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Risk factors for anal HPV infection and anal precancer in HIV-infected men who have sex with men

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Abstract

Background: Carcinogenic human papillomaviruses (HPV) cause a large proportion of anal cancers. HIV-infected men who have sex with men (MSM) are at increased risk of HPV infection and anal cancer compared to HIV-negative men. We evaluated risk factors for HPV infection and anal precancer in a population of HIV-infected MSM.

Methods: Our study included 305 MSM at an HIV/AIDS clinic in the Kaiser Permanente Northern California Health Maintenance Organization. Logistic regression was used to estimate associations of risk factors comparing (1) men without anal HPV infection, (2) men with anal HPV infection, but no precancer, and (3) men with anal precancer.

Results: Low CD4 count (<350 cells/mm^3) and previous Chlamydia infection were associated with an increased risk of carcinogenic HPV infection (OR 3.65 95%CI 1.28 – 10.40, OR 4.24 95%CI 1.16 – 15.51, respectively). History of smoking (OR 2.71 95%CI 1.43 – 5.14), duration, recency, and dose of smoking, increased the risk of anal precancer among carcinogenic HPV-positive men, but had no association with HPV infection.

Conclusions: We found distinct risk factors for anal HPV infection and anal precancer. Risk factors for HPV infection and anal precancer are similar to established risk factors for cervical cancer progression.

Introduction

Anal cancer is relatively uncommon in the United States, with an expected 6,000 new cases in 2012 and an overall annual incidence rate of 2/100,000[1-2]. Most anal cancers are caused by human papillomavirus (HPV) infection [3]. Certain populations such as human immunodeficiency virus (HIV)-infected and other immunosuppressed individuals appear to have an increased risk of both human papillomavirus (HPV) infection and anal cancer. A recent world-wide meta-analysis reported that HIV-infected men who have sex with men (MSM) have an estimated 46/100,000 incident anal cancer cases per year compared to HIV-negative MSM who have 5/100,000 incident cases per year [4]. Since the advent of highly active antiretroviral therapy (HAART), the life expectancy for HIV-infected individuals has significantly increased. This may have allowed time for progression of carcinogenic HPV infections to anal cancer and possibly explaining an increase in anal cancer incidence in the US [5-8].

Anal cancer natural history presumably follows the same steps as cervical cancer natural history: HPV acquisition, HPV persistence, progression of persistent HPV infections to high-grade anal intraepithelial neoplasia (AIN), and invasion to anal cancer [5]. Studies have identified risk factors for each stage in the progression from cervical HPV infection to cervical cancer [9-11], and we are beginning to find similar risk factors in the anal cancer natural history. Risk factors for anal HPV infection are related to sexual behavior such as lifetime number of sexual partners, as well as low CD4 count in HIV-infected populations [12-15]. Smoking history has been shown to be associated with anal HPV persistence [16-17] and anal cancer [18-19], but it is unclear at what stage of the natural history smoking acts. To add to our understanding of risk factors that influence the natural history of HPV and anal cancer, we conducted a study of lifestyle

characteristics and clinical parameters for HIV-infected MSM undergoing routine anal cancer screening at Kaiser Permanente Northern California (KPNC) Health Maintenance Organization.

Methods

Study population

The study was based at the San Francisco KPNC Anal Cancer Screening Clinic. Using the Kaiser HIV registry, we identified men 18 years or older, who were not diagnosed with anal cancer prior to enrollment and provided informed consent as eligible for the study. The study was reviewed and approved by both institutional review boards at KPNC and at the National Cancer Institute (NCI). In total, 363 men were enrolled between August 2009 and June 2010. To collect risk factor information, participants completed a self-administered questionnaire. Additional information on HIV status and medication, sexually transmitted diseases, and histopathology results were abstracted from the KPNC clinical database. 271 had no high-grade AIN detected at the enrollment visit but we were able to obtain follow-up information from 86 of these subjects from additional clinic visits up to December 2011. This follow-up information was included in the analysis to improve ascertainment of prevalent disease as anoscopy has less than perfect sensitivity. We excluded 54 participants with no questionnaire data and 4 men with invalid HPV DNA resulting in a final population of 305 men.

