The Association between Smoking and Anal Human Papillomavirus in the HPV Infection in Men Study

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ABSTRACT

Background: Previous studies show an association between smoking and anal cancer. The objective of this study was to assess the association between smoking and anal HPV (human papillomavirus) prevalence, incidence, and persistence in men.

Methods: The HPV Infection in Men (HIM) Study is a multinational study that enrolled HIV-negative men. At baseline and follow-up visits, anal specimens were collected. HPV genotyping was assessed by linear array. Prevalence ratios (PR) were used to assess the association between smoking and anal HPV prevalence. Odds ratios (OR) were used to assess the association between smoking and anal HPV incidence and ≥12-months persistence.

Results: Current smokers have a higher prevalence [adjusted PR (aPR), 1.36; 95% confidence interval (CI), 1.06–1.73) and incidence [adjusted OR (aOR), 1.74; 95% CI, 1.26–2.39] and ≥12-months persistence (aOR, 1.67; 95% CI, 1.19–2.33) of any anal HPV compared with never smokers. There were no differences in the prevalence, incidence, or persistence of anal HPV between former and never smokers. Smoking status was not associated with the prevalence or persistence of anal HPV among men who have sex with men but was associated with higher incidence of HR-HPV. Among men that have sex with women (MSW), current smokers had an increased prevalence and incidence of LR-HPV compared with never smokers.

Conclusions: Current smokers had a higher prevalence, persistence, and incidence of HPV compared with never smokers. Further research is needed to assess the role smoking in anal HPV persistence and progression to disease.

Impact: Prevention initiatives should raise awareness about smoking and the risk factor of anal HPV infection and anal cancer.

Introduction

Anal cancer incidence has been shown to be increasing over time in both men and women from high-resource countries (1). People living with HIV, men that have sex with men (MSM), women with prior vulvar cancer, and solid organ transplant recipients are at higher risk for anal cancer compared with the general population (2). In the US, anal cancer incidence has increased 2.7% per year between 2001 and 2015 with increases in both men (2.2%) and women (3.1%; ref. 3). The Center for Disease Control reports that there are 4,700 new cases of human papillomavirus (HPV)–associated anal cancers in women and 2,300 in men each year in the US (4).

Risk factors for anal cancer include HPV infection, tobacco smoking, HIV infection, and a high number of sexual partners (5). Worldwide, 88% of anal carcinoma are caused by high-risk HPV (6) and 79% of those are attributable to HPV16 and HPV18 (1, 7). The attributable fraction of HPV16 to anal cancer is greater in persons without HIV than persons with HIV (1).

Previous studies have shown an association between smoking and cervical and penile HPV infection (8–12). Smoking suppresses the local immune response, increases cellular turnover and proliferation, upregulates proinflammatory cytokines, and can damage host DNA that could increase the likelihood of HPV persisting (13). Studies among women have shown HPV infection is associated with cigarette smoking and a higher HPV16 and HPV18 viral load in current smokers compared with never smokers (8). Smoking at least one pack year of cigarettes was associated with prevalence of anal HPV DNA (14). We have previously shown an increased prevalence of genial HPV among men that smoke (9). The objective of this study was to assess the association between tobacco smoking and the prevalence, incidence, and persistence of anal HPV among men residing in Tampa, Florida, US; Sao Paulo, Brazil; and Cuernavaca, Mexico. Analyses were stratified by sexual orientation given that anal HPV prevalence differs by sexual orientation.

Materials and Methods

Study population

The HPV Infection in Men (HIM) Study recruited 4,123 HIV-negative men between 18 and 70 years old and residing in Tampa, Florida, US; Cuernavaca, Mexico; and Sao Paulo, Brazil from July 2005 to June 2009. We provided a complete report of study methods in previous publications (15). Men returned to clinic every 6 months for up to 10 years. From the initial and subsequent visits, participants were given a physical exam where multiple samples were obtained for laboratory examination and a self-administered questionnaire looking at sociodemographic characteristics, sexual history, tobacco, and alcohol use.

