

Seroprevalence of Herpes Simplex Virus Type 2 Antibodies among HIV- Infected Individuals Attending Lautech Teaching Hospital, Ogbomoso, Oyo State

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ABSTRACT

Background: Herpes Simplex Virus type 2 (HSV-2) is an opportunistic infection of public health concern globally among people living with HIV (PLWHIV). This study focused on the prevalence of and risk factors for HSV among PLWHIV to help them live a healthier life.

Methods: A cross-sectional study was conducted at the Retroviral Clinic of LAUTECH Teaching Hospital, Ogbomosho, between December 2021 and June 2022. A total of 208 consenting individuals were enrolled in the study. Type-specific anti-HSV-2 IgG and IgM antibodies were detected using ELISA. Risk factors and the prevalence of HSV type 2 were analyzed using SPSS version 23.

Results: The prevalence rates of HSV-2 IgG among 104 PLWHIV and 104 non-HIV were 40.4% and 0%, respectively. The prevalence of HSV-2 IgM among 104 PLWHIV and 104 non-HIV was 7.7% and 4.8%, respectively. A significant HSV-2 prevalence (3.8%, $P=0.049$) was reported among those with one or more sexual partners. The percentages of females and males who were positive for HSV-2 IgG were 40.0% and 41.0%, respectively.

Conclusion: HSV-2 infection remains a significant comorbidity among PLWHIV. Increased awareness, screening, and prevention efforts are essential to reduce the dual burden.

Keywords: STDs; Serology; Immunology

Word count: 189

INTRODUCTION

Herpes simplex virus-2 (HSV-2) related genital herpes is a worldwide problem, with an estimated 491 million (13%) people aged 15 to 49 years infected in 2016. (WHO, 2021). The seroprevalence of HSV-2 is greater in PLWHA than in HIV-negative people globally, and it is especially high in sub-Saharan Africa (Harfouche *et al.*, 2021). Multiple reasons could explain these findings: vaginal ulceration provides a location for HIV entrance in HIV-positive people, and the associated inflammation increases the number of activated cells that HIV can target (WHO, 2019). Both symptomatic and asymptomatic HSV-2 reactivation may stimulate HIV shedding in the vaginal tract and increase HIV levels in the blood (Mawak *et al.*, 2012). Because silent HSV-2 infections in HIV-positive people are linked to increased HIV transmission and can hasten the progression of HIV/AIDS illness, screening should be administered to all patients with HIV who do not have a history of genital herpes and all individuals at risk should receive HSV-2-specific education and counseling, as HSV-2-negative HIV-infected patients have a significantly greater risk of HSV-2 acquisition (Mawak *et al.*, 2012).

Studies on HSV-2 conducted in Nigeria have focused mostly on pregnant women, and very few studies have compared HSV-2 among PLWHA; therefore, the goal of this study This study aims to determine the prevalence of HSV-2 antibodies and identify associated risk factors among PLWHIV in Ogbomoso, Nigeria.

METHODS

Study Design

This descriptive cross-sectional study was carried out at the Retroviral Clinic of LAUTECH Teaching Hospital (LTH), Ogbomoso, Oyo State, Nigeria, using a random sampling technique. Sample size was determined using Fisher's formula, and ethical approval was obtained from the ethical committee of the hospital in line with the code of ethics for biomedical research involving human subjects. Informed consent was obtained from each participant after carefully explaining the study to them, and an open-ended questionnaire was self-administered.

Sample collection and HSV-2 antibody testing

Blood samples were collected aseptically from 208 consenting participants (104 HIV and 104 non-HIV participants) via venipuncture into plain sterile bottles. Serum samples were separated and stored at -70°C until used for analysis. Serological testing for HSV-2 IgG and IgM was performed using FDA-certified commercial ELISA kits manufactured by Diagnostics Automation/Cortez with a sensitivity and specificity rate of 100% and 94.1% respectively (Diagnostics, Inc., 23961 Craftsman Road, Suite E/F, Calabasas, California 91302, USA). All laboratory analysis was carried out at the Metabolic Laboratory of LAUTECH Teaching Hospital.

Data analysis

Statistical analysis: All the data were analysed using the Statistical Package for Social Sciences (SPSS) version 23. The seroprevalence for HSV-2 was expressed as a percentage for the entire study group, with the P-value set at ($P < 0.05$) for significance at the 95% confidence interval.