Cytology, anoscopy, histology, and HPV detection

The clinical procedures used in this study have been previously described [20-23]. In brief, during the clinical examination, two cytology specimens were collected by inserting a wetted swab into the anal canal up to the distal rectal vault and withdrawing with rotation and lateral pressure. A third anal specimen was collected for routine *Chlamydia Trachomatis* and *Neisseria*

Gonorrhea testing, using the Gen-Probe® nucleic acid amplification testing. After specimen collection, participants received a digital anorectal exam followed by high resolution anoscopy (HRA). At most two suspicious-appearing lesions identified during HRA were biopsied and sent for routine histopathological evaluation. From the first specimen, a ThinPrep slide was prepared for routine Pap staining; cytology results were reported using a modification of the Bethesda System classification for cervical cytology [24], using the categories NILM (Negative for intraepithelial lesion or malignancy), ASC-US (atypical squamous cell of undetermined significance), ASC-H (atypical squamous cells-cannot exclude high grade lesion), LSIL (low-grade squamous intraepithelial lesion), HSIL-AIN2 (high grade squamous intraepithelial lesions-AIN2) and HSIL-AIN3. Histology results were reported as negative, condyloma acuminata, and anal intraepithelial neoplasia (AIN) grades 1-3. HPV DNA testing was performed using the Cobas 4800 HPV test (Roche, Pleasanton, CA, USA) blinded to all study data as previously described [25].

Statistical analysis

Risk factor associations at different stages of anal carcinogenesis were estimated using combined endpoints based on cytology, histology and HPV testing: (i) <AIN2, HR-HPV-negative: No high-grade AIN, including men with no biopsy, non-dysplastic biopsy or AIN1 histology, <HSIL cytology and HR-HPV-negative; (ii) <AIN2, HR-HPV-positive: No high-grade AIN, including men with no biopsy, non-dysplastic biopsy or AIN1 histology, <HSIL cytology and HR-HPV-positive; (iii) AIN2+: AIN2 and AIN3, including men with AIN2 or AIN3 histology or with lower grade, normal, or no histology and with HSIL-AIN2 or HSIL-AIN3 cytology (precancer).

Univariate analyses, adjusted for age at enrollment, were conducted to compare a) AIN2+ vs. <AIN2, HR-HPV-negative, b) <AIN2, HR-HPV-positive vs. <AIN2, HR-HPV-negative, and c) AIN2+ vs. <AIN2, HR-HPV-positive for each of the lifestyle characteristics asked in the questionnaire. Odds ratios and 95% confidence intervals were estimated using logistic regression models and only variables that were significant ($p<0.05$) from univariate analyses were included in the multivariate models. We conducted a sensitivity analysis and raised the p-value to 0.10 from the univariate models as a cutoff to include in the multivariate models, but this did not affect the main findings (data not shown).

We report estimates from multivariate logistic regression models for comparisons b) and c) from above, adjusting for age at enrollment, ethnicity (non-Hispanic, Hispanic), CD4 count (<350, ≥350 cells/mm^3), number of lifetime male partners (<5, ≥5), history of Chlamydia at any site (no, yes) and smoking status (never, ever). We then further examined specific smoking variables including smoking in the last 12 months (no, yes), number of years smoked (non-smoker, ≤10 years, >10 years) and cigarette packs smoked per day (non-smoker, ≤1/2 packs, >1 packs). If models were unstable due to low numbers, variables were excluded from the model. We also evaluated the associations using a polytomous regression model which gave similar results. All statistical tests were two-sided and considered to be statistically significant at $p<0.05$. Analyses were conducted using SAS version 9.2 (SAS Institute, Cary, NC).

Results

Table 1 shows the distribution of population characteristics of the 305 HIV-infected MSM included in this analysis across the three disease endpoints and results from age-adjusted univariate analyses of individual risk factors. Overall, the population was largely white and non-

Hispanic with a median (interquartile range) age of 53 (47-60) years at enrollment. All participants were HIV-infected with 94% of men using HAART, 81% of men with CD4 counts ≥350 cells/mm^3, and 91% of men with HIV viral load ≤75 copies. We found 21.0% (n=64) were <AIN2, HR-HPV-negative, 49.8% (n=152) were <AIN2, HR-HPV-positive, and 29.2% had (n=89) anal precancer (AIN2+). All men with high grade anal lesions were HR-HPV-positive and no anal cancer cases were identified in this population. Men with AIN2+ had a higher number of lifetime male partners. HPV-positive men, regardless of histology and cytology, were more likely to have a previous Chlamydia infection. AIN2+ men also had more lifetime smoking exposure with a higher number of years smoking and more cigarette packs smoked per day.

