Published in final edited form as:
Cancer Epidemiol Biomarkers Prev. 2012 January ; 21(1): 102–110. doi: 10.1158/1055-9965.EPI-11-0591.

Smoking and Human Papillomavirus (HPV) Infection in the HPV in Men (HIM) Study

Matthew B. Schabath[1], **Luisa L. Villa**[2], **Eduardo Lazcano-Ponce**[3], **Jorge Salmerón**[3,4], **Manuel Quiterio**[3], and **Anna R. Giuliano**[1] **for the HIM Study**

[1]H. Lee Moffitt Cancer Center and Research Institute, Tampa, FL

[2]Ludwig Institute for Cancer Research

[3]Instituto Nacional de Salud Pública

[4]Instituto Mexicano del Seguro Social.

Abstract

Background—The influence of smoking on the natural history of HPV infection in men is not well-understood. Smoking could influence the incidence and persistence of HPV infections by suppressing local immune function, increase cellular proliferation, up-regulate pro-inflammatory factors, or cause host DNA damage resulting in increased susceptible to infection. The purpose of this analysis is to assess prevalent HPV infections by smoking status in men, and to determine baseline risk of HPV infection associated with smoking.

Methods—The HPV in Men (HIM) study is a multinational prospective study of the natural history of HPV infections in men. Samples from the coronal sulcus, glans penis, shaft, and scrotum were combined for HPV DNA testing. Multivariable logistic regression was used to assess the association between smoking and any-, oncogenic-, and non-oncogenic HPV infections.

Results—Our analyses revealed that current smoking was associated with an increased risk of any HPV infection (OR = 1.19; 95% CI 1.01 – 1.41) and oncogenic HPV infection (OR = 1.24; 95% CI 1.05 – 1.47). However, the association between smoking and any HPV infection (OR = 1.35; 95% CI 1.05 – 1.73) and oncogenic HPV infection (OR = 1.46; 95% CI 1.11 – 1.92) was only evident among men reporting fewer lifetime sexual partners.

Discussion—These results suggest that current smokers with the fewest number of sexual partners are associated with an increased risk for oncogenic HPV infection.

Impact—The relationship between smoking and HPV infection remains understudied in men; these data sheds new light on the interplay between smoking, sexual activity, and risk of HPV infection.

Keywords

Smoking; HPV; Men; Genital Infection; Oncogenic HPV

INTRODUCTION

Human papillomavirus (HPV) is one the most common sexually transmitted infections, with over 6 million new infections occurring annually in the US (1, 2). Over 120 different HPV

Corresponding Author: Matthew B. Schabath, Ph.D H. Lee Moffitt Cancer Center and Research Institute Cancer Epidemiology Program 12902 Magnolia Drive MRC-CANCONT, Tampa, FL 33612 Fax: 1-813-745-6525 Matthew.Schabath@Moffitt.org.

types have been identified, more than 30 of which are transmitted through sexual contact. In addition to the diseases HPV causes directly in men, including genital warts and various cancers, the HPV virus is readily transmitted from men to women and can affect disease risk in women (3-6). However, most HPV infections are transient and asymptomatic or subclinical, do not result in disease, and are usually self-cleared.

Previous epidemiology studies have provided widely varied estimates of HPV prevalence in men ranging from 0 to 73% (7). While prevalence findings are mixed, even less is known about the relationship between HPV and smoking in men. In studies conducted among women, smoking has been associated with longer duration of oncogenic HPV infections as well as increased risk of invasive cervical cancer (8-16). Thus, since approximately one third of men worldwide are active smokers (17) and HPV appears to be highly prevalent in men, it is important to determine the potential influence of smoking on the natural history of HPV infection in men. Smoking could influence the incidence and persistence of HPV infections by suppressing local immune function, increase cellular proliferation and turnover, up-regulate pro-inflammatory factors, or cause host DNA damage resulting in increased susceptible to infection (18-34). The purpose of this analysis was to assess prevalent HPV infections by smoking status in men and determine the baseline risk of HPV infection associated with smoking.

MATERIALS AND METHODS

Study Population

The HPV in Men (HIM) study is a prospective study of the natural history of HPV infections in men in three countries. A full description of cohort methods and procedures has been previously published (35, 36). Briefly, men aged 18 to 70 years were recruited from Tampa, Florida, USA, São Paulo, Brazil, and Cuernavaca, Mexico. Eligibility criteria included no previous diagnosis of penile or anal cancers, no previous diagnosis of genital or anal warts, had not participated in an HPV vaccine study, no previous diagnosis of HIV, no current penile discharge or burning during urination, not under current treatment for sexually transmitted infection, no history of imprisonment or homelessness during the past 6 months, no drug treatment during the past 6 months, no plans to relocate in the next 4 years, and willingness to attend ten visits scheduled every 6 months for 4 years.

Men were recruited from three different population sources: the general population, universities, and organized health-care systems. In Brazil, men were recruited from a large clinic in São Paulo that provides genitourinary healthcare and from the general population through television, radio, and newspaper advertisements. In Mexico, men were recruited in Morelos state, through a large government health care system, from factories, and the military. In the USA, men were mainly recruited from the University of South Florida and the general community in Tampa, FL. Human subjects' committees of the University of South Florida, Tampa, FL, the Ludwig Institute for Cancer Research, São Paulo, Brazil, the Centro de Referencia e Treinamento de Doencas Sexualmente Transmissiveis e AIDS, São Paulo, Brazil, the Instituto Mexicano del Seguro Social, and the National Institute of Public Health of Mexico, Cuernavaca, Mexico, approved all study procedures.