The study team obtained written informed consent from all subjects. Study procedures were authorized by the Institutional Review

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Boards at the University of South Florida (Tampa, FL), the Ludwig Institute for Cancer Research, the Centro de Referencia e Treinamento em Doencas Sexualmente Transmissiveis e AIDS (São Paulo, Brazil), and the Instituto Nacional de Salud Pública (Cuernavaca, Mexico).

Anal specimen collection for HPV detection

At each visit, participants were screened for anal HPV infections. A pre-wetted Dacron swab was used to swab 360 degrees of the anal canal epithelium between the anal opening and the anal canal dentate line. The sample was then placed into a standard transport medium and preserved at −80°C (16). DNA extraction (Qiagen Media Kit), PCR analysis, and HPV genotyping (Roche Linear Array) were conducted on all samples (17). Only samples that tested positive for beta-globin or an HPV genotype were included in the analysis. The linear array assay tests for 37 HPV types, classified as high-risk (HR-HPV; types 16, 18, 31, 33, 35, 39, 45,51, 52, 56, 58, 59, and 68) or low-risk (LR-HPV; types 6, 11, 26, 40, 42, 53, 54, 55, 61, 62, 64, 66, 67, 69, 70, 71, 72, 73,81, 82, 82 subtype IS39, 83, 84, and 89; ref. 18).

Statistical analysis

Participants with missing smoking data ($n = 39$) were excluded. At baseline, participants self-reported the lifetime number of sexual partners and sex was defined as oral, anal, or vaginal sex. On the basis of the participant's responses, sexual orientation was categorized into men who have sex with women (MSW) only and men who have sex with men regardless of having sex with women (MSM). Men that did not report any sexual partners at baseline were classified as unknown for sexual orientation. Tobacco smoking status was categorized as: Current smoker, former smoker, and never smoker. Smoking pack-years were calculated by taking number of packs smoked per year (number of cigarettes per day divided by 20 cigarettes per pack) multiplied by the number of years smoked. The median pack-years smoked were assessed. For current and former smokers separately, tertiles for "number of cigarettes smoked per day," "number of years smoked," and "smoking pack-years" were calculated.

Prevalence

For this analysis, participants who had an anal specimen ($n = 1,994$) at baseline were included. The outcome of interest was prevalence of: any HPV, LR-HPV, HR-HPV, and multiple infections. In this analysis, any HPV refers to the presence of any of the 37 HPV genotypes. Prevalence of any HPV, HR-HPV, and LR-HPV represented having at least one of those HPV types present at the anal canal. Infection with multiple types, defined as two or more HPV genotypes was also assessed compared with persons with one or no HPV infections at baseline. Multiple anal HPV infections have been previously shown to be a risk factor anal pre-cancer among MSM with HIV (19, 20).

To assess the association between HPV prevalence and smoking status, prevalence ratios (PR) and 95% confidence intervals (CI) using Poisson regression with robust variance estimation were calculated. The following are the outcome variables for the models: (i) Any HPV versus No HPV, (ii) HR-HPV versus no HR-HPV, (iii) LR-HPV versus no LR-HPV, (iv) multiple HPV infections versus no HPV/only one HPV infection.

Known potential confounders were selected *a priori* to be used for adjustment in multivariable models. In the overall cohort, PRs were adjusted for age, country, sexual orientation (MSM or MSW) and lifetime female sexual partners. The MSW only model was adjusted for age, country, and lifetime female sexual partners. The MSM only model was adjusted for country, age, lifetime female, and male sexual partners.

Incidence and duration

Participants could elect to have anal specimens collected at each visit. Men with anal HPV specimens collected at follow-up visits were included ($n = 1,427$). The reduction in sample size resulted in the sexual orientation proportions significantly differing between those with baseline specimens (20.3% MSMW and 68.1% MSW) and those with follow-up specimens (27.1% MSMW and 61.6%). Incident infections were defined as new type specific infection that was not present at baseline and was detected at follow-up visits. Incidence of any HPV, HR-HPV, and LR-HPV represented having at least one of those HPV types newly detected at the anal canal. For example, a participant could have HPV16 at baseline and an incident HPV18 detected 6-months later, this participant would have an incident any HPV and HR-HPV. Odds ratios (OR) and 95% CIs were calculated using a multivariable logistic regressions to assess the association between HPV incidence and smoking status. Models were adjusted for baseline known potential confounders that were previously described.