When comparing men with anal precancer (AIN2+) to <AIN2, HR-HPV-negative men, low CD4 count (<350 cells/mm^3) (OR 3.26 95%CI 1.22-8.74), a higher number of lifetime male partners (≥5, OR 2.49 95%CI 1.12-5.58), history of Chlamydia (OR 4.46 95%CI 1.23-16.18), and all smoking variables (ever smoker, smoked within the last 12 months, total number of smoking years, and cigarette packs smoked per day), were all significantly associated with an increased risk of anal precancer. When comparing <AIN2, HR-HPV-positive men to <AIN2, HR-HPV-negative men, history of Chlamydia was associated with an increased risk of <AIN2, HR-HPV-positive (OR 3.96 95%CI 1.13-13.90). When comparing men with anal precancer to <AIN2, HR-HPV-positive men, being Hispanic decreased risk of anal precancer (OR 0.35 95%CI 0.13-0.97), whereas all smoking variables were significantly associated with an increase in anal precancer.

In the multivariate models we included age at enrollment, ethnicity, CD4 count, number of lifetime male partners, history of Chlamydia and smoking status (Table 2). After adjusting for all these factors, ethnicity and number of lifetime partners were no longer statistically significant in both of the final multivariate models (p-value >.05).

However, when comparing <AIN2, HR-HPV-positive and <AIN2, HR-HPV-negative men, both low CD4 count and previous Chlamydia infection remained statistically significant (OR 3.65 95%CI 1.28 – 10.40, OR 4.24 95%CI 1.16 – 15.51, respectively). When comparing AIN2+ and <AIN2, HR-HPV-positive, smoking remained statistically significant (OR 2.71 95%CI 1.43 – 5.14).

In Table 3 associations for smoking variables, including recency, duration, and intensity of smoking from the multivariate model adjusting for age at enrollment, ethnicity, CD4 count, lifetime number of male partners, and history of Chlamydia infection are presented. Smoking within the past 12 months (OR 3.20 95%CI 1.45-7.09), number of years smoked (>10 yrs, OR 3.09 95%CI 1.33-7.18, p-trend=0.005), and cigarette packs smoked per day (>1 pack, OR 3.50 95%CI 1.19-10.28, p-trend=0.005) were associated with a statistically significant increased risk for anal precancer (AIN2+) compared to <AIN2, HR-HPV-positive men. These factors were not significant when comparing <AIN2, HR-HPV-positive and <AIN2, HR-HPV-negative.

Conclusions

In the present study we assessed characteristics in an HIV-infected MSM population to define co-factors for anal carcinogenic HPV infection and for high grade AIN. Many of the significant risk factors found are similar to established risk factors for cervical HPV infection and cervical precancer.

We confirmed previous findings that low CD4 count (<350 cells/mm^3) was strongly associated with carcinogenic HPV infection [12, 15, 26]. Some studies have reported that number of lifetime sexual partners is a risk factor for anal HPV infection in HIV-infected MSM [13, 27-29] but others have not found this association [30]. While our univariate models found a significantly

increased risk associated with number of lifetime partners, the finding was not significant in the multivariate model. History of Chlamydia infection was associated with anal HPV infection, in both univariate and multivariate analyses. In contrast, history of Gonorrhea, Syphilis, or Herpes was not associated with HPV infection. It has been suggested that Chlamydia may increase persistence of HPV in the cervix, but there is no evidence for a biological interaction [31]. Lifetime sexual partners and history of Chlamydia infection are both indicators of sexual behavior and may just be surrogate markers for increased exposure to HPV.

We found history of ever smoking, smoking in the last 12 months, smoking more than 10 years, and increased cigarette packs smoked per day, to be significantly associated with anal precancer among HR-HPV-positive men. We saw a significant relationship in overall trend between both the number of smoking years and cigarette packs smoked per day and risk of anal precancer. Other studies have found smoking to be a risk factor for anal cancer [18 , 19], but our study is the first to demonstrate that smoking is a co-factor for anal precancer, using rigorous histology-confirmed endpoints. In a prospective study of 247 HIV-infected MSM followed for 3 years for development of anal precancers, smoking at baseline was not associated with increased risk of AIN2 or AIN3 compared to never smokers in univariate analyses [32], but this population was younger and smoking variables did not include duration and intensity. More studies are needed to assess the relationship between smoking and anal precancer, especially with regard to the timing of smoking exposures.