Data and Sample Collection

Eligible men provided written informed consent and underwent a clinical examination at a visit 2 weeks before the enrollment visit and every 6 months thereafter. Only men who returned for the enrollment visit were included in the study. At each visit, participants completed a computer-assisted self-interview questionnaire. Samples of penile and scrotal cells were obtained at each visit for detection of HPV DNA by use of PCR and subsequent

genotyping. To encourage compliance with follow-up, men in the US and Brazil were compensated for their participation.

Risk Factor Questionnaire

At enrollment an extensive sexual history and health questionnaire was administered which collected sociodemographic characteristics, sexual history, condom use practices, and alcohol and tobacco use. The questionnaire was self-administered using computer assisted self-interviewing (CASI) and was typically completed in approximately 20 minutes. Never smokers were defined as men who had smoked less than 100 cigarettes in their lifetime. Former smokers were defined as men who had smoked at least 100 cigarettes in their lifetime but quit smoking at least 1 year before the baseline interview. Pack-years smoked were calculated using the average number of cigarette packs smoked per day and the numbers of years smoked.

HPV Penile and Scrotal Sampling

To maximize sampling and prevent fraying of applicators, three different pre-wetted Dacron applicators were used to sample the external genitalia and combined into a single sample for HPV detection. This method has been shown to maximize HPV detection and result in reproducible detection of genital HPV in men (37, 38). The study clinician at each site first swept 360° around the coronal sulcus and then another 360° around the glans penis and placed the swab into a collection vial containing Specimen Transport Medium (STM, Digene. Corp., Gaithersburg, MD). A second swab was used to sample the entire skin surface of each of the quadrants of the shaft of the penis (left and right ventral, and left and right dorsal) and placed into a separate collection vial. A third swab was used for scrotum sampling. All 3 swabs were placed in separate collection vial. Among men who were uncircumcised, the foreskin was sampled at the time of collection of the coronal sulcus/glans penis sample. All HPV samples were stored at −70°C until PCR analyses and genotyping were conducted. Prior to DNA extraction, the three samples were combined to produce one DNA extract per participant clinic visit.

DNA Extraction and HPV Genotyping

DNA extraction was accomplished using the QIAamp DNA Mini Kit (Qiagen, Valencia, CA) and HPV testing of the combined DNA sample was conducted using PCR for amplification of a fragment of the HPV L1 gene. Briefly, 200 μL aliquots of clinical material were digested with 20 μL of proteinase K solution for 1 h at 65°C, followed by 200 μL of lysis buffer. Specimens were tested for the presence of HPV by amplifying 50 μL of the DNA extracts using the Linear Array HPV genotyping test following manufacturer instructions (Roche Diagnostics, San Francisco, CA). Samples were amplified using Perkin-Elmer GeneAmp PCR System 9700 as directed by the linear array protocol. HPV genotyping was conducted on all samples regardless of HPV PCR result. 96.8% of specimens obtained were positive for β-Globin.

Statistical Analysis

Three HPV categories (i.e., “Any HPV”, “Oncogenic HPV”, and “Only non-oncogenic HPV”) were used as the dependent variables in our analyses. A participant was considered positive for “any HPV” if he tested HPV-positive by PCR or tested positive for at least one genotype. The “Oncogenic HPV” category included men who were positive for at least one of the 13 oncogenic types tested for (HPV types 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, and 66) and included men infected with both oncogenic and non-oncogenic types. “Non-oncogenic HPV” infections included single or multiple infections with only non-oncogenic HPV types (6, 11, 26, 40, 42, 53, 54, 55, 61, 62, 64, 67–73, 81–84, IS39, and CP6108) The

Pearson's X^2 test was used to test for differences for the prevalence of HPV by smoking status and multivariable logistic regression models were used to estimate odds ratios (ORs) and 95% confidence intervals (CIs) for the association between smoking and HPV prevalence. Two different logistic regression models were used to generate multivariable ORs (mORs). The first model adjusted for only study design variables (i.e., age and country) and the second model adjusted for the study design variables and a common HIM study list of covariates that are associated with HPV and/or are potential confounders including, where appropriate, ethnicity, circumcision, total number of female partners in the last 3 months, and total number of vaginal sex partners. Interaction was tested with a multiplicative interaction term included in the full multivariate model. All statistical tests were two-sided. All statistical analyses were performed using Stata Intercooled v10.1 (StataCorp, College Station, Texas).

RESULTS

The study population demographic characteristics are presented by smoking status in Table 1. Never smokers (mean age = 30.9; SD 10.7) were younger than former (mean age = 37.3; SD 12.2) and current smokers (mean age = 32.6; SD 10.1) ($P < 0.05$). Among never smokers, 36.2% were from the US, 37.7% from Mexico and 26.1% from Brazil. Compared to current smokers, never smokers had a higher frequency of self-indentified Whites (46.7% versus 39.5%), Blacks (18.1% versus 13.7%), Asian/Pacific Islanders (3.8% versus 1.3%), and Non-Hispanics (59.3% versus 44.6%) ($P < 0.05$). Never- versus current smokers also reported a slightly higher frequency of circumcision (35.9% versus 32.3%). Current smokers self-reported a higher frequency of ten or more lifetime vaginal sex partners (43.8% versus 31.2%) and a lower frequency of no vaginal sex partners (7.7% versus 11.8%; $P < 0.05$). Current smokers also reported a higher frequency of total number of female partners in the last 3 months (≥ 3 female partners: 14.5% versus 13.2%; 2 female partners: 13.4% versus 12.9%; 1 female partner: 60.6% versus 59.2%; $P < 0.05$).

