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Human Papillomavirus Seroprevalence and Seroconversion Among Men Living With HIV: Cohort Study in South Africa

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Background: Men living with HIV (MLHIV) have a high burden of human papillomavirus (HPV)-related cancer. Understanding serological dynamics of HPV in men can guide decisions on introducing HPV vaccination and monitoring impact. We determined HPV seroprevalence and evaluated factors associated with HPV seroconversion among MLHIV in Johannesburg, South Africa.

Methods: We enrolled 304 sexually active MLHIV 18 years and older and collected sociobehavioral data, blood samples ($CD4^+$ counts, HIV-1 plasma viral load, and HPV serology), and genital and anal swabs [HPV DNA and HPV viral load (VL)] at enrollment and 6-monthly for up to 18 months. Antibodies to 15 HPV types were measured using HPV pseudovirions. Generalized estimating equations were used to evaluate correlates of HPV seroconversion.

Results: Median age at enrollment was 38 years (IQR: 22–59), 25% reported >1 sexual partner in the past 3 months, and 5% reported ever having sex with other men. Most participants (65%) were on antiretroviral therapy (ART), with median $CD4^+$ count of 445 cells/μL (IQR: 328–567). Seroprevalence for any HPV type was 66% (199/303). Baseline seropositivity for any bivalent (16/18), quadrivalent (6/11/16/18), and nonavalent (6/11/16/18/31/33/45/52/58) vaccine types was 19%, 37%, and 60%, respectively. At 18 months, type-specific seroconversion among 59 men whose genital samples were HPV DNA positive but seronegative for the same type at enrollment was 22% (13/59). Type-specific seroconversion was higher among men with detectable HIV plasma viral load (adjusted odds ratio = 2.78, 95% CI: 1.12 to 6.77) and high HPV VL (adjusted odds ratio = 3.32, 95% CI: 1.42 to 7.74).

Conclusions: Seropositivity and exposure to nonavalent HPV types were high among MLHIV. HPV vaccination of boys before they become sexually active could reduce the burden of HPV infection among this at-risk population.

Key Words: HPV, vaccine, serology, HIV, Africa

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INTRODUCTION

Men living with HIV (MLHIV) have a high burden of human papillomavirus (HPV) infections and related diseases, such as anal and oropharnygeal cancers as well as anogenital warts.[1,2] Although antiretroviral therapy (ART) attenuates risks of some sequelae of HPV infections, MLHIV remain a risk of these diseases and some evidence even suggests an increase in anal and oropharyngeal cancers.[3,4] Therefore, they are an important target group to consider when planning HPV prevention strategies.

HPV vaccines have been produced with bi/quadrivalent (16/18) and nonavalent (16/18/31/33/45/52/58) vaccines estimated to protect against 70% and 90% of cancer-causing high-risk (HR) infections.[5–7] However, because of the World Health Organization's (WHO) recommendation that limited public health funds should be directed primarily toward achieving a high coverage among girls to prevent cervical cancer, in most low- and middle-income countries, HPV vaccines are only routinely provided to girls.[8,9]

Natural infection with HPV can elicit immunoglobulin G antibodies to the L1 capsid protein of the HPV viral envelope known as a serological response.[10] The potential protective effect of naturally acquired type-specific antibodies against subsequent infection with same-type HPV among men is uncertain.[11] A systematic review based on 3 studies, all conducted in high-income countries, suggested that there was no protective effect among men.[12,13] However, more data are needed, particularly in low- to middle-income countries.[12] A better understanding of HPV type-specific serological responses to natural infection in MLHIV and the risk of subsequent infection is important in guiding targeted HPV control efforts such as vaccination. This information, especially if gathered before widespread vaccine introduction, could help improve our understanding of the HPV serological dynamics among this group and be useful for monitoring vaccine effectiveness and understanding of the impact of vaccination programs among vulnerable populations over time. It could also help account for any protection (if at all) after natural infection when modeling future impacts of the HPV vaccination programs.[14]

To address these knowledge gaps, we followed a cohort of predominately heterosexual MLHIV in South Africa. We estimated the HPV seroprevalence and concordance with same-type DNA at enrollment in the cohort, evaluated HIV and HPV virological factors associated with HPV seroconversion at follow-up 18 months later, and explored the risk of HPV DNA incident infection among men seropositive for the same type at enrollment.