Type-specific HPV infection duration was calculated as the time from prevalent or incident infection detection with observations right-censored at the last positive visit for that infection. HPV clearance is defined as two consecutive negative visits after detection (or a single negative result at the participant's last visit). Persistent infections were defined as having a type-specific infection, prevalent or incident infection, for ≥12 months. Any persistent infections were defined as a participant with ≥1 of 37 HPV types persisting for ≥12 months. Men with no persistent infections for ≥1 of 37 HPV types were defined as non-cases for any HPV. Similar groupings were done for HR-HPV and LR-HPV. We assessed the difference in duration of anal HPV infections between smoking groups using ORs and 95% CIs were calculated using a multivariable logistic regressions. Models were adjusted for baseline known potential confounders that were previously described.

All statistical analyses were conducted using STATA software, version 15.1 (StataCorp. LLC).

Data availability

Data that support this study are only available upon requests. They are not publicly accessible due to the delicate nature of the data presented in this study. All data pertinent to this study are incorporated in the article or uploaded as Supplementary Material. Study dataset with deidentified participant data and protocol can be requested from Anna.Giuliano@moffitt.org. Data requests can also be directed to the Center for Immunization and Infection Research, which is the institutional organization that handles data requests. Their email is: CIIRC@moffitt.org.

Results

Among the men with baseline anal HPV samples ($n = 1,994$), 23% ($n = 454$) were current smokers, 19% ($n = 379$) were former smokers, and 58% ($n = 1,161$) had never smoked. The median pack-years smoked among current and former smokers were 3 years. **Table 1** shows the demographic characteristics of study participants stratified by smoking status. In this study population, 45% of participants were between 18 and 30 years old, 39% were between 31 and 44 years old, and 15% were between 45 and 70 years old. Mexico had the highest proportion of participants that are current and former smokers (45% and 37%, respectively). About 42% of MSW and 44% of MSM were ever smokers. Lifetime number of female sexual partners differed by smoking status, but lifetime male sexual partners did not differ. Former smokers had been smoking for an average of 11 years and 10 cigarettes

Table 1. Demographic characteristic stratified by smoking status.

Characteristic	Current ($n = 454$) *N* (%)	Former ($n = 379$) *N* (%)	Never ($n = 1,161$) *N* (%)
Age categories			
18–30	202 (44.5)	105 (27.7)	602 (51.8)
31–44	193 (42.5)	163 (43.0)	431 (37.1)
45–70	59 (13.0)	111 (29.3)	128 (11.0)
Country of residence			
US	110 (24.2)	120 (31.7)	396 (34.1)
Mexico	205 (45.1)	142 (37.5)	304 (26.2)
Brazil	139 (30.6)	117 (30.9)	461 (39.7)
Race			
White	170 (37.4)	182 (48)	562 (48.4)
Black	63 (13.9)	39 (10.3)	204 (17.6)
Asian/PI	6 (1.3)	3 (0.8)	32 (2.8)
Other	205 (44.1)	149 (39.3)	328 (28.2)
Refused	10 (2.2)	6 (1.6)	35 (3)
Ethnicity			
Non-Hispanic	198 (43.6)	186 (49.5)	667 (58.4)
Hispanic	256 (56.4)	190 (50.5)	476 (41.6)
Marital status			
Divorced/separated	52 (11.5)	44 (11.6)	88 (7.6)
Married/cohabitating	205 (45.4)	228 (60.2)	497 (43.0)
Single	195 (43.1)	107 (28.2)	572 (49.4)
Sexual orientation			
MSM	97 (21.4)	84 (22.2)	224 (19.3)
MSW	303 (66.7)	267 (70.5)	787 (67.8)
Unknown	54 (11.9)	28 (7.4)	150 (12.9)
Lifetime male sexual partners			
0–1	370 (81.5)	316 (83.38)	982 (84.58)
2–9	37 (8.15)	35 (9.23)	91 (7.84)
10–49	28 (6.17)	15 (3.96)	49 (4.22)
50+	11 (2.42)	9 (2.37)	28 (2.41)
Missing	8 (1.8)	4 (1.1)	11 (0.9)
Lifetime female sexual partners			
0–1	62 (13.7)	33 (8.7)	292 (25.1)
2–9	163 (36.0)	141 (37.2)	477 (41.1)
10–49	159 (35.0)	161 (42.5)	297 (25.6)
50+	37 (8.1)	25 (6.6)	50 (4.3)
Missing	33 (7.3)	19 (5.0)	45 (3.9)
Years smoked			
Mean (std)	14.7 (11.3)	10.8 (9.2)	n/a
Median (IQR)	12 (7–20)	9 (3–15)	n/a
Average cigarettes per day			
Mean (std)	8.5 (7.7)	10.4 (11.7)	n/a
Median (IQR)	6 (3–12)	5 (2–15)	n/a
Pack years			
Mean (std)	7.5 (10.4)	8.1 (15.7)	n/a
Median (IQR)	3.6 (1.0–9.8)	2.4 (0.5–10)	