Prevalence of any HPV (Table 2) was statistically significantly ($P = 0.015$) lower among never smokers (63.2%) compared to former- (66.7%) and current smokers (68.2%). Similar trends were also observed for prevalence of HPV infection with at least one oncogenic type ($P < 0.001$) and only non-oncogenic types ($P = 0.009$). These three HPV categories (i.e., "Any HPV", "Oncogenic HPV", and "Only non-oncogenic HPV") were used as the dependent variables for the analyses in Tables 3 to 5. Additionally, the prevalence of infection for the other HPV categories in Table 2 were all statistically significantly lower for never smokers except for only oncogenic types which revealed no statistically significant different ($P = 0.241$).

To determine if there were differences (i.e., potential confounding) in HPV positivity with increasing smoking intensity, we assessed the prevalence of HPV infection across tertiles of cigarettes smoked/day, years smoked, and pack-years smoked for current- and former smokers. Among former smokers, there was no statistically significant difference in the prevalence of HPV infection by the tertile distributions of cigarettes smoked per day and pack-years smoked (Table 3). However, prevalence of any HPV infection was higher for the third tertile of years smoked versus the first tertile (73.3% versus 62.9%; $P = 0.025$), as was prevalence of only non-oncogenic HPV (23.9% versus 21.7%; $P = 0.041$). We also assessed mean number of cigarettes smoked per day and mean pack-years smoked 1) between former and current smokers for each HPV category (i.e., "Any HPV", "Oncogenic HPV", and "Only non-oncogenic HPV"), and 2) among former and current smokers by HPV status (compared to participants who were HPV negative) and found no statistically significant differences (data not shown).

When we assessed the association between smoking and HPV infection (Table 4), we found that current smokers were statistically significantly associated with an increased risk for any HPV (OR = 1.19; 95% CI 1.01 – 1.41) and oncogenic HPV (OR = 1.24; 95% CI 1.05 – 1.47) after adjusting for age, race, ethnicity, country, circumcision, total number of female partners in the last 3 months, and total number of vaginal sex partners. Compared to never smokers, current smokers who smoked greater than 5 pack-years exhibited an increased risk of oncogenic HPV (OR = 1.29; 95% CI 1.02 – 1.64). We found no statistically significant association between smoking and non-oncogenic HPV infection or among men who smoked < 5 pack-years.

We assessed the interplay of sexual activity, smoking, and risk of HPV infection (Table 5) and found that current smokers with the fewest number of partners (1 to 9 partners) were at increased risk of any HPV infection (OR = 1.35; 95% CI 1.05 – 1.73) and oncogenic HPV infection (OR = 1.46; 95% CI 1.11 – 1.92) after adjusting for age, race, ethnicity, country, circumcision, total number of female partners in the last 3 months, and total number of vaginal sex partners. Among men with zero lifetime partners, elevated ORs that were not statistically significant were observed for any- and oncogenic HPV infection. As noted in Table 4, we found no statistically signification associations for only non-oncogenic HPV infection for any of the strata. We also found that men with ≥ 10 lifetime partners consistently exhibited ORs near the null. We also stratified number of partners by more narrow categories (i.e., 1 to 3; 4 to 6, 7 to 9; 10 to 49, and ≥ 50), but found no appreciable differences in the results (data not shown).

Finally, we also analyzed the data by country, sexual orientation, and circumcision and found that the results were relatively consistent for: all three countries, by sexual orientation (i.e., No sex, men who have sex with women and men [MSWM], and men who have sex with men [MSM}, and among non-circumcised men (data not shown). However, because of the reduced sample size the point estimates were not statistically significant for the three countries and among non-circumcised men. Moreover, the number of MSM for each stratum was quite small which yielded imprecise and uninterruptable point estimates.

DISCUSSION

In this multinational cohort study of HPV in men, our analyses suggest that current smoking was associated with an increased risk of any HPV infection and oncogenic HPV infection. However, the observed association with smoking was limited to men reporting fewer lifetime female sexual partners.

Limited data exist on the association between HPV infection and smoking in men. Our results are generally consistent with prior findings in women that cigarette smoking is associated with HPV load (39), HPV prevalence (8, 12-15), incidence (11, 16), and persistence (9, 10). At present it is unclear how smoking may influence HPV infection in men, but many possible mechanisms exist. For instance, laboratory studies have demonstrated that smoking increases cellular proliferation and metaplasia in various tissues and cell types (23, 29, 31, 33, 34), which in turn could result in an increase in replication or production of HPV due to smoking-induced cell proliferation. Constituents of cigarette smoke have also been shown to modify the function of immune cells (24, 25). For example, acrolein, an aldehyde found in tobacco smoke, affects neutrophil function (20), causes DNA damage (19) and has been shown to suppress resistance to pulmonary infections (27). Smoking could also potentially increase viral load by weakening the cellular immune response since previous studies have demonstrated that smoking has deleterious effects on both systemic and local immunity (18, 26, 30, 32). Smoking results in the recruitment of inflammatory cells and subsequent release of pro-inflammatory cytokines, chemotactic

factors, oxygen radicals, and proteases which alter the function of immune cells (28). Nicotine, the main compound responsible for the dependence-forming properties of smoking, has also been to be immunosuppressive in both animals (21) and in humans (22).