Anal cancer and cervical cancer are initiated in similar epithelial junctions between squamous and glandular tissue [5, 33]. Indicators of sexual behavior such as number of lifetime partners, condom use, history of sexually transmitted infections, and age at sexual debut are highly associated with cervical HPV infection [34]. Analogously, we found that number of lifetime

partners (in univariate analysis) and history of Chlamydia infection was associated with anal HPV infection. Similar to our analysis, low CD4 count has also been identified as a risk factor for cervical HPV infection in HIV-infected women [35-36]. Smoking has been established as a risk factor for cervical precancer [9-11]; in our study of MSM HIV-infected men we also found that smoking status, recency, duration and dose of smoking increased risk of progression to anal precancer among carcinogenic HPV-positive men.

The recognition that co-factors for anal cancer precursors are very similar to those of cervical precancers together with previous observations that the same biomarkers are associated with precancers at both sites [20-22] further corroborate the similarity between CIN3 and AIN3. This could facilitate development of anal cancer early detection efforts, since established tools and approaches from cervical cancer screening can be adapted for a population at risk of anal cancer [37-38]. However, it has been suggested that the risk of invasion is lower for AIN3 compared to CIN3, which affects decisions about expectant management versus immediate treatment of AIN3 [4, 37, 39].

Our study has several strengths including a population highly representative of the HIV-infected MSM community. We had a good assessment of anal histology and cytology [23], high rates of HRA and biopsy, and we performed state-of-the-art HPV testing. A limitation of our study is the cross-sectional study design, which did not allow us to evaluate progression from HPV infection to precancer. We addressed the limited sensitivity of HRA by using a composite histology-cytology endpoint as previously described [21]. In order to best compare anal precancer endpoints to CIN3, we would have preferred to use AIN3 as an endpoint, but the samples size was not sufficient. It is necessary to evaluate AIN3 endpoints in larger studies in the future.

In summary, our results confirm previous studies that have found low CD4 count and indicators of sexual behavior, such as history of Chlamydia, are strong risk factors for anal carcinogenic HPV infection. Using a composite precancer endpoint based on cytology and histology, we determined that several smoking characteristics such as ever smoking, smoking in the last 12 months, lifetime duration of smoking, and cigarettes smoked per day, are risk factors for anal precancer among HPV-positive men. In summary, we demonstrated that risk factors for HPV infection and progression to anal precancer are similar to established risk factors for cervical cancer progression.

Footnote page:

Conflict of Interest Statement: Dr. Castle serves as a paid consultant of BD, Gen-Probe/Hologic, GE Healthcare, and Cepheid; he received a Speaker's honorarium from Roche and he serves as a paid member of a Data and Safety Monitoring Board for HPV Vaccines for Merck. All other authors did not report a conflict of interest.

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Table 1. Among 305 HIV-infected MSM, univariate analyses (adjusted by age), of selected HPV cofactors for three disease endpoints