Abbreviations: std, standard deviation; IQR, interquartile range.

per day whereas current smokers had been smoking for an average of 14 years with 9 cigarettes per day.

Anal HPV genotype prevalence by smoking status is described in **Table 2**. Overall, the prevalence of any anal HPV was 23.6% (95% CI, 19.9–27.2) among current smoker, 15.8% (95% CI, 12.5–19.9) among former smoker and 17.4% (95% CI, 15.3–19.7) among never smoker. Any LR-HPV prevalence was highest in current smoker (18.1%; 95% CI, 14.8–21.9) followed by former smoker (12.7%; 95% CI, 9.6–16.4) and never smoker (11.5%; 95% CI, 9.8–13.5). When stratified by sexual orientation, there were no statistically significant differences in the prevalence of anal HPV groupings among MSM by smoking status. However, among MSW the prevalence of any anal HPV was 16.8% (95% CI, 13.0–21.5) in current smoker, 9.7% (95% CI, 6.7–13.9) among former smoker and 10.3% (95% CI, 8.3–12.6) among never smoker. Anal LR- and HR-HPV prevalence were also higher among current MSW smokers compared with never smokers. The prevalence of multiple infections was highest among current smokers in all three groups: Overall, MSM, and MSW. Prevalence of individual HPV genotypes by smoking status is shown in Supplementary Table S1. There were no differences in anal HPV prevalence by smoking intensity among current or former smokers (Supplementary Table S2).

The association between anal HPV prevalence and smoking status is presented in **Table 3**. The unadjusted PRs for HPV prevalence and smoking are shown in Supplementary Table S3. Current smokers had a

Table 2. Prevalence and 95% CIs of HPV genotypes by smoking status overall and stratified by sexual orientation.

	Overall ($n = 1,994$)				MSM ($n = 405$)			MSW ($n = 1,357$)		
	Current ($n = 454$)	Former ($n = 379$)	Never ($n = 1,161$)	Current ($n = 97$)	Former ($n = 84$)	Never ($n = 224$)	Current ($n = 303$)	Former ($n = 267$)	Never ($n = 787$)
HPV Status									
Negative	76.4 (72.3–80.1)	84.2 (80.1–87.5)	82.6 (80.3–84.7)	54.6 (44.6–64.3)	66.7 (55.9–75.9)	54.5 (47.9–60.9)	83.2 (78.5–86.9)	90.3 (86.1–93.3)	89.7 (87.3–91.6)
Any[a]	23.6 (19.9–27.7)	15.8 (12.5–19.9)	17.4 (15.3–19.7)	45.4 (35.7–55.3)	33.3 (24.1–44.1)	45.5 (39.1–52.1)	16.8 (13.0–21.5)	9.7 (6.7–13.9)	10.3 (8.3–12.6)
Low-risk[a]	18.1 (14.8–21.9)	12.7 (9.6–16.4)	11.5 (9.8–13.5)	37.1 (28.1–47.1)	27.4 (18.9–37.8)	31.7 (25.9–38.1)	12.2 (8.9–16.4)	7.1 (4.6–10.9)	6.3 (4.8–8.3)
High-risk[a]	13.6 (10.8–17.1)	7.6 (5.4–10.8)	11 (9.3–12.9)	27.8 (19.8–37.6)	20.2 (12.9–30.2)	31.7 (25.9–38.1)	8.91 (6.2–12.7)	4.1 (2.29–7.3)	5.7 (4.3–7.6)
Multiple infections[b]	13.2 (10.4–16.6)	5.5 (3.6–8.3)	8.3 (6.8–9.9)	36.1 (27.1–46.1)	19 (11.9–28.9)	27.2 (21.8–33.4)	6.3 (4.0–9.6)	1.5 (0.6–3.9)	3.2 (2.1–4.6)