Although in our analysis current smoking was associated with a statistically significantly elevated risk of any HPV infection (OR = 1.19), it is likely that this effect is largely driven by the elevated risk of oncogenic HPV infection among current smokers (OR = 1.24). Consistent with the observation that current smokers may be at an increased risk of oncogenic HPV, we also found a statistically significant increased risk of oncogenic HPV among current smokers with a ≥ 5 pack-year history (OR = 1.29). No statistically significant effects were found among former smokers or among men who smoked < 5 pack-years, possibly due to the immunomodulatory properties observed among individuals who are actively exposed to cigarette smoke (18, 26, 30, 32). We also observed a novel interplay between smoking and number of sexual partners revealing that men with fewer or no sexual partners were at an increased risk for HPV infection versus men with a greater number of sexual partners. Specifically, men with zero or less than nine lifetime sexual partners exhibited modest elevated risks for any- and oncogenic HPV infection; however, the ORs were only statistically significant for the strata of 1 to 9 sexual partners. Moreover, no statistically significant effects were found among men with greater than 10 lifetime sexual partners. Increased sexual activity may result in higher HPV exposure which would then mask the influence of cigarette smoking. However, this is merely speculative and, moreover, none of the interaction tests were statistically significant. Thus, this finding warrants further analysis in other cohorts and longitudinal analyses.

The major strengths of this study are the inclusion of well-characterized international cohort of men of a wide age range (i.e., 18 to 70 years of age), and the availability of extensive and previously validated participant information (40). Another strength of the study is the large sample size of the baseline cohort, although stratification by smoking and sexual behavior did result in smaller subgroups in the present analysis. There was little evidence of confounding for the oncogenic HPV results as demonstrated by the relative consistency between the two multivariable models for each analysis. One model included only the study design variables and the other model included both the study design variables and potential confounders. None of the ORs between the two models differed by more than 10% for the oncogenic HPV results. Hence, presenting data from the more inclusive multivariable model is likely more conservative. Additionally, the data in Table 3 suggests there was no evidence that smoking intensity was confounding the results among current smokers since there were no differences in prevalence of HPV infection with increasing smoking intensity (cigarettes smoked/day, years smoked, and pack-years) among current smokers. Yet, we do acknowledge that we cannot account for bias due to unmeasured or unknown confounding cannot be accounted for. Sexual behavior is potentially an important confounder in the association between smoking and HPV positivity. Although we accounted for potential confounding by adjusting for self-reported sexual behavior and stratified by number of female partners, residual confounding still may exist which could potentially inflate the observed point estimates. Hence, our results should be interpreted with caution. Although this is a baseline cross-sectional analysis, these data are important and novel since there is limited information on the association between smoking and HPV in men. Future longitudinal analyses will be performed to assess whether smoking influences HPV acquisition and clearance. We do acknowledge that the cohort may not be a representative sample of the general male population of the participating countries, which limits the generalizability of our findings.

The biological role that smoking plays in HPV infection in men remains understudied, and limited association data exist on the association between smoking and HPV infection. This

analysis thus provides important data on the interplay between smoking, sexual activity, and men's risk of HPV infection in men. Overall, these results demonstrated that current smokers are associated with an increased risk oncogenic HPV infection.

Acknowledgments

The authors would like to thank the following staff members for their dedication in recruiting, examining, and maintaining data on cohort participants, as well as conducting HPV DNA laboratory analyses: Kathy Eyring, CCRP; Christine Gage, ARNP; Nadia Lambermont, ARNP; Kim Isaacs, BA; Andrea M. Leto, BA; Emily Jolles, MPH, Kayoko Kennedy, BA; Amanda Sivia; Pauline Schwalm-Andel, BS; Rana Zaki, MPH, Sireesha Banduvula, MS, Kyle Wolf, Steven McAnany, Shannon McCarthy, and the Tissue Core staff of the Moffitt Cancer Center for their help managing biological samples from the US site; MLuiza Baggio, Roberto Silva, Lenice Galan, Elimar Gomes, Ricardo Cintra, Viviane Relvas, Filomena Cernicchiaro, Raquel Hessel, Sandra Araujo, Graça Ribeiro, Rosária Otero, Roberta Bocalon, Juliana Antunes, Rossana Terreri, Fernanda Silva, Rubens Matsuo, Ricardo Cunha, Vera Souza, Elisa Brito, Birgit Fietzek, from the Brazil site; Verónica Chávez, Aurelio Cruz, María Griselda Díaz, Rossana del Carmen González, Pilar Hernández, Ana Laura Landa, Alicia Rodríguez and Oscar Rojas from the Mexico site. The authors would also like to thank the Digene Corporation for kindly providing STM for the collection and storage of samples at no charge to the study.

GRANT SUPPORT This project was supported through a grant from the National Cancer Institute, National Institutes of Health, CA#RO1CA098803.