RESULTS

Table 1: Sociodemographic Findings on the study population

Variables	Control group n (%)	Cases n (%)	χ^2	p-value
Age (Years)				
18 - 28	39(37.5)	18(17.3)	22.455	*0.001
29 - 38	19(18.3)	26(25.0)		
38 - 48	19(18.3)	21(20.2)		
49 - 58	10(9.6)	29(27.9)		
59 - 68	7(6.7)	7(6.7)		
69 - 78	8(7.7)	3(2.9)		
Above 78	2(1.9)	0(0.0)		
Gender				
Male	46(44.2)	39(37.5)	0.975	0.323
Female	58(55.8)	65(62.5)		
Religion				
Christianity	69(66.3)	73(70.2)	2.675	0.444
Islam	35(33.7)	29(27.8)		
Traditional	0(0.0)	1(1.0)		

Others	0(0.0)	1(1.0)		
Marital Status				
Single	40(38.4)	17(16.3)		
Married	57(54.8)	59(56.7)		
Cohabiting	0(0.0)	1(1.0)	23.147	*0.000
Divorced	1(1.0)	6(5.8)		
Separated	0(0.0)	4(3.9)		
Widowed	6(5.8)	17(16.3)		
Ethnicity				
Yoruba	99(95.2)	87(83.7)		
Hausa	1(1.0)	3(2.8)	7.441	0.059
Igbo	1(1.0)	5(4.8)		
Others	3(2.8)	9(8.7)		
Level of education				
No education	7(6.7)	4(3.9)		
Primary education	10(9.6)	10(9.6)	1.569	0.667
Secondary education	48(46.2)	44(42.3)		
Tertiary education	39(37.5)	46(44.2)		

χ^2 : Chi square test, p-value < 0.05 indicates significance

Table 2: Relationship between HSV-2 IgM serostatus and risk factors.

Risk Factors	Control			Cases		
	Status		χ^2 (p-value)	Status		χ^2 (p-value)
	Positive n (%)	Negative n (%)		Positive n (%)	Negative n (%)	
No of Sex Partners						
1	4 (4.3)	90 (95.7)	9.392	8 (8.8)	83 (91.2)	1.238
2	0 (0.0)	8 (100)	(0.009)	0 (0.0)	8 (100)	(0.744)
3	1 (50.0)	1 (50.0)		0 (0.0)	3 (100)	
More	0 (0.0)	0 (0.0)		0 (0.0)	2 (100)	
Age (First Sex)						
18 – 28	4 (4.6)	83 (95.4)	0.051	5 (5.8)	81 (94.2)	2.469
29 – 38	1 (5.9)	16 (94.1)	(0.821)	3 (16.7)	15 (83.3)	(0.116)
Change Sexual Partners						
Never	5 (6.8)	69 (93.2)	2.059	6 (8.7)	63 (91.3)	0.498
Occasionally	0 (0.0)	28 (100)	(0.357)	2 (6.5)	29 (93.5)	(0.779)

Often	0 (0.0)	1 (100)		0 (0.0)	4 (100)	
Polygamous						
Yes	1 (6.7)	14 (93.3)	0.132	3 (10.0)	27 (90.0)	0.316
No	4 (4.5)	85 (95.5)	(0.716)	5 (6.8)	69 (93.2)	(0.574)
Condom						
Yes	2 (11.8)	15 (88.2)	2.149	3 (5.7)	50 (94.3)	0.628
No	3 (3.4)	84 (96.6)	(0.143)	5 (9.8)	46 (90.2)	(0.428)
Sexually Active						
Yes	4 (4.2)	91 (95.8)	0.855	4 (5.5)	69 (94.5)	1.689
No	1 (11.1)	8 (88.9)	(0.355)	4 (12.9)	27 (87.1)	(0.194)
Sex Practice						
Oral Sex	0 (0.0)	0 (0.0)	0.156	0 (0.0)	2 (100)	0.722
Anal Sex	0 (0.0)	3 (100)	(0.693)	0 (0.0)	6 (100)	(0.697)
Vaginal Sex	5 (5.0)	96 (95.0)		8 (8.3)	88 (91.7)	

χ^2 : Chi square test, p-value < 0.05 indicates significance

Table 3: Relationships between HSV-2 IgG serostatus and risk factors.