[a]Any, high-risk, low-risk prevalence of one or more anal HPV infections for that grouping.
[b]Multiple infections, having two or more prevalent any anal HPV infections.

higher prevalence of any anal HPV [adjusted PR (aPR), 1.36, 95% CI, 1.06–1.73] and LR-HPV (aPR, 1.59; 95% CI, 1.20–2.12) compared with never smokers after adjusting for age, country, lifetime female sexual partners, and sexual orientation. Current smokers had a significantly higher prevalence of multiple infections (aPR, 1.57; 95% CI, 1.12–2.18) compared with never smokers. There was no significant difference between former smoker and never smokers in the prevalence of anal HPV. The association between smoking intensity, defined as ≥3 pack-years or <3 pack-years among current and former smokers and the prevalence of anal HPV was assessed (Supplementary Table S4). Current smokers with ≥3 pack-years had a significantly higher prevalence of LR-HPV (aPR, 1.66; 95% CI, 1.18–2.34) and multiple infections (aPR, 1.63; 95% CI, 1.09–2.45) compared with never smokers. Current smokers with <3 pack-years had a significantly higher prevalence of any HPV and LR-HPV. In a *post hoc* analysis, we stratified by country and found that current smokers in the US had a significantly higher prevalence of any (aPR, 2.04; 95% CI, 1.24–3.36), LR-HPV (aPR, 3.25; 95% CI, 1.67–6.29), and multiple infections (aPR, 2.63; 95% CI, 1.11–6.27) compared with never smokers (Supplementary Table S5). There were no significant differences in Brazil or Mexico comparing the prevalence of anal HPV by smoking status (Supplementary Table S5).

The association between smoking status and anal HPV infection stratified by sexual orientation is presented in **Table 3**. Anal HPV prevalence did not statistically differ by smoking status among MSM. Among MSW, current smokers had an increased prevalence of LR-HPV (aPR, 1.60; 95% CI, 1.02–2.50) compared with never smokers after adjusting for age, country, and lifetime female sexual partners. Current MSW smokers were more likely to have multiple HPV infections (aPR, 1.79; 95% CI, 1.35–2.39) compared with never smokers. In addition, when smoking intensity (<3 or ≥3 pack-years) was assessed among current smokers, both were not associated with increased prevalence of multiple infections compared with never smokers. There were no significant differences between never and former smokers and anal HPV prevalence among MSW.

Incidence of any HPV was 37% among current smokers, 25% among former, and 28% among never smokers. The association between smoking and the incidence of HPV infections is presented in **Table 4**. The unadjusted ORs for incident infections are shown in Supplementary Table S6. Overall, current smokers were significantly more likely to have an incident any HPV (aOR, 1.74; 95% CI, 1.26–2.39), incident HR-HPV (aOR, 1.63; 95% CI, 1.15–2.31) and incident LR-HPV (aOR, 1.62; 95% CI, 1.16–2.27) infection compared with never smokers. Among MSM, current smokers were significantly more likely to have an incident HR-HPV infection [adjusted OR (aOR), 1.73; 95% CI, 1.00–2.99]. Among MSW, current smokers were significantly more likely to have an incident any HPV (aOR, 1.82; 95% CI, 1.17–2.82) and incident LR-HPV (aOR, 1.99; 95% CI, 1.23–3.23) infection compared with never smokers. There were no significant differences between never and former smokers and anal HPV incidence.