REFERENCES

1. Weinstock H, Berman S, Cates W. Sexually transmitted diseases among American youth: Incidence and prevalence estimates, 2000. Perspect Sex Repro H. 2004; 36:6–10.
2. CDC. Centers for Disease Control and Prevention. [cited 2011 June 7] Genital HPV Infection - Fact Sheet. 2010. Available from: http://www.cdc.gov/std/hpv/hpv-fact-sheet-press.pdf
3. Buckley JD, Harris RW, Doll R, Vessey MP, Williams PT. Case-control study of the husbands of women with dysplasia or carcinoma of the cervix uteri. Lancet. 1981; 2:1010–5. [PubMed: 6118477]
4. Zunzunegui MV, King MC, Coria CF, Charlet J. Male influences on cervical cancer risk. Am J Epidemiol. 1986; 123:302–7. [PubMed: 3753819]
5. Agarwal SS, Sehgal A, Sardana S, Kumar A, Luthra UK. Role of male behavior in cervical carcinogenesis among women with one lifetime sexual partner. Cancer. 1993; 72:1666–9. [PubMed: 8348498]
6. Bosch FX, Castellsague X, Munoz N, de Sanjose S, Ghaffari AM, Gonzalez LC, et al. Male sexual behavior and human papillomavirus DNA: key risk factors for cervical cancer in Spain. J Natl Cancer Inst. 1996; 88:1060–7. [PubMed: 8683637]
7. Dunne EF, Nielson CM, Stone KM, Markowitz LE, Giuliano AR. Prevalence of HPV infection among men: A systematic review of the literature. J Infect Dis. 2006; 194:1044–57. [PubMed: 16991079]
8. Bauer HM, Hildesheim A, Schiffman MH, Glass AG, Rush BB, Scott DR, et al. Determinants of genital human papillomavirus infection in low-risk women in Portland, Oregon. Sex Transm Dis. 1993; 20:274–8. [PubMed: 8235925]
9. Giuliano AR, Sedjo RL, Roe DJ, Harri R, Baldwi S, Papenfuss MR, et al. Clearance of oncogenic human papillomavirus (HPV) infection: effect of smoking (United States). Cancer Causes Control. 2002; 13:839–46. [PubMed: 12462549]
10. Koshiol J, Schroeder J, Jamieson DJ, Marshall SW, Duerr A, Heilig CM, et al. Smoking and time to clearance of human papillomavirus infection in HIV-seropositive and HIV-seronegative women. Am J Epidemiol. 2006; 164:176–83. [PubMed: 16775041]
11. Minkoff H, Feldman JG, Strickler HD, Watts DH, Bacon MC, Levine A, et al. Relationship between smoking and human papillomavirus infections in HIV-infected and -uninfected women. J Infect Dis. 2004; 189:1821–8. [PubMed: 15122518]
12. Sellors JW, Karwalajtys TL, Kaczorowski J, Mahony JB, Lytwyn A, Chong S, et al. Incidence, clearance and predictors of human papillomavirus infection in women. CMAJ. 2003; 168:421–5. [PubMed: 12591782]

13. Sellors JW, Mahony JB, Kaczorowski J, Lytwyn A, Bangura H, Chong S, et al. Prevalence and predictors of human papillomavirus infection in women in Ontario, Canada. Survey of HPV in Ontario Women (SHOW) Group. CMAJ. 2000; 163:503–8. [PubMed: 11006760]
14. Syrjanen K, Shabalova I, Petrovichev N, Kozachenko V, Zakharova T, Pajanidi J, et al. Smoking is an independent risk factor for oncogenic human papillomavirus (HPV) infections but not for high-grade CIN. Eur J Epidemiol. 2007; 22:723–35. [PubMed: 17828436]
15. Vaccarella S, Herrero R, Snijders PJ, Dai M, Thomas JO, Hieu NT, et al. Smoking and human papillomavirus infection: pooled analysis of the International Agency for Research on Cancer HPV Prevalence Surveys. Int J Epidemiol. 2008; 37:536–46. [PubMed: 18316350]
16. Winer RL, Lee SK, Hughes JP, Adam DE, Kiviat NB, Koutsky LA. Genital human papillomavirus infection: incidence and risk factors in a cohort of female university students. Am J Epidemiol. 2003; 157:218–26. [PubMed: 12543621]
17. Mackay, J.; Eriksen, M. Male Smoking. World Health Organization; 2002. The Tobacco Atlas-Chapter 3.
18. Barton SE, Maddox PH, Jenkins D, Edwards R, Cuzick J, Singer A. Effect of cigarette smoking on cervical epithelial immunity: a mechanism for neoplastic change? Lancet. 1988; 2:652–4. [PubMed: 2901519]
19. Feng ZH, Hu WW, Hu Y, Tang MS. Acrolein is a major cigarette-related lung cancer agent: Preferential binding at p53 mutational hotspots and inhibition of DNA repair. P Natl Acad Sci USA. 2006; 103:15404–9.
20. Finkelstein EI, Nardini M, van der Vliet A. Inhibition of neutrophil apoptosis by acrolein: a mechanism of tobacco-related lung disease? Am J Physiol Lung Cell Mol Physiol. 2001; 281:L732–9. [PubMed: 11504702]
21. Geng Y, Savage SM, Razani-Boroujerdi S, Sopori ML. Effects of nicotine on the immune response. II. Chronic nicotine treatment induces T cell anergy. J Immunol. 1996; 156:2384–90. [PubMed: 8786295]
22. Guslandi M. Long-term effects of a single course of nicotine treatment in acute ulcerative colitis: remission maintenance in a 12-month follow-up study. Int J Colorectal Dis. 1999; 14:261–2. [PubMed: 10647637]
23. Harris TG, Kulasingam SL, Kiviat NB, Mao C, Agoff SN, Feng Q, et al. Cigarette smoking, oncogenic human papillomavirus, Ki-67 antigen, and cervical intraepithelial neoplasia. Am J Epidemiol. 2004; 159:834–42. [PubMed: 15105176]
24. Holt PG. Immune and inflammatory function in cigarette smokers. Thorax. 1987; 42:241–9. [PubMed: 3303428]
25. Holt PG, Keast D. Environmentally induced changes in immunological function: acute and chronic effects of inhalation of tobacco smoke and other atmospheric contaminants in man and experimental animals. Bacteriol Rev. 1977; 41:205–16. [PubMed: 405003]
26. Kalra R, Singh SP, Savage SM, Finch GL, Sopori ML. Effects of cigarette smoke on immune response: chronic exposure to cigarette smoke impairs antigen-mediated signaling in T cells and depletes IP3-sensitive Ca(2+) stores. J Pharmacol Exp Ther. 2000; 293:166–71. [PubMed: 10734166]
27. Li L, Holian A. Acrolein: a respiratory toxin that suppresses pulmonary host defense. Rev Environ Health. 1998; 13:99–108. [PubMed: 9718625]
28. Mehta H, Nazzal K, Sadikot RT. Cigarette smoking and innate immunity. Inflamm Res. 2008; 57:497–503. [PubMed: 19109742]
29. Peters EJ, Morice R, Benner SE, Lippman S, Lukeman J, Lee JS, et al. Squamous metaplasia of the bronchial mucosa and its relationship to smoking. Chest. 1993; 103:1429–32. [PubMed: 8486022]
30. Poppe WA, Ide PS, Drijkoningen MP, Lauweryns JM, Van Assche FA. Tobacco smoking impairs the local immunosurveillance in the uterine cervix. An immunohistochemical study. Gynecol Obstet Invest. 1995; 39:34–8. [PubMed: 7890250]
31. Sekhon HS, Wright JL, Churg A. Cigarette smoke causes rapid cell proliferation in small airways and associated pulmonary arteries. Am J Physiol. 1994; 267:L557–63. [PubMed: 7977766]
32. Sopori M. Effects of cigarette smoke on the immune system. Nat Rev Immunol. 2002; 2:372–7. [PubMed: 12033743]