	Total N=305	<AIN2, HR-HPV-negative (n=64)	<AIN2, HR-HPV-positive (n=152)	AIN2+, HR-HPV-positive (n=89)	AIN2+, HR-HPV-positive vs <AIN2, HR-HPV-negative		<AIN2, HR-HPV-positive vs <AIN2, HR-HPV-negative		AIN2+, HR-HPV-positive vs <AIN2, HR-HPV-positive	
Characteristics	n (%)	n (%)	n (%)	n (%)	OR	95% CI	OR	95% CI	OR	95% CI
Race										
White	260 (89.66)	51 (89.47)	124 (87.32)	81 (93.1)	1.00	Reference	1.00	Reference	1.00	Reference
Non White	30 (10.34)	6 (10.53)	18 (12.68)	6 (6.90)	0.76	(0.23-2.57)	1.22	(0.45-3.30)	0.50	(0.19-1.32)
Ethnicity										
Non Hispanic	251 (88.07)	50 (86.21)	118 (84.89)	79 (94.05)	1.00	Reference	1.00	Reference	1.00	Reference
Hispanic	34 (11.93)	8 (13.79)	21 (15.11)	5 (5.95)	0.37	(0.11-1.23)	1.07	(0.44-2.60)	**0.35**	**(0.13-0.97)**
Education										
High School	50 (16.39)	13 (20.63)	26 (17.33)	10 (11.36)	1.00	Reference	1.00	Reference	1.00	Reference
College	139 (45.57)	27 (42.86)	64 (42.67)	48 (54.55)	2.45	(0.93-6.46)	1.17	(0.52-2.63)	1.99	(0.87-4.51)
Graduate School	116 (38.03)	23 (36.51)	60 (40.00)	30 (34.09)	1.86	(0.68-5.10)	1.42	(0.62-3.29)	1.32	(0.56-3.10)
HIV viral load, copies										
<75	268 (90.54)	59 (93.65)	130 (89.04)	77 (92.77)	1.00	Reference	1.00	Reference	1.00	Reference
≥75	28 (9.46)	4 (6.35)	16 (10.96)	6 (7.23)	0.48	(0.08-3.05)	0.88	(0.21-3.71)	0.57	(0.11-2.90)
CD4 count, cells/mm^3										
≥350	240 (81.08)	57 (90.48)	118 (80.82)	62 (74.70)	1.00	Reference	1.00	Reference	1.00	Reference
<350	56 (18.92)	6 (9.52)	28 (19.18)	21 (25.3)	**3.26**	**(1.22-8.74)**	2.53	(0.97-6.55)	1.39	(0.73-2.67)
Taking HIV medication										
No	19 (6.23)	4 (6.35)	13 (8.67)	2 (2.27)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	286 (93.77)	59 (93.65)	137 (91.33)	86 (97.73)	2.57	(0.45-14.86)	0.71	(0.22-2.29)	4.11	(0.91-18.65)
Age at first anal intercourse										
<20	123 (42.41)	25 (45.45)	62 (42.18)	35 (41.67)	1.00	Reference	1.00	Reference	1.00	Reference
≥20	167 (57.59)	30 (54.55)	85 (57.82)	49 (58.33)	1.31	(0.65-2.65)	1.22	(0.64-2.31)	1.04	(0.60-1.8)
Previous anal cancer screen										
No	116 (37.66)	25 (39.06)	64 (42.11)	26 (29.55)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	192 (62.34)	39 (60.94)	88 (57.89)	62 (70.45)	2.01	(0.97-4.19)	1.02	(0.55-1.90)	1.77	(0.99-3.14)
Previous anal cancer screen time										
Never	116 (39.19)	25 (40.98)	64 (43.24)	26 (31.33)	1.00	Reference	1.00	Reference	1.00	Reference
≤12 months	131 (44.26)	26 (42.62)	60 (40.54)	43 (51.81)	2.16	(0.98-4.80)	1.07	(0.54-2.13)	1.86	(0.99-3.47)
>12 months	49 (16.55)	10 (16.39)	24 (16.22)	14 (16.87)	1.52	(0.56-4.15)	0.97	(0.40-2.32)	1.44	(0.65-3.23)
Previous anal dysplasia										