Persistence of any HPV greater than 12 months among current smokers was 26%, 19% among former smokers and 19% among never smokers. The effect of smoking on HPV infections persisting ≥12 months is presented in **Table 5**. The unadjusted ORs for persistent infections are shown in Supplementary Table S7. Overall, current smokers were significantly more likely to have an HPV infection persist greater than 12 months compared with never smokers (aOR, 1.67; 95% CI, 1.19–2.33). This same association was seen among MSW (aOR, 1.83, 95% CI, 1.12–2.98). However, smoking was not significantly associated with HPV persistence among MSM.

Table 3. PRs for HPV infection by smoking status and sexual orientation.

	Any HPV[a] aPR (95% CI)[c]	High-risk HPV[a] aPR (95% CI)[c]	Low-risk HPV[a] aPR (95% CI)[c]	Multiple infections[b] aPR (95% CI)[c]
Overall				
Smoking status				
Never	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)
Former	0.97 (0.72–1.31)	0.77 (0.51–1.17)	1.21 (0.86–1.71)	0.78 (0.48–1.28)
Current	**1.36 (1.06–1.73)**	1.24 (0.91–1.70)	**1.59 (1.20–2.12)**	**1.57 (1.12–2.18)**
MSM				
Smoking status				
Never	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)
Former	0.93 (0.60–1.45)	0.81 (0.46–1.42)	1.08 (0.66–1.77)	0.92 (0.69–1.24)
Current	0.93 (0.65–1.35)	0.79 (0.50–1.25)	1.10 (0.73–1.66)	0.94 (0.75–1.19)
MSW				
Smoking status				
Never	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)
Former	0.81 (0.51–1.28)	0.68 (0.35–1.35)	0.94 (0.54–1.63)	0.77 (0.52–1.14)
Current	1.39 (0.96–2.02)	1.41 (0.85–2.33)	**1.60 (1.02–2.50)**	**1.79 (1.35–2.39)**

[a]Any, high-risk, low-risk, models assessed the prevalence of one or more anal HPV infections for that grouping.
[b]Multiple infections, models assessed having two or more prevalent any anal HPV infections.
[c]aPR, multivariable PR adjusted for age, country, sexual orientation and lifetime female sexual partners (overall); age, country, lifetime female sexual partners, and lifetime male sexual partners (MSM); age, country, and lifetime female sexual partners (MSW).

Discussion

In this multinational cohort of HIV-negative men, the prevalence, incidence, and duration of anal HPV by smoking status and smoking intensity was assessed. Current smokers had a significantly higher prevalence and incidence of any anal HPV and LR-HPV compared with never smokers and were more likely to have prevalent multiple HPV infections than never smokers. Interestingly, there were no differences in prevalence, incidence, or duration of anal HPV between former and never smokers. When models were stratified by sexual orientation, MSW that currently smoke had a significantly higher prevalence and incidence of anal any LR-HPV and prevalent multiple HPV infections compared with never smokers. There was no difference between HPV prevalence and duration among MSM that currently smoke compared with never smokers, but current smokers had a significantly higher HR-HPV incidence compared with never.

Among MSM without HIV, the prevalence of anal HPV is common with 47% being infected with any HPV in a recent meta-analysis (21). In this study, 43% of MSM were infected with any HPV and HPV prevalence was not associated with smoking status. These results are consistent with two studies among MSM that found that current smoking was not associated with the prevalence of any HPV compared with those not currently smoking (22, 23) and our prior work among MSM in the *HIM Study* (24). Although Goldstone and colleagues (25) reported a significant increased prevalence of any HPV among MSM former smokers compared with never smokers but no difference between current and never smokers. Smoking may not be associated with prevalent HPV infection, but there was a significantly higher

Table 4. ORs for incident infections by smoking status and sexual orientation.

