also be spread through oropharyngeal sections, urine, breast milk, and blood [10]. Most patients with AIDS who develop clinical signs and symptoms of CMV infection probably have reactivation of previous infection rather than primary infection [10]. The prevalence of CMV in Sub Saharan Africa is high but it usually presents as an opportunistic infection in adults [10].

The prevalence of CMV IgG among HIV positive patients in this study was 368(92.0%). This finding compared favourably with the prevalence of 89.4% reported from HIV seropositive patients in Khammam, India. Research conducted in Benin City, Edo State reported a prevalence of 98.8% while another research in Iran reported 94%. In USA, a study reported a prevalence of 91.6%. Similarly, other researches in Botswana and in Lagos Nigeria reported prevalence of reported 95.3% and 100% respectively [11,12]. They all reported higher prevalence respectively corresponding to result in Kwara State, North-central Nigeria. However, research reported an unusually low prevalence of 59.9% in Ghana [13].

The age distribution of CMV among HIV positive patients in this study shows that participants 36-45 years (28.3%) had the highest prevalence of CMV infection this in agreement with research conducted in India who observed the highest prevalence of CMV infections among participants aged between 31 and 40 years [14].

Findings in this study are not in agreement with publications in USA where higher prevalence was recorded among younger participants [15]. Gender- related distribution of CMV IgG among HIV positive patients in this study showed higher prevalence in females 135 (73.4%) than males 49 (26.6%). This finding is in agreement with a 2013 research conducted in Benin city, in which the prevalence of CMV infection in female were higher than male counterparts i.e. 252 (73.7%) in female and male with 90 (26.3%) [16]. And also in agreement with the report by 2017 research in Bida, showed that the prevalence in female is higher than male with IgG CMV among HIV positive patients i.e. 218(83.8%) and 106(84.8%) [17]. This finding is however in contrast with some 2010 and 2015 reports which both reported higher prevalence in male than in females [18,19].

The prevalence of CMV and associated risk factors among HIV positive patients based on educational level is closely related with research conducted in Sokoto [19]. The finding is also in agreement with previous 2010 report in Iran who reported the Elementary level (Primary) with 130(96.2%), which had the highest prevalence and University (Tertiary) with 12(85.7%) had the lowest prevalence of cytomegalovirus among HIV positive patients [20]. Other researchers reported contrasting prevalence patterns in relation to educational status of their sampled population in 2013 [21].

The patterns of prevalence of CMV in relation to occupation as a potential risk factor for the transmission of CMV in this study is related to the patterns described in a 2015 report which stated that traders 100(100%) had the highest prevalence while both civil servants 16(80.0%) and the unemployed 16(80.0%) had the least prevalence of CMV [22].

The prevalence of CMV based on marital status in this research which concluded that the married participants had the highest rate (57.1%) of infection is in exact agreement with previous report by Fowotade *et al*. (2015) who reported the married patients with 118(92.9%) had the highest prevalence and others (Widowed) with 13(92.7%) had the lowest prevalence of CMV among HIV positive patients based on marital status [23] and also in agreement with a 2017 findings showing that married 25(53.2%) as the highest prevalence of CMV followed by single 11(52.3%) which has the lowest prevalence of CMV among HIV positive patients based on marital status [24].

Ojide et al. (2013) in Benin had the highest prevalence of CD4 count < 200 (50.8%) and lower prevalence of >200 (49.2%) as well as research by **Omosigho et al.** (2017) in Bida showed similar patterns of < 200 (88.4%) with the findings in this research on the prevalence of CMV infection in relation with the CD4 counts of the participants.

Of the participants in this current research, 21.7% had received blood transfusion and this finding is in agreement with previous reports in 2015 [23]. In Bida, Niger State, in 2015 research reported that there was higher prevalence of IgG CMV among HIV positive patients with no history of blood transfusion 140(193.6%) than those with history of blood transfusion 38 (90.0%), while in 2017 another researcher also reported that there was higher prevalence of IgG CMV among HIV positive patients with no history of blood transfusion

128(83.0%) than those with history of blood transfusion 35(94.6%) [25].

Sexual transmission is one of the methods implicated in the transmission of HIV as well as CMV and this is likely the single most important method of transmission in this research as most of the participants were sexually active. This finding is related with previous 2015 reports in Ilorin which discussed on sexual partners (single and multiple) which the multiple partners had the highest prevalence of IgG CMV among HIV positive patients with 118(97.5%) and the lowest with single partners had 51(86.4%) [23].

Conclusion

This study revealed an overall high prevalence of CMV infection among HIV positive patients on HAART in Ilorin, Kwara State as 368(92.0%). This possibly implies that both HIV and CMV share similar routes of transmission and as such, more public enlightenment programs should be undertaken by relevant authorities as a conduit to improve the awareness levels of the residents within the study area on adequate preventive and control measures to alleviate the burden of HIV and CMV coinfections among residents of Ilorin, Kwara State. The research also put forth relevant information about the risk factors associated with the spread of CMV as well as the socio demographic characteristics of the participants with the view to deduce common patterns implicated with lifestyle of individuals and the transmission of the disease.

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Conflict of interest

The authors declare no conflict of interest.

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