No	232 (81.98)	52 (85.25)	117 (83.57)	59 (75.64)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	51 (18.02)	9 (14.75)	23 (16.43)	19 (24.36)	1.79	(0.73-4.37)	0.97	(0.41-2.29)	1.68	(0.84-3.36)
Previous anal warts										
No	159 (52.65)	39 (61.9)	76 (51.01)	43 (49.43)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	143 (47.35)	24 (38.1)	73 (48.99)	44 (50.57)	1.55	(0.79-3.03)	1.35	(0.73-2.51)	1.10	(0.64-1.89)
Male Partners, Lifetime										
<5	50 (17.12)	16 (27.12)	26 (17.93)	8 (9.52)	1.00	Reference	1.00	Reference	1.00	Reference
≥5	242 (82.88)	43 (72.88)	119 (82.07)	76 (90.48)	**2.49**	**(1.12-5.58)**	1.45	(0.73-2.88)	1.70	(0.83-3.48)
Male Partners in the Past 6 months										
0	158 (53.74)	36 (61.02)	78 (53.06)	41 (48.81)	1.00	Reference	1.00	Reference	1.00	Reference
≥1	136 (46.26)	23 (38.98)	69 (46.94)	43 (51.19)	1.39	(0.67-2.85)	1.21	(0.64-2.30)	1.23	(0.70-2.17)
Condom Use among anal sex participants										
No	54 (19.57)	11 (19.64)	26 (19.12)	16 (19.75)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	222 (80.43)	45 (80.36)	110 (80.88)	65 (80.25)	1.10	(0.46-2.63)	0.99	(0.45-2.21)	0.97	(0.49-1.95)
Chlamydia										
No	233 (82.92)	55 (94.83)	110 (80.29)	64 (78.05)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	48 (17.08)	3 (5.17)	27 (19.71)	18 (21.95)	**4.46**	**(1.23-16.18)**	**3.96**	**(1.13-13.90)**	1.12	(0.56-2.27)
Gonorrhea										
No	159 (54.08)	32 (52.46)	81 (56.64)	45 (52.33)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	135 (45.92)	29 (47.54)	62 (43.36)	41 (47.67)	1.00	(0.51-1.95)	0.84	(0.46-1.55)	1.20	(0.70-2.05)
Syphilis										
No	214 (75.35)	45 (76.27)	105 (75.54)	60 (73.17)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	70 (24.65)	14 (23.73)	34 (24.46)	22 (26.83)	1.29	(0.59-2.86)	1.10	(0.53-2.28)	1.16	(0.62-2.16)
Herpes										
No	197 (69.37)	41 (68.33)	97 (70.80)	58 (69.88)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	87 (30.63)	19 (31.67)	40 (29.20)	25 (30.12)	0.97	(0.47-2.02)	0.83	(0.43-1.62)	1.06	(0.58-1.94)
Ever Smoker										
No	136 (44.74)	30 (49.18)	78 (51.32)	25 (28.74)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	168 (55.26)	31 (50.82)	74 (48.68)	62 (71.26)	**2.48**	**(1.23-4.98)**	0.98	(0.53-1.79)	**2.65**	**(1.50-4.67)**
Smoked in the last 12 months										
Non smoker	136 (45.18)	30 (50.85)	78 (51.32)	25 (29.07)	1.00	Reference	1.00	Reference	1.00	Reference
No	98 (32.56)	20 (33.90)	46 (30.26)	31 (36.05)	2.04	(0.92-4.51)	1.03	(0.52-2.06)	**2.11**	**(1.10-4.04)**
Yes	67 (22.26)	9 (15.25)	28 (18.42)	30 (34.88)	**3.86**	**(1.53-9.78)**	1.11	(0.46-2.67)	**3.41**	**(1.71-6.80)**
Number of years smoked										
Non smoker	136 (57.63)	30 (66.67)	78 (65.00)	25 (36.76)	1.00	Reference	1.00	Reference	1.00	Reference

≤10 years	35 (14.83)	6 (13.33)	15 (12.50)	14 (20.59)	2.53	(0.83-7.72)	1.02	(0.35-2.91)	**2.88**	**(1.22-6.78)**
>10 years	65 (27.54)	9 (20.00)	27 (22.50)	29 (42.65)	**4.17**	**(1.63-10.66)**	1.16	(0.48-2.81)	**3.43**	**(1.71-6.88)**
Cigarette Packs per Day										
Non smoker	136 (59.39)	30 (66.67)	78 (67.24)	25 (38.46)	1.00	Reference	1.00	Reference	1.00	Reference
≤1/2	63 (27.51)	9 (20.00)	28 (24.14)	26 (40.00)	**3.42**	**(1.34-8.71)**	1.11	(0.46-2.66)	**2.99**	**(1.48-6.05)**
>1	30 (13.1)	6 (13.33)	10 (8.62)	14 (21.54)	**3.24**	**(1.04-10.08)**	0.76	(0.24-2.35)	**4.16**	**(1.63-10.59)**
Alcohol in the last 12 months										
No	51 (16.67)	10 (15.87)	24 (15.79)	16 (18.39)	1.00	Reference	1.00	Reference	1.00	Reference
Yes	255 (83.33)	53 (84.13)	128 (84.21)	71 (81.61)	0.85	(0.35-2.06)	1.01	(0.45-2.29)	0.80	(0.40-1.62)

Combined Disease Endpoints: <AIN2, HR-HPV-negative: No dysplasia and AIN1, including men with no biopsy, non-dysplastic biopsy or AIN1 histology, <HSIL cytology and HR-HPV-negative; <AIN2, HR-HPV-positive: No dysplasia and AIN1, including men with no biopsy, non-dysplastic biopsy or AIN1 histology, <HSIL cytology and HR-HPV-positive; AIN2+, HR-HPV-positive:AIN2 and AIN3, including men with AIN2 or AIN3 histology or with lower grade, normal, or no histology and with HSIL-AIN2 or HSIL-AIN3 cytology. These are all HR-HPV-positive.