	Any HPV[a] aOR (95% CI)[b]	High-risk HPV[a] aOR (95% CI)[b]	Low-risk HPV[a] aOR (95% CI)[b]
Overall			
Smoking status			
Never	1.0 (ref)	1.0 (ref)	1.0 (ref)
Former	1.06 (0.73–1.54)	0.87 (0.56–1.35)	1.366 (0.923–2.0)
Current	**1.74 (1.26–2.39)**	**1.63 (1.15–2.31)**	**1.62 (1.16–2.27)**
MSM			
Smoking status			
Never	1.0 (ref)	1.0 (ref)	1.0 (ref)
Former	0.76 (0.4–1.45)	0.72 (0.37–1.4)	0.96 (0.5–1.86)
Current	1.21 (0.67–2.18)	**1.73 (1.00–2.99)**	0.87 (0.49–1.55)
MSW			
Smoking status			
Never	1.0 (ref)	1.0 (ref)	1.0 (ref)
Former	0.99 (0.59–1.7)	0.79 (0.39–1.59)	1.34 (0.76–2.36)
Current	**1.82 (1.17–2.82)**	1.42 (0.82–2.47)	**1.99 (1.23–3.23)**

[a]Any, high-risk, low-risk, models assessed the incidence of one or more anal HPV infections for that grouping.
[b]aOR, multivariable OR adjusted for age, country, sexual orientation and lifetime female sexual partners (overall); age, country, lifetime female sexual partners, and lifetime male sexual partners (MSM); age, country, and lifetime female sexual partners (MSW).

Table 5. ORs for persistent infection lasting ≥12 months by smoking status and sexual orientation.

	Any HPV[a] aOR (95% CI)[b]	High-risk HPV[a] aOR (95% CI)[b]	Low-risk HPV[a] aOR (95% CI)[b]
Overall			
Smoking status			
Never	1.0 (ref)	1.0 (ref)	1.0 (ref)
Former	1.19 (0.80–1.76)	0.72 (0.42–1.24)	1.06 (0.69–1.64)
Current	**1.67 (1.19–2.33)**	1.40 (0.94–2.12)	1.32 (0.91–1.91)
MSM			
Smoking status			
Never	1.0 (ref)	1.0 (ref)	1.0 (ref)
Former	1.26 (0.66–2.41)	0.78 (0.38–1.62)	1.13 (0.59–2.16)
Current	1.10 (0.63–1.91)	0.88 (0.49–1.59)	0.93 (0.53–1.62)
MSW			
Smoking status			
Never	1.0 (ref)	1.0 (ref)	1.0 (ref)
Former	0.9 (0.49–1.64)	0.52 (0.19–1.44)	0.96 (0.50–1.87)
Current	**1.83 (1.12–2.98)**	1.89 (0.94–3.78)	1.51 (0.84–2.69)

[a]Any, high-risk, low-risk, models assessed the persistence of one or more anal HPV infections lasting ≥12 months for that grouping.
[b]aOR, multivariable OR adjusted for age, country, sexual orientation and lifetime female sexual partners (overall); age, country, lifetime female sexual partners, and lifetime male sexual partners (MSM); age, country, and lifetime female sexual partners (MSW).

incidence of HR-HPV among smokers compared with never smokers. The association between anal HPV incidence and persistence and smoking among MSM without HIV has not previously been reported in the literature. Further studies are needed to assess the role of smoking in anal HPV persistence and development of anal disease among MSM without HIV.

On the basis of our knowledge, we are the first to report here the role of smoking in anal HPV prevalence, incidence, and persistence among MSW without HIV. As expected, the prevalence of anal HPV among MSW (12%) was significantly lower compared with MSM (47%). Current smokers had a significantly higher prevalence of LR-HPV and multiple HPV infections compared with never smokers. We previously reported risk factors associated with anal HPV prevalence among a smaller subset of MSW in the *HIM Study*, and in adjusted models found that formers smokers were significantly less likely to have a prevalent HPV infection than never smokers (24). Although there are no other published studies among MSW without HIV, among MSW with HIV, current smokers were significantly more likely to have prevalent HPV infection compared with nonsmokers (26).