METHODS

Study Design, Population, and Data Collection

This cohort study has been described previously.[15] Briefly, 304 HIV-1–seropositive men (MLHIV) aged 18 years or older who reported sexual activity in the past 3 months were recruited from ART clinics in inner-city Johannesburg, South Africa. After enrollment, participants were followed up every 6 months for up to 18 months. An interviewer-administered questionnaire was used to collect data on sociodemographic, behavioral, and clinical characteristics. Furthermore, to optimize privacy, participants completed sensitive questions on sexual behavior using a computer-assisted self-interview.

Sample Collection

At each visit, venous blood was collected to test for $CD4^+$ cell count (FACScount, BD; BD Biosciences, San Jose, CA); HIV-1 plasma viral load (PVL) using Roche Taqman (Roche Diagnostics, Mannheim, Germany) and HPV serology. A genital sample for HPV DNA testing was collected by rubbing a cotton swab around the glans penis, coronal sulcus, and ventral surface of the penis as previously described.[15] An anal sample for HPV DNA testing was collected by blindly (ie, without anoscopy) inserting a Dacron swab 3 centimeters into the anal canal and removing it while rotating and applying pressure on the walls of the canal. Swabs were stored at −70°C before HPV DNA testing.

Laboratory Testing

HPV detection and genotyping were performed using an identical method at enrollment and at last follow-up visit. The MagNA Pure LC DNA Isolation Kit I (Roche Diagnostics) was used to extract HPV DNA from the swabs. HPV genotype distributions were then assessed by the Roche Linear Array assay (RLA; Roche Diagnostics). HPV16 and HPV18 genital viral loads (VL, copies per million human cells) were quantified at enrollment on samples that were positive for these types using quantitative duplex real-time PCR method.[16] Genital and anal samples were tested separately. This method allows the HPV16, HPV18, and albumin gene copy number to be quantified in the same assay. The human β-globin gene served as an internal control for cellular adequacy and extraction efficiency. For the purposes of this analysis, HPV DNA positivity was limited to the 15 types covered by the serology assay (HPV6/11/16/18/31/33/35/39/45/52/56/58/59/68/73) with HR-HPV being similarly defined with the following 12 types: HPV16/18/31/33/35/39/45/52/56/58/59/68.[17]

HPV antibodies were detected using a multiplexed binding assay and classified as seropositive or seronegative based on the preassigned cutoff. This uses pseudovirions (PsV) as antigens and detects HPV type-specific immunoglobulin G antibodies (PsV-Luminex). Serology was performed to detect the following HR types HPV16/18/31/33/35/39/45/52/56/58/59/68 and the low-risk (LR) types HPV6/11/73 at Karolinska Institute, Stockholm, Sweden.[18,19] Serum samples were analyzed in 1:50 and 1:150 dilutions. Seropositivity cutoff values were independently determined for each HPV type by analyzing the mean fluorescence intensity unit (MFI) values obtained from a panel of 100 children's sera (≤12 years old and presumed virgins). The cutoff algorithm was as recommended by the global HPV LabNet (mean MFI value of a negative control serum panel plus 3 SDs).[20] In cases where the cutoff value was unexpectedly low (ie, <400 MFI), 400 MFI was used as cutoff to have a sensitivity and specificity similar to classical HPV ELISAs.[21]

Definition of HPV DNA and Serology Status

HPV DNA Status

Genital swab results were used to determine the HPV DNA status. There were more genital samples (304) as only 250 (82%) men accepted anal swabbing. Men were

considered "HPV DNA positive" if positive by RLA for any of the HPV types included in the serology assay, and "HPV DNA negative" if negative for all these types. HPV DNA genotype-specific persistence was defined as being positive for the same type at enrollment and 18-month visit. Type-specific clearance was defined as being positive for a specific type at enrollment and negative for that type at 18-month visit.