33. Wright JL, Jeng AY, Battistini B. Effect of ECE and NEP inhibition on cigarette smoke-induced cell proliferation in the rat lung. Inhal Toxicol. 2001; 13:497–511. [PubMed: 11445889]
34. Wright JL, Lawson LM, Pare PD, Wiggs BJ, Kennedy S, Hogg JC. Morphology of peripheral airways in current smokers and ex-smokers. Am Rev Respir Dis. 1983; 127:474–7. [PubMed: 6838053]
35. Giuliano AR, Lazcano E, Villa LL, Flores R, Salmeron J, Lee JH, et al. Circumcision and sexual behavior: factors independently associated with human papillomavirus detection among men in the HIM study. Int J Cancer. 2009; 124:1251–7. [PubMed: 19089913]
36. Giuliano AR, Lee JH, Fulp W, Villa LL, Lazcano E, Papenfuss MR, et al. Incidence and clearance of genital human papillomavirus infection in men (HIM): a cohort study. Lancet. 2011; 377:932–40. [PubMed: 21367446]
37. Flores R, Abalos AT, Nielson CM, Abrahamsen M, Harris RB, Giuliano AR. Reliability of sample collection and laboratory testing for HPV Detection in Men. J Virol Methods. 2008; 149:136–43. [PubMed: 18279976]
38. Giuliano AR, Nielson CM, Flores R, Dunne EF, Abrahamsen M, Papenfuss MR, et al. The optimal anatomic sites for sampling heterosexual men for human papillomavirus (HPV) detection: The HPV detection in men study. Journal of Infectious Diseases. 2007; 196:1146–52. [PubMed: 17955432]
39. Xi LF, Koutsky LA, Castle PE, Edelstein ZR, Meyers C, Ho J, et al. Relationship Between Cigarette Smoking and Human Papilloma Virus Types 16 and 18 DNA Load. Cancer Epidem Biomar. 2009; 18:3490–6.
40. Nyitray AG, Kim J, Hsu CH, Papenfuss M, Villa L, Lazcano-Ponce E, et al. Test-retest reliability of a sexual behavior interview for men residing in Brazil, Mexico, and the United States: the HPV in Men (HIM) Study. Am J Epidemiol. 2009; 170:965–74. [PubMed: 19741044]

Table 1

Demographic characteristics by smoking status

Characteristic	Overall (n = 4,054)	*Smoking Status* Never (n = 2,346)	Former (n = 748)	Current (n = 960)
Age				
Mean, (SD)[a]	32.5 (11.1)	30.9 (10.7)	37.3 (12.2)	32.6 (10.1)
Categorical				
18-24	1,227 (30.3)	847 (36.10)	132 (17.7)	248 (25.8)
25-29	621 (15.3)	349 (14.9)	93 (12.4)	179 (18.7)
30-34	610 (15.1)	330 (14.1)	112 (15.0)	168 (17.5)
35-39	538 (13.3)	310 (13.2)	105 (14.0)	123 (12.8)
40-44	421 (10.4)	227 (9.7)	96 (12.8)	98 (10.2)
45-74	637 (15.7)	283 (12.1)	210 (28.1)	144 (15.0)
Country of Birth, N (%)[a]				
USA	1,338 (33.0)	850 (36.2)	209 (27.9)	279 (29.0)
Mexico	1,394 (34.4)	884 (37.7)	251 (33.6)	259 (27.0)
Brazil	1,322 (32.6)	612 (26.1)	288 (38.5)	422 (44.0)
Race, N (%)[a]				
White	1,819 (44.9)	1,097 (46.7)	343 (45.9)	379 (39.5)
Black	633 (15.6)	425 (18.1)	777 (10.3)	131 (13.7)
Asian/Pacific Islander	111 (2.7)	91 (3.8)	8 (1.1)	12 (1.3)
American Indian/Alaska Native	80 (2.0)	47 (2.0)	18 (2.4)	15 (1.6)
Mixed/Other/Not Reported	1,411 (4.4)	686 (29.2)	302 (40.3)	423 (44.1)
Ethnicity, N (%)[a]				
Hispanic	1,828 (45.1)	932 (39.7)	373 (49.9)	523 (54.5)
Non-Hispanic	2,191 (54.1)	1,392 (59.3)	371 (49.6)	428 (44.6)
Not Reported/Refused	35 (0.8)	22 (0.94)	4 (0.5)	9 (0.9)
Circumcision, N (%)				
Yes	1,402 (34.6)	842 (35.9)	250 (33.4)	310 (32.3)
No	2,568 (63.3)	1,457 (62.1)	479 (64.0)	632 (65.8)
Partial	84 (2.1)	47 (2.0)	19 (2.5)	18 (1.9)
Lifetime number of vaginal sex partners[a]				
0	386 (9.5)	276 (11.8)	36 (4.8)	74 (7.7)
1	325 (8.0)	250 (10.7)	31 (4.1)	44 (4.6)
2 to 9	1,617 (39.9)	977 (41.7)	291 (38.9)	349 (36.4)
≥ 10	1,501 (37.0)	732 (31.2)	348 (46.5)	703 (43.8)
Refused	225 (5.6)	111 (4.7)	42 (5.6)	72 (7.5)
Total number of female partners last 3 months, N (%)[a]				
None	393 (9.7)	278 (11.9)	38 (5.1)	77 (8.0)
1	2,496 (61.6)	1,388 (59.2)	526 (70.3)	582 (60.6)
2	514 (12.7)	303 (12.9)	85 (11.4)	129 (13.4)