We also found that MSW that are current smokers were more likely to have an incident LR-HPV infection compared with never smokers and any HPV infections were more likely to persist >12 months in current smokers compared with never. LR-HPV infections are common and are associated with condylomas within the anal canal and genitals (27). Although the majority of anal cancers are caused by HPV16, low-risk HPV genotypes like HPV6 have been shown to cause anal cancer and anal warts (1). MSW are likely not having anal intercourse, yet we are detecting HPV within the anal canal. It is possible that these infections are occurring through autoinoculation or partner inoculation. It is interesting that the LR-HPV prevalence and incidence were strongly associated with smoking status among MSW, but we did not see a significant association with HR-HPV. Within the *HIM Study*, we have reported MSW with genital HPV infections are more likely to acquire anal HPV compared with men without genital infections (28). Current smokers are also more likely to have prevalent genital HPV infection compared with never smokers (9) and current smokers had a higher incidence of LR-HPV at the genitals compared with never smokers (29).

Tobacco smoking has been shown to be associated with anal cancer development in several studies (30–34). Several case–control studies in both men and women have reported a 2-fold increased odds of anal cancer among smokers compared with non-smokers (30–33). Recent data from the Surveillance, Epidemiology, and End Results showed counties with high smoking prevalence have higher anal cancer incidence in both males (4% per year) and females (5% per year) compared with counties with low smoking prevalence (34). Similarly, studies among people living with HIV also found an association between smoking and anal cancer (35). Smoking has also been shown to increase the risk for other HPV-associated cancers (36–40). The tobacco use could be affecting HPV persistence or progression of HPV-related lesions to cancer by reducing the immune response to HPV infections (13, 38).

Strengths of this study include large sample size, data from three international sites and ability to assess the association among both MSW and MSM. Very few studies have assessed the prevalence of anal HPV among MSW populations or MSM without HIV. The three sites had standard protocols for specimen processing and data collection. To interpret our results, it is important to keep in mind some of the limitations of this study. The results may not be generalizable to the broader male population within these communities given that the HIM Study is not a population-based cohort study. The demographics such as age, ethnicity, and education of the participants included at each site are similar to communities that they were enrolled from. Lifetime number of sexual partners were adjusted for the models, but this does not capture the different types of sexual intercourse. Although these data are unique, there were only 405 MSM included in these analyses, and we may have been limited in the power to detect significant differences. There are differences in anal HPV prevalence among MSM by type of anal intercourse, that is, either receptive or insertive. Detection of HPV infections does not mean that the infection is active and could cause disease. Further research is needed to understand fully the role of smoking in progression of HPV infections to disease in both MSM and MSW.

In conclusion, our study found currently smoking was associated with a higher prevalence and incidence of anal HPV. Although we did not see a difference in the prevalence and duration of anal HPV among

MSM that currently smoke, currently smoking was associated with incident HR-HPV infections. MSW that currently smoked had a higher prevalence and incidence of LR-HPV. There were no differences in the prevalence, incidence, or duration of anal HPV between former and never smokers. Smoking may alter the immune response to HPV infection and smoking cessation could be an important step in preventing anal cancer. Therefore, it is important to conduct more studies on the effects of smoking on anal HPV persistence and progression to anal disease and to educate the public about the increased risk for anal HPV infection and anal cancer among smokers.

Authors' Disclosures

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Authors' Contributions

V. Umutoni: Conceptualization, formal analysis, validation, investigation, methodology, writing–original draft. **M.B. Schabath:** Conceptualization, investigation, methodology, writing–review and editing. **A.G. Nyitray:** Conceptualization, funding acquisition, investigation, writing–review and editing. **T.J. Wilkin:** Conceptualization, methodology, writing–review and editing. **L.L. Villa:** Resources, data curation, funding acquisition, methodology, writing–review and editing. **E. Lazcano-Ponce:** Resources, data curation, funding acquisition, writing–review and editing. **A.R. Giuliano:** Conceptualization, resources, data curation, supervision, funding acquisition, investigation, methodology, writing–review and editing. **S.L. Sudenga:** Conceptualization, resources, data curation, formal analysis, supervision, funding acquisition, methodology, writing–original draft, writing–review and editing.

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