HPV Serology Status

Overall and type-specific or group (ie, vaccine targets) HPV seroprevalences were defined as being seropositive for any/type-specific/grouped types, respectively. HPV type-specific seroconversion was defined as being HPV DNA positive and same-type seronegative at enrollment that became same-type seropositive at 18-month visit, regardless of the HPV DNA status at 18-month visit. We also computed type-specific seropersistence (having same-type detectable antibody at enrollment and 18-month visit) among all men who were seropositive for any HPV type at enrollment, and calculated seroincidence (seronegative for a specific HPV type at enrollment and seropositive for the same type at 18-month visit) among all men who were seronegative at enrollment, irrespective of their HPV DNA status.

Statistical Analysis

To compare HPV seropositivity among type-specific HPV DNA positive and DNA negative MLHIV at enrollment, prevalence ratios (PRs) were obtained from logistic regression using marginal standardization and the accompanying 95% confidence intervals (CI) were estimated using the delta method.[22] Factors associated with HPV seroconversion were explored using generalized estimating equations to account for seropositivity by multiple types within MLHIV and presented as adjusted odds ratios (OR) and their 95% CI.[23] To explore HIV-related factors associated with HPV seroconversion, preplanned analyses included stratification by ART status (ART or no ART), ART duration ($\leq$1 or $>$1 year) and $CD4^+$ cell counts/μL at enrollment ($<$200, 201–350, 351–500, $>$500), and HIV-1 PVL (plasma HIV-1 RNA $<$40 or $\geq$40 copies/mL) at enrollment. Stable high $CD4^+$ count was defined as having $CD4^+$ counts $>$500 cells/μL at all 3 follow-up visits. Sustained HIV virological control was defined as HIV-1 PVL $<$40 copies/mL at all the 3 follow-up visits. Controlled disease status was defined as being on ART for $>$6 months, $CD4^+$ $>$350 cells/μL, and HIV-1 RNA $<$40 copies/mL. High HPV VL was defined as viral load $>$ 5.3 $\log_{10}/10^6$ cells as previously described.[16] Multivariable analyses were adjusted for sociodemographic and behavioral factors which were associated with HPV seroconversion at bivariate analysis at $P < 0.10$.[24] Risk ratios (RRs) were calculated to explore the association between HPV seropositivity at enrollment and HPV DNA infection incidence at 18-month follow-up. Data were analyzed using Stata version 15 (Stata Statistical Software; Stata Corporation, College Station, TX).

Ethics Statement

Ethical approval for the study was obtained from the Wits Human Research Ethics Committee (reference numbers: M111191 and M160859). All study participants provided written, informed consent after explanation of the study objectives and testing procedures.

RESULTS

Study Population

A detailed description of the cohort is provided elsewhere.[15] Of the 304 MLHIV enrolled, the median age at enrollment was 38 years (IQR: 22–59), 25% reported $>$1 sexual partner in the past 3 months, and 5% reported ever having sex with other men. At enrollment, most participants (65%) were on ART, with median $CD4^+$ count of 445 cells/μL (IQR: 328–567), and 54% were virologically suppressed (plasma HIV RNA $<$40 copies/mL).

HPV Seroprevalence at Enrollment

Enrollment HPV serology results were available for 99% (303/304) of the enrolled men. Seroprevalence of any HPV type was 66% (199/303), of whom 67% (134/199) were seropositive for multiple (ie, $>$1) types. The seroprevalence of any 12 HR-HPV types was 61%. For the HR types, seroprevalence was highest for HPV58 (30%), followed by HPV52 (27%) and HPV35 (18%). Seropositivity for any HPV types of the bivalent (HPV16/18), quadrivalent (HPV6/11/16/18), and nonavalent (HPV6/11/16/18/31/33/45/52/58) vaccine types was 19%, 37%, and 60%, respectively. No participants were seropositive for all HPV types included in the nonavalent vaccine (Fig. 1).