Characteristic	Overall (n = 4,054)	*Smoking Status*		
		Never (n = 2,346)	Former (n = 748)	Current (n = 960)
≥ 3	529 (13.1)	309 (13.2)	81 (10.8)	139 (14.5)
Refused	119 (2.9)	68 (2.9)	18 (2.4)	33 (3.4)

[a] $P < 0.05$ comparing never, former, current. The chi^2 test was used to test for differences by smoking status for categorical variables and analysis of variance (ANOVA) was used for continuous variables (i.e., age).

Cancer Epidemiol Biomarkers Prev. Author manuscript; available in PMC 2013 January 01.

Table 2

Prevalence of HPV infection by smoking status

		Smoking Status			
	Overall	**Never**	**Former**	**Current**	***P*-value**
	No. = 4,054	**No. = 2,348**	**No. = 748**	**No. = 960**	
HPV Status[a]					
Negative	1,435 (34.9)	861 (36.7)	249 (33.3)	305 (31.8)	
Positive for[b]:					
Any HPV (HPV Positive by PCR or Genotyping)[c]	2,639 (65.1)	1,485 (63.2)	499 (66.7)	655 (68.2)	**0.015**
At least 1 oncogenic type[c]	1,226 (30.2)	664 (28.3)	228 (30.5)	334 (34.8)	**< 0.001**
Only non-oncogenic types[c]	845 (20.8)	475 (20.3)	157 (20.9)	213 (22.2)	**0.009**
Only oncogenic types	476 (11.7)	269 (11.5)	91 (12.2)	116 (12.1)	0.241
Non-oncogenic types	1,595 (39.3)	870 (37.1)	294 (39.3)	431 (44.9)	**0.001**
> 1 genotype	1,142 (28.2)	605 (25.8)	208 (27.8)	329 (34.3)	**< 0.001**
Oncogenic and non-oncogenic genotypes	750 (18.5)	395 (16.8)	137 (18.3)	218 (22.7)	**< 0.001**
At least one genotype	2,071 (51.1)	1,139 (48.6)	385 (51.5)	547 (56.9)	**< 0.001**
Type 6, 11, 16, or 18	624 (15.4)	333 (14.2)	108 (14.4)	183 (19.1)	**0.001**
HPV 16 Only	305 (7.5)	161 (6.9)	53 (7.1)	91 (9.5)	**0.006**

[a]The percentages presented are prevalence using the total number of subjects by smoking status strata as the denominator (e.g., for any HPV positivity among never smokers: 1,485/2,348 = 63.2%)

[b]P-values were calculated by a chi^2 test for the distribution of HPV positivity compared to HPV negativity by smoking status

[c]Any HPV, Oncogenic HPV, and Only non-oncogenic HPV were used as the dependent variables for the analyses in Tables 3 to 5. Any HPV positivity is defined as HPV-positive by PCR or tested positive for at least one genotype. Oncogenic HPV positivity was defined as possessing at least one of the 13 oncogenic types tested for (HPV types 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, and 66), and included men infected with both oncogenic and non-oncogenic types. Non-oncogenic HPV positivity included single or multiple infections with only non-oncogenic HPV types (6, 11, 26, 40, 42, 53, 54, 55, 61, 62, 64, 67–73, 81–84, IS39, and CP6108).