Risk Factors	Cases		
	Status		χ^2 (p-value)
	Positive n (%)	Negative n (%)	
No of Sex Partners			
1	39 (42.9)	52 (57.1)	7.799 (0.049)
2	1 (12.5)	7 (87.5)	
3	0 (0.0)	3 (100)	
More	2 (100)	0 (0.0)	
Age (First Sex)			
18 – 28	36 (41.9)	50 (58.1)	0.450 (0.503)
29 – 38	6 (33.3)	12 (66.7)	
Change Sex Partners			
Never	26 (37.7)	43 (62.3)	2.231 (0.328)
Occasionally	13 (41.9)	18 (58.1)	
Often	3 (75.0)	1 (25.0)	
Polygamous			
Yes	8 (26.7)	22 (73.3)	3.296 (0.069)
No	34 (45.9)	40 (54.1)	
Condom			

Yes	22 (41.5)	31 (58.5)	0.057 (0.812)
No	20 (39.2)	31 (60.8)	
Sexually Active			
Yes	29 (39.7)	44 (60.3)	0.044 (0.834)
No	13 (41.9)	18 (58.1)	
Sex Practice			
Oral Sex	0 (0.0)	2 (100)	1.588 (0.452)
Anal Sex	3 (50.0)	3 (50.0)	
Vaginal Sex	39 (40.6)	57 (59.4)	

χ^2 : Chi square statistic, p-value < 0.05 indicates significance

Table 4: Relationship between HSV-2 IgG serostatus and clinical features

Clinical Features	Cases		
	Status		χ^2 (p-value)
	Positive n (%)	Negative n (%)	
Genital Lesion			
Current	3 (60.0)	2 (40.0)	2.216 (0.330)
Before	7 (29.2)	17 (70.8)	
Never	32 (42.7)	43 (57.3)	
Dysuria			
Yes	3 (30.0)	7 (70.0)	0.496 (0.481)
No	39 (41.5)	55 (58.5)	1.815 (0.178)
Abnormal Discharge			
Yes	7 (58.3)	5 (41.7)	
No	35 (38.0)	57 (62.0)	0.871 (0.351)
Vaginal Infection			
Yes	10 (33.3)	20 (66.7)	0.023 (0.880)
No	32 (43.2)	42 (56.8)	3.507 (0.061)
Dyspareunia			
Yes	5 (38.5)	8 (61.5)	
No	37 (40.7)	54 (59.3)	3.558 (0.059)
Diabetes			
Yes	1 (11.1)	8 (88.9)	

No	41 (43.2)	54 (56.8)	
Genital Ulcer			
Yes	0 (0.0)	5 (100)	
No	42 (42.4)	57 (57.6)	

Table 5: Relationships between HSV-2 IgM serostatus and Clinical features.

Clinical Features	Control			Cases		
	Status		χ^2 (p-value)	Status		χ^2 (p-value)
	Positive n (%)	Negative n (%)		Positive n (%)	Negative n (%)	
Genital Lesion						
Current	0 (0.0)	2 (100)	2.322	1 (20.0)	4 (80.0)	2.436
Before	3 (9.7)	28 (90.3)	(0.313)	3 (12.5)	21 (87.5)	(0.296)
Never	2 (2.8)	69 (97.2)		4 (5.3)	71 (94.7)	
Dysuria						
Yes	1 (12.5)	7 (87.5)	1.121	1 (10.0)	9 (90.0)	0.083
No	4 (4.2)	92 (95.8)	(0.290)	7 (7.4)	87 (92.6)	(0.773)
Abnormal Discharge						
Yes	1 (3.7)	26 (96.3)	0.097	2 (16.7)	10 (83.3)	1.539
No	4 (5.2)	73 (94.8)	(0.755)	6 (6.5)	86 (93.5)	(0.215)
Vaginal Infection						
Yes	3 (7.)	35 (92.1)	1.247	6 (20.0)	24 (80.0)	8.995
No	2 (2.0)	64 (97.0)	(0.264)	2 (2.7)	72 (97.3)	(*0.003)
Dyspareunia						
Yes	0 (0.0)	8 (100)	0.438	2 (15.4)	11 (84.6)	1.238
No	5 (5.2)	91 (94.8)	(0.508)	6 (6.6)	85 (93.4)	(0.266)
Diabetes						
Yes	0 (0.0)	10 (100)	0.559	0 (0.0)	9 (100)	0.821
No	5 (5.3)	89 (94.7)	(0.455)	8 (8.4)	87 (91.6)	(0.365)
Genital Ulcer						
Yes	0 (0.0)	1 (100)	0.051	3 (60.0)	2 (40.0)	20.24 (*0.000)
No	5 (4.9)	98 (95.1)	(0.821)	5 (5.1)	94 (94.9)	