Relationship Between HPV DNA and HPV Antibody-Specific Prevalence at Enrollment

Of the 184/257 (71%) men who were HPV DNA positive for any of the 15 HPV types at enrollment ("HPV DNA positive"), 125 (68%) were seropositive for the same HPV type at enrollment (ranging by type from 0% for HPV73 (0/12) to 50% for HPV31 (3/6); Table 1). The type-specific HPV seroprevalence was significantly higher among those with same-type HPV DNA positive compared with same-type DNA negative for the HR types HPV31 after adjusting for employment status and age at sexual debut [adjusted prevalence ratio (aPR) = 4.31; 95% CI: 1.80 to 10.31], HPV45 (aPR = 3.68; 95% CI: 1.07 to 12.68), HPV59 (aPR = 2.29; 95% CI: 1.86 to 6.10), and LR types HPV6 (aPR = 2.75; 95% CI: 1.65 to 4.57) and HPV11 (aPR = 3.73; 95% CI: 1.90 to 7.32; Table 1).

HPV Seroconversion and Association With DNA Persistence and Clearance

A total of 59 MLHIV were HPV DNA positive and same-type seronegative at enrollment. Among these 59, 13 (22%) seroconverted for their type(s), giving a total of 18

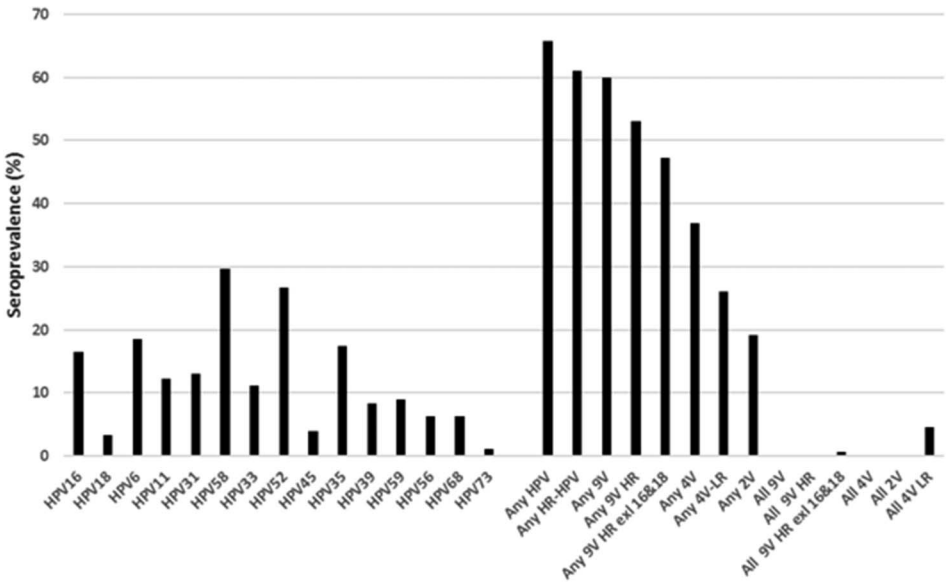

FIGURE 1. HPV seroprevalence* any HPV type prevalence defined as positive for at least one HPV type (6/11/16/18/31/33/35/39/45/52/56/58/59/68/73) at enrollment; any HR-HPV type prevalence defined as positive for at least one HR-HPV type (16/18/31/33/35/39/45/52/56/58/59/68) at baseline; any 9V = positive for any of HPV6/11/16/18/31/33/45/52/58 (nonavalent HPV vaccine); any 9V-HR = positive for any of HPV16/18/31/33/45/52/58; any 4V = positive for any of HPV6/11/16/18 (quadrivalent HPV vaccine); any 4V-LR = positive for any of HPV6/11; any 2V = positive for any of HPV16/18 (bivalent HPV vaccine); all 9V = positive for all of HPV6, 11, 16, 18, 31, 33, 45, 52, and 58; all 9V-HR = positive for all of HPV16, 18, 31, 33, 45, 52, and 58; all 4V = positive for all of HPV6, 11, 16, and 18; all 2V = positive for BOTH HPV16 and 18; all 4V-LR = positive for BOTH HPV 6 and 11.