Missing values are not included in this table. Mean age (range) for each disease endpoint are 1) <AIN2, HR-HPV-negative: 55.7 (39 - 79), 2) <AIN2, HR-HPV-positive: 52.8 (32 - 78), 3) AIN2+, HR-HPV-positive: 52.9 (37 - 72).

Table 2. Mutivariate logistic regression model of HPV cofactors among HIV-infected MSM*

	<AIN2, HR-HPV-positive vs <AIN2, HR-HPV-negative		AIN2+, HR-HPV-positive vs <AIN2, HR-HPV-positive	
Characteristics	OR	95% CI	OR	95% CI
Ethnicity				
Non Hispanic	1.00	Referent	1.00	Referent
Hispanic	0.88	(0.32-2.43)	0.48	(0.16-1.42)
CD4 count, cells/mm^3				
≥350	1.00	Referent	1.00	Referent
<350	**3.65**	**(1.28-10.40)**	1.40	(0.67-2.93)
Male Partners, Lifetime				
<5	1.00	Referent	1.00	Referent
≥5	1.61	(0.68-3.77)	1.68	(0.71-3.96)
Chlamydia				
No	1.00	Referent	1.00	Referent
Yes	**4.24**	**(1.16-15.51)**	1.15	(0.52-2.57)
Ever Smoker				
No	1.00	Referent	1.00	Referent
Yes	0.89	(0.44-1.78)	**2.71**	**(1.43-5.14)**

*Adjusted for age at enrollment and all other variables in the table.
Combined Disease Endpoints: <AIN2, HR-HPV-negative: No dysplasia and AIN1, including men with no biopsy, non-dysplastic biopsy or AIN1 histology, <HSIL cytology and HR-HPV-negative; <AIN2, HR-HPV-positive: No dysplasia and AIN1, including men with no biopsy, non-dysplastic biopsy or AIN1 histology, <HSIL cytology and HR-HPV-positive; AIN2+, HR-HPV-positive:AIN2 and AIN3, including men with AIN2 or AIN3 histology or with lower grade, normal, or no histology and with HSIL-AIN2 or HSIL-AIN3 cytology. These are all HR-HPV-positive.

Table 3. Multivariate logistic regression of detailed smoking covariates among HIV-infected MSM*

	<AIN2, HR-HPV-positive vs <AIN2, HR-HPV-negative		AIN2+, HR-HPV-positive vs <AIN2, HR-HPV-positive	
Characteristics	OR	95% CI	OR	95% CI
Ever Smoker				
No	1.00	Referent	1.00	Referent
Yes	0.89	(0.44-1.78)	**2.71**	**(1.43-5.14)**
Smoked in the last 12 months				
Non smoker	1.00	Referent	1.00	Referent
No	0.82	(0.38-1.81)	**2.30**	**(1.11-4.8)**
Yes	1.36	(0.47-3.94)	**3.20**	**(1.45-7.09)**
Number of years smoked**				
Non smoker	1.00	Referent	1.00	Referent
≤10 years	1.15	(0.35-3.75)	**3.39**	**(1.29-8.93)**
>10 years	1.44	(0.53-3.93)	**3.09**	**(1.33-7.18)**
p-trend		*0.47*		***0.005***
Cigarette Packs per Day				
Non smoker	1.00	Referent	1.00	Referent
≤1/2	1.00	(0.35-2.86)	**2.90**	**(1.27-6.60)**
>1	0.84	(0.21-3.39)	**3.50**	**(1.19-10.28)**
p-trend		*0.84*		***0.005***

*Adjusted for age at enrollment, ethnicity, CD4 count, number of male partners and history of Chlamydia infection.
** Not adjusted for history of Chlamydia in the first comparison due to missing data.
Combined Disease Endpoints: <AIN2, HR-HPV-negative: No dysplasia and AIN1, including men with no biopsy, non-dysplastic biopsy or AIN1 histology, <HSIL cytology and HR-HPV-negative; <AIN2, HR-HPV-positive: No dysplasia and AIN1, including men with no biopsy, non-dysplastic biopsy or AIN1 histology, <HSIL cytology and HR-HPV-positive; AIN2+, HR-HPV-positive:AIN2 and AIN3, including men with AIN2 or AIN3 histology or with lower grade, normal, or no histology and with HSIL-AIN2 or HSIL-AIN3 cytology. These are all HR-HPV-positive.