 χ^2 : Chi square test, p-value < 0.05 indicates significance

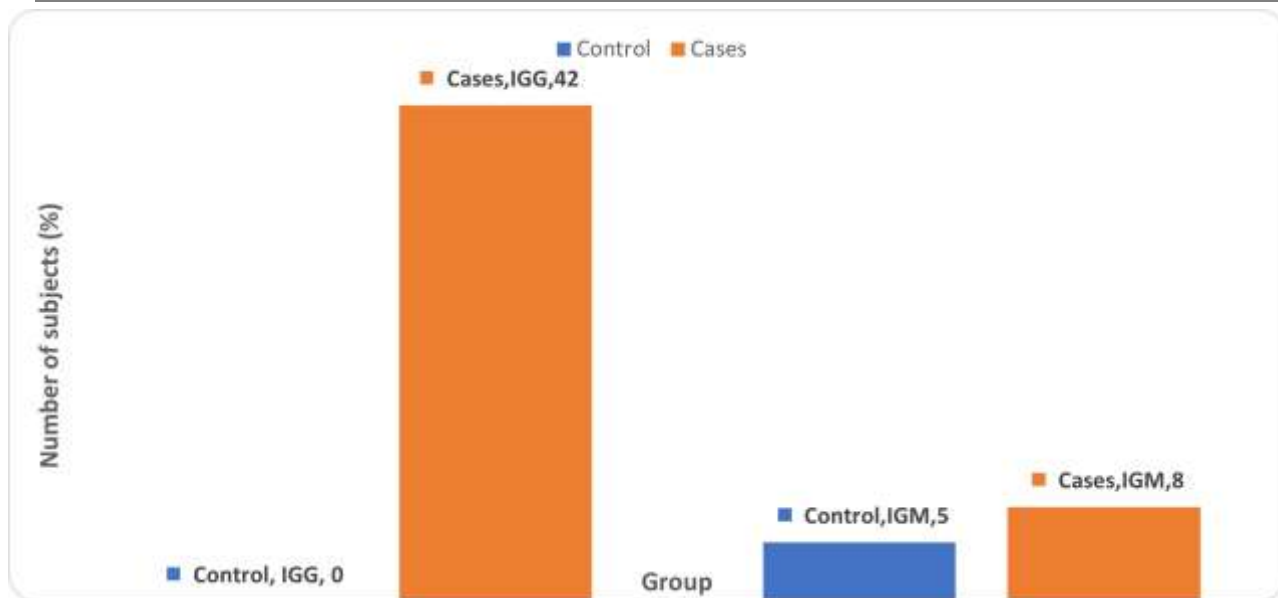


Figure 1: Prevalence of HSV-2 IgM and IgG among subjects in the study population

DISCUSSION

Serological testing, which detects antibodies or antigens in blood, is widely used in diagnosing infectious diseases and monitoring immune responses. However, it has several limitations that can impact its accuracy and reliability. For instance, variability in immune response, where individuals vary in how strongly and how quickly they produce antibodies, as well as variations in the sensitivity and specificity of different commercial test kits, can affect diagnostic accuracy, particularly in low-resource settings. However, the importance of assessing the prevalence of HSV in a developing country like Nigeria, particularly among HIV-infected people who may be at risk of various complications due to their immunosuppressed state, cannot be overemphasised. The best strategy for identifying suspected sexually transmitted illnesses and creating a way for possible interventions is to screen people living with HIV.