seroconversions events (5 men had >1 type-specific seroconversion event). Type-specific seroconversion was highest for HPV58 (29%) and HPV16 (22%) while there were no seroconversion events for the HR types 31, 33, and 45. Overall, type-specific seroconversion was more frequent with same-type cleared DNA infection compared with persistent infection (any HPV type seroconversion: 33% versus 27%; *P* value = 0.03; Table 2).

HIV-Related Factors Associated With HPV Seroconversion

The risk of seroconversion was higher among MLHIV with detectable HIV PVL at enrollment compared to those with undetectable HIV PVL after adjusting for employment status and age at sexual debut [adjusted odds ratio (aOR) = 2.78, 95% CI: 1.12 to 6.77, *P* value = 0.03]. Similarly, there was a higher risk of seroconversion among men who had high enrollment HPV18 viral load (≤5.3 $\log_{10}$ vs. >5.3 $\log_{10}$/10^6cells; aOR = 3.32, 95% CI: 1.42 to 7.74, *P* value = 0.01). There was marginal evidence that MLHIV who were less than a year on ART were more likely to seroconvert compared with those at least one year on ART (aOR = 6.21, 95% CI: 0.76 to 50.8, *P* value = 0.09). $CD4^+$ cell count, clearance of genital HPV infection at 18 months, and anal HPV infection status at enrollment were not independently associated with seroconversion (Table 3).

Incident HPV DNA Infection, Seroincidence, and Seropersistence Over 18 Months

We further evaluated the impact of baseline seroprevalence status on incident HPV DNA infections during 18-month follow-up. There were only 6 incident genital HPV DNA infections among the 119 MLHIV who were negative for any of the 15 HPV DNA infection types at enrollment. These were distributed as 4 infections among 74 men who were seropositive for any of the 15 types and 2 infections among the 45 men seronegative for all the 15 types. There was no significant difference in incidence of HPV DNA infection between these 2 groups (risk ratio of incident infection among those HPV seropositive at baseline compared with those seronegative was RR = 1.21, 95% CI: 0.23 to 6.30, *P* value = 0.59; see Supplementary Table 1, Supplemental Digital Content, http://links.lww.com/QAI/B444). Among the 83 men who were seronegative for any HPV type at enrollment, 28 (34%) had seroincidence (seronegative for a specific HPV type at enrollment and seropositive for the same type at 18-month visit). A total of 148 (89%) among the 167 men with detectable antibodies at enrollment had seropersistence (having same-type detectable antibody at enrollment and 18-month visit) (see Supplementary Table 2, Supplemental Digital Content, http://links.lww.com/QAI/B444).

DISCUSSION

In this cohort study of predominantly heterosexual African MLHIV, we report an overall HPV seroprevalence of 66%, and 60% for the nonavalent vaccine types. Although there are difficulties in directly comparing these findings with previous studies due to differences in the methods, serotypes assessed, sexual behavior, and HIV status, these levels are higher than in most previous studies. For instance, the seroprevalence of quadrivalent vaccines types of 37% that we found is much higher than 13% and 21% reported among HIV-negative men in the United States.[25,26] Similarly, the seroprevalence of nonavalent vaccines types is 3-fold higher than the 20% reported

TABLE 1. Comparison of HPV Seropositivity Among Type-Specific HPV DNA Positive and DNA Negative MLHIV (N = 257) at Enrollment

	DNA Positive, (N)	DNA and Antibody Positive, (n)	DNA and Antibody Positive/DNA Positive (n/N), (%)	DNA Negative, (N)	DNA Negative and Antibody Positive, (n)	DNA Negative and Antibody Positive/ DNA Negative (n/N), (%)	aPR (95% CI)*
Bivalent /quadrivalent types							
HPV6	34	14	41.2	223	34	15.3	2.75 (1.65 to 4.57)
HPV11	18	8	44.4	239	27	11.3	3.73 (1.90 to 7.32)
HPV16†	28	5	17.9	229	39	17.0	1.09 (0.47 to 2.54)
HPV18†	20	1	5.0	237	8	3.4	1.64 (0.22 to 12.47)
Additional nonavalent types							
HPV31†	6	3	50.0	251	30	12.0	4.31 (1.80 to 10.31)
HPV33†	11	3	27.3	246	27	11.0	2.54 (0.90 to 7.14)
HPV45†	20	3	25.0	237	9	3.8	3.68 (1.07 to 12.68)
HPV52†	17	6	35.3	240	63	26.3	1.34 (0.68 to 2.65)
HPV58†	20	6	30.0	237	68	28.7	0.91 (0.42 to 1.99)
Nonvaccine types							
HPV35†	32	5	15.6	225	39	17.3	0.92 (0.39 to 2.16)
HPV39†	12	1	8.3	245	20	8.2	0.89 (0.13 to 6.26)
HPV56†	10	1	10.0	247	14	5.7	1.65 (0.24 to 11.56)
HPV59†	21	5	23.8	236	20	8.5	2.29 (1.86 to 6.10)
HPV68†	20	3	15.0	237	14	5.9	2.27 (0.83 to 8.79)
HPV73	12	0	0.0	245	4	1.6	—
Any HPV	184	125	67.9	73	45	62.2	1.08 (0.91 to 1.29)
Any HR-HPV†	117	78	66.7	140	80	57.5	1.16 (0.97 to 1.40)

*Adjusted prevalence ratio [PR] from logistic regression using marginal standardization for type-specific seroprevalence if same-type DNA positive compared with DNA negative, adjusted for employment status and age at sexual debut.

†Denotes high-risk (HR) HPV types.

among HIV-negative men in the United States, but similar to the 61% reported among HIV-positive men who have sex with men in Amsterdam, the Netherlands.[11,27]

Our analysis showed that almost 70% of men with HPV DNA infection at baseline were seropositive. This is keeping with previous reports that L1 seroconversion does not always occur after sexually transmitted HPV infection.[28,29] Although the reason for this observation is unclear, it may be because the humoral response against HPV L1 capsid protein is very weak, thus unmeasurable with the current serological tests. Previous studies have reported that men have a considerably lower HPV seroprevalence than women,[30,31] and a recent study suggested that HPV-seropositive women may have higher antibody levels than HPV-seropositive men.[32] This may be because HPV infections in women have more access to the mucosal immune system than HPV infections on the keratinized surface of the male genitals.[12]

We found that HPV seroconversion was associated with detectable HIV PVL and higher genital HPV viral load. This

TABLE 2. Type-Specific Seroconversion at 18 Months Among 59 MLHIV Who Were DNA Positive and Seronegative at Enrollment