Serological testing in this study revealed that the prevalence of HSV-2 IgG among HIV-positive individuals was 40.4%, which is consistent with the 38.8% prevalence of HIV/HSV type 2 co-infection reported in a multi-center study among pregnant women by Mathew *et al.* (2019) and 36.4% in central Nigeria by Yunusa *et al.* (2019). All these studies took place in hospital settings, which could potentially explain their similarities. However, Nwadike *et al.* (2020) reported a higher prevalence of HSV-2 IgG antibodies in HIV patients, with an increase of 70.0%. The difference in this prevalence could be attributable to the variation in the study population. Most affected were the 29- to 38-year-old age groups, suggesting high infection rates among young adults and economically productive groups. This conclusion is in line with the findings of Nwadike *et al.* (2020), who reported that the most affected group was the 31- to 35-year-old group.

In this study, the prevalence of anti-HSV-2 antibodies did not vary significantly between males and females; however, a higher prevalence in males was reported by Karad and Khade (2016). Triggers for the recurrence of HSV-2 infection, such as illness, stress, and fatigue, could account for the slightly greater percentage recorded among the males. Such an increase may be because of a high level of sexual activity or having multiple sexual partners.

The rate of seropositivity for HSV-2 was highest (41.9%) among people who had their first sex within the 18-to-28-year-old group, which is consistent with the findings of the Karad and Khade (2016) study carried out in India. This shows that the younger a person is at the time of their first sexual encounter, the greater their risk of acquiring HSV-2. A total of 41.5% of the participants used a condom and had an HSV-2 infection, while 39.2% were in the group that did not use a condom, which contrasts with the findings of Ameh *et al.* (2016), who reported that the use of protection was found to protect against infection. The increase in HSV-2 infection among those who use condoms could be as a result of prior exposure to the infection. With respect to education, the seroprevalence (IgG or IgM of HSV-2) was highest among those with no education. Education has been

acknowledged to be advantageous in all facets of life, increasing the likelihood of making informed decisions as well as finding useful health information. Seropositivity for HSV-2 IgG was greater (50%) in participants who practiced anal sex than in those who did not. Anal sex is a risk factor for both HIV and HSV-2 infection because the lining of the anus is thin and can be easily damaged, which increases the vulnerability to infection (Shayo et al. 2017).

Seropositivity was highest (42.4%) among people with no genital ulcers, which proves that the absence of an ulcer does not mean the absence of HSV-2. This finding is consistent with the findings of Karad and Khade's (2016) study, which showed that the absence of genital ulcers does not preclude HSV-2 IgG seropositivity, as HIV infection promotes the shedding of HSV-2 in people who are even asymptomatic.

The prevalence of HSV-2 IgM detected in this study among non-HIV-coherent individuals was 4.8%, which is in line with the 2.7% reported by Enitan *et al.* (2020) and the 2.2% reported by Oluboyo *et al.* (2020). The prevalence of 7.7% in this study among HIV patients was quite low when compared with the 49.5% prevalence in a study carried out by Ige and Kolawole (2020). The rate of exposure to the virus and its prevalence in an area could potentially explain the variations.

This study revealed that 20.0% of HIV-infected individuals had vaginal infections (past and present) and that vaginal infection had a significant effect on both HSV-2 IgG and IgM serostatus among HIV-infected individuals ($p=0.003$). This indicates that people dually infected with HIV and HSV-2 suffer increased microbial diversity in their genital areas, according to Keller *et al.*, (2019).

The prevalence of HSV-2 IgM (8.3%) was greater among participants who practiced vaginal sex than among those who did not. This can be attributed to the larger mucosal surface area of the female genital tract, which is more prone to infection.

A greater percentage (10.0%) of individuals who practice polygamy had HSV-2 IgM antibodies for both HIV and non-HIV patients in this study, which agrees with the findings of Nwadike *et al.*, (2020). This result shows that multiple sexual partners increase the chance of contracting HSV-2 infection.

CONCLUSION

Although, most patients are asymptomatic for HSV-2 infection they can still serve as carrier of HSV-2 virus in transmission to uninfected people. Therefore, the creation of awareness of HSV-2 through health education among high-risk groups as well as an emphasis on the need to practice safe sex will play a key role, in limiting the spread of infection.

Conflict of Interest

The authors declare no conflict of interest.

Funding

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Ethics Statement:

Ethical approval was obtained from the ethical committee of LAUTECH Teaching Hospital, Ogbomoso, Nigeria (No: LTH/OGB/EC/2022/255), in line with the code of ethics for biomedical research involving human subjects. Informed consent was also obtained from each of the subjects after carefully explaining the concept of the study to them and questionnaires were self-administered.

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