	All Participants*		Participants With HPV DNA Persistence†		Participants With HPV DNA Clearance‡	
	DNA Positive and Antibody Negative at Enrollment	Seroconversion Events	DNA Positive and Antibody Negative at Enrollment	Seroconversion Events	DNA Positive and Antibody Negative at Enrollment	Seroconversion Events
	N Infections	n (%)*	N Infections	n (%)†	N Infections	n (%)‡
Bivalent/ quadrivalent						
HPV6	20	1 (1.5)	2	0 (0.0)	18	1 (5.6)
HPV11	10	2 (20.0)	1	0 (0.0)	9	2 (22.2)
HPV16§	23	5 (21.7)	7	2 (28.6)	16	3 (18.8)
HPV18§	19	2 (10.5)	0	0 (0.0)	19	2 (10.5)
Additional nonavalent						
HPV31§	3	0 (0.0)	0	0 (0.0)	3	0 (0.0)
HPV33§	8	0 (0.0)	1	0 (0.0)	7	0 (0.0)
HPV45§	17	0 (0.0)	1	0 (0.0)	16	0 (0.0)
HPV52§	11	1 (9.1)	1	0 (0.0)	10	1 (10.0)
HPV58§	14	4 (28.6)	0	0 (0.0)	14	4 (30.8)
Nonvaccine HR-HPV						
HPV35§	27	2 (7.4)	1	0 (0.0)	26	2 (7.8)
HPV39§	11	0 (0.0)	2	0 (0.0)	9	0 (0.0)
HPV56§	9	0 (0.0)	0	0 (0.0)	9	0 (0.0)
HPV59§	16	0 (0.0)	5	0 (0.0)	11	0 (0.0)
HPV68§	17	0 (0.0)	2	0 (0.0)	15	0 (0.0)
Nonvaccine LR-HPV						
HPV73	12	1 (8.3)	4	0 (0.0)	8	1 (12.5)
Any HPV	59	13 (22.2)	**26**	2 (7.7)	**33**	11 (33.3)
Any HR-HPV§	39	11 (28.2)	**6**	2 (33.3)	**33**	9 (27.3)

*Seroconversion calculated among 59 men with DNA positive and antibody negative status at enrollment.
†Seroconversion calculated among men with DNA positive and antibody negative at baseline and with type-specific persistence at 18 months.
‡Seroconversion calculated among men with DNA positive and antibody negative at baseline and type-specific clearance at 18 months.
§High-risk (HR) HPV types (NOTE: the total is not the sum of individual infections, but only infected men, some having >1 infection).

is probably related to the fact that a higher HPV viral load is required to induce a measurable type-specific serum antibody response against epitopes of the HPV L1 capsid protein. This is also aligned with our finding that HPV seroconversion was more common among MLHIV who had cleared HPV DNA, perhaps indicating the important role of a competent immune system in seroconversion and the subsequent clearance of HPV infection.

Our results show that HPV seropositivity at baseline was not protective of type of incident infection at 18 months. Although this finding need to be interpreted with caution owing to the small sample size, it is consistent with the conclusions of a systematic review that naturally acquired HPV antibodies are not protective of subsequent HPV infections among men.[12] The reasons for this are not fully understood. However, it has been hypothesized that the observed HPV DNA infections are probably a combination of newly acquired, reactivated latent and transient infections.[33] Although the currently available commercial PCR-based assay methods cannot distinguish between a potential acquisition versus a reactivation, new evidence supports the hypothesis that natural immunity protects against new acquisition but not reactivation.[34] Protection against new HPV infections is believed to be largely antibody mediated immunity, whereas control of existing infections is probably more cell-mediated. It is therefore plausible that protection from serum antibodies is only a surrogate marker for other local mechanisms that may still need further elucidation.[12]

This cohort study was limited by the fact that HPV serology and HPV DNA were only tested at 2 time points (enrollment and 18 months). It is therefore difficult to determine whether the specific seroconversion events occurred in response to either an infection which persisted during follow-up or a prevalent HPV infection at enrollment which was cleared followed by same-type reinfection or reactivation of a latent infection (ie, transient infections). Similarly, when evaluating the risk of reinfection according to same-type seropositivity at baseline, we cannot be certain if the HPV DNA infection detected at 18-month follow-up was

TABLE 3. HIV and HPV-Related Correlates of HPV Seroconversion at 18 Months Using 18 Events of DNA Positive/Same-Type Seronegative at Enrollment

	N = 202 n (%)	18 Seroconversion Events n (%)	Crude OR (95% CI)	*P*	Adjusted* aOR (95% CI)	*P*
ART status at enrollment						
No ART	76 (37)	8 (10.5)	1		1	
ART	126 (63)	10 (7.9)	0.75 (0.33 to 1.71)	0.49	0.91 (0.34 to 2.34)	0.83
Duration on ART at enrollment (mo)						
>12	80 (40)	3 (3.8)	1		1	
6–12	25 (12)	4 (16.0)	4.62 (0.70 to 30.47)	0.11	6.21 (0.76 to 50.8)	0.09
<6	14 (7)	0 (0.0)	—	—	—	—
Enrollment $CD4^+$ count (cells/μL)						
>500	60 (30)	2 (11.6)	1		1	
351–500	70 (35)	4 (5.7)	0.43 (0.14 to 1.39)	0.16	0.50 (0.14 to 1.83)	0.29
201–350	29 (14)	3 (10.3)	0.84 (0.17 to 4.12)	0.84	0.99 (0.22 to 4.58)	0.99
<200	33 (16)	3 (9.1)	0.76 (0.22 to 2.69)	0.67	1.09 (0.27 to 4.44)	0.91
High stable $CD4^+$ count†	42 (21)	5 (11.9)	1.60 (0.84 to 3.03)	0.15	1.42 (0.74 to 2.78)	0.29
Detectable HIV PVL at enrollment‡	111 (55)	13 (11.7)	2.60 (1.05 to 6.46)	0.04	2.78 (1.12 to 6.77)	0.03
Sustained HIV virological control§	22 (11)	1 (4.6)	0.48 (0.05 to 4.13)	0.50	0.76 (0.10 to 5.88)	0.79
Disease control status at enrollment‖						
ART naive	76 (38)	8 (10.5)	1		1	
Poorly controlled	79 (39)	9 (11.4)	1.16 (0.46 to 2.90)	0.78	1.72 (0.62 to 4.74)	0.30
Well controlled	43 (21)	0 (0.0)	—	—	—	—
High genital HPV viral load¶						
HPV16 viral load	24 (12)	5 (20.8)	2.65 (0.69 to 10.16)	0.16	2.67 (0.45 to 15.55)	0.29
HPV18 viral load	10 (5)	2 (20.0)	1.93 (0.58 to 6.50)	0.28	3.32 (1.42 to 7.74)	0.01
Anal HPV infection	88 (44)	10 (11.4)	1.81 (0.74 to 4.46)	0.20	1.41 (0.54 to 3.70)	0.49
Cleared genital HPV infection	104 (51)	10 (9.6)	1.12 (0.26 to 4.89)	0.88	1.16 (0.26 to 5.17)	0.85
Anogenital warts	48 (23)	3 (6.3)	0.63 (0.28 to 1.42)	0.26	0.68 (0.29 to 1.49)	0.23

*Adjusted odds ratio (aOR): associations adjusted for age at sexual debut and employment status, from generalized estimating equations.
†$CD4^+$ count was > 500 cells/μL for all the 3 follow-up visits.
‡HIV PVL ≥ 40 copies/mL.
§HIV PVL was undetectable (<40 copies/mL) for all the follow-up visits.
‖Controlled disease was defined as on ART for >6 months, $CD4^+$ >350 cells/μL and undetectable PVL.
¶HPV genital viral load > 5.3 $\log_{10}/10^6$ cells.

a new acquisition or a reactivation of a latent infection. Moreover, the low incidence of HPV DNA infections made exploring the impact naturally acquired antibodies have on incident infections challenging, and thus, the results presented need to be interpreted with caution. Despite these limitations, our study also has several strengths which include its longitudinal design, the availability of serology and HPV DNA genotyping data of 15 serotypes at both enrollment and 18 months. This is also the first study to explore HPV seroepidemiology using longitudinal data among MLHIV in Africa.

CONCLUSIONS

Overall seropositivity and exposure to nonavalent HPV types was high among MLHIV. The lack of protection from naturally acquired infection supports the hypothesis that HPV vaccination of boys before they become sexually active could reduce the burden of HPV infection and related disease among this population. The high seroprevalence of the nonavalent types supports the need to make this multivalent vaccine more widely available.

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