Table 3

Prevalence of HPV infection by smoking status and tertiles[b] of smoking characteristics

	Former smokers							Current smokers						
HPV Status	**T_1**		**T_2**		**T_3**		**P-value[a]**	**T_1**		**T_2**		**T_3**		***P*-value[a]**
Total	***N = 255***		***N = 258***		***N = 215***			***N = 320***		***N = 352***		***N = 275***		
Tertiles cigarettes smoked/day														
Negative	97	(38.0%)	86	(37.7%)	62	(28.8%)		107	(33.4%)	117	(33.2%)	79	(28.7%)	
Any HPV	158	(62.0%)	172	(75.4%)	153	(71.2%)	0.108	213	(66.6%)	235	(66.8%)	196	(71.3%)	0.386
Oncogenic HPV	73	(28.6%)	80	(35.1%)	68	(31.6%)	0.265	103	(32.2%)	128	(36.4%)	96	(34.9%)	0.520
Only non-oncogenic HPV	48	(18.8%)	53	(21.5%)	50	(23.3%)	0.168	74	(23.1%)	68	(19.3%)	67	(24.4%)	0.244
Total	***N = 240***		***N = 249***		***N = 243***			***N = 142***		***N = 321***		***N = 479***		
Tertiles of years smoked														
Negative	89	(37.1%)	91	(36.5%)	65	(26.7%)		50	(35.2%)	96	(29.9%)	156	(32.6%)	
Any HPV	151	(62.9%)	158	(63.5%)	178	(73.3%)	**0.025**	92	(64.8%)	225	(70.1%)	323	(67.4%)	0.500
Oncogenic HPV	67	(27.9%)	74	(29.7%)	81	(33.3%)	0.064	46	(32.4%)	124	(38.6%)	155	(32.4%)	0.235
Only non-oncogenic HPV	52	(21.7%)	43	(17.3%)	58	(23.9%)	**0.041**	25	(14.6%)	69	(21.5%)	114	(23.8%)	0.363
Total	***N = 262***		***N = 233***		***N = 222***			***N = 215***		***N = 359***		***N = 362***		
Tertiles of pack-years														
Negative	95	(36.3%)	85	(36.5%)	61	(27.5%)		75	(34.9%)	121	(33.7%)	104	(28.7%)	
Any HPV	167	(63.7%)	148	(63.5%)	161	(72.5%)	**0.066**	140	(65.1%)	217	(66.3%)	225	(71.3%)	0.497
Oncogenic HPV	79	(30.2%)	68	(29.2%)	72	(32.4%)	0.199	71	(33.0%)	141	(34.5%)	111	(35.4%)	0.602
Only non-oncogenic HPV	51	(19.5%)	45	(19.3%)	52	(23.4%)	0.117	44	(31.0%)	85	(26.5%)	77	(16.1%)	0.617

[a] P-values were calculated by a chi^2 test for the distribution comparing each HPV positivity category (i.e., “Any HPV”, “Oncogenic HPV”, and “Only non-oncogenic HPV”) to HPV negativity.

[b] Tertiles were defined by overall smoking characteristic distributions by smoking status.

Table 4

Risk of HPV infection by smoking characteristics

	Any HPV		Oncogenic HPV		Only non-oncogenic HPV	
	mOR (95% CI)[a]	**mOR (95% CI)[b]**	**mOR (95% CI)[a]**	**mOR (95% CI)[b]**	**mOR (95% CI)[a]**	**mOR (95% CI)[b]**
Smoking Status						
Never smoker	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)
Former smoker	1.14 (0.95 – 1.37)	1.07 (0.88 – 1.29)	1.14 (0.95 – 1.38)	1.06 (0.87 – 1.29)	0.96 (0.77 – 1.18)	0.95 (0.77 – 1.18)
Current smoker	**1.32 (1.12 – 1.56)**	**1.19 (1.01 – 1.41)**	**1.42 (1.21 – 1.68)**	**1.24 (1.05 – 1.47)**	1.10 (0.91 – 1.33)	1.05 (0.87 – 1.27)
Pack-years smoked						
Never smokers	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)
< 5 pack-years (current smokers)	**1.25 (1.01 – 1.54)**	1.13 (0.92 – 1.40)	**1.35 (1.09 – 1.67)**	1.19 (0.96 – 1.48)	1.10 (0.86 – 1.40)	1.05 (0.82 – 1.35)
≥ 5 pack-years (current smokers)	**1.39 (1.09 – 1.77)**	1.23 (0.96 – 1.57)	**1.52 (1.21 – 1.91)**	**1.29 (1.02 – 1.64)**	1.11 (0.86 – 1.44)	1.06 (0.82 – 1.38)
Never smokers	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)	1.0 (ref)
< 5 pack-years (former smokers)	1.06 (0.86 – 1.31)	0.99 (0.80 – 1.23)	1.10 (0.88 – 1.38)	1.04 (0.83 – 1.31)	0.95 (0.74 – 1.23)	0.94 (0.73 – 1.21)
≥ 5 pack-years (former smokers)	1.30 (0.97 – 1.75)	1.16 (0.85 – 1.57)	1.20 (0.90 – 1.60)	1.08 (0.81 – 1.46)	0.92 (0.67 – 1.26)	0.89 (0.65 – 1.24)

[a] mOR = multivariable odds ratio adjusted for age and country

[b] mOR = multivariable odds ratio adjusted for age, race, ethnicity, country, circumcision, total number of female partners in the last 3 months, and total number of vaginal sex partners

Table 5

Risk of HPV infection by sexual activity and smoking status

	Lifetime number of female sex partners						
	0		**1 to 9**		**≥ 10**		$P_{\text{for interaction}}$
	mOR (95% CI)[a]	**mOR (95% CI)[b]**	**mOR (95% CI)[a]**	**mOR (95% CI)[b]**	**mOR (95% CI)[a]**	**mOR (95% CI)[b]**	
Any HPV by Number of female partners							
Never smokers	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	
Former smokers	1.32 (0.61 – 2.85)	1.48 (0.65 – 3.34)	1.11 (0.85 – 1.43)	1.08 (0.83 – 1.40)	0.87 (0.64 – 1.17)	0.83 (0.61 – 1.13)	0.123
Current smoker	1.29 (0.76 – 2.21)	1.31 (0.76 – 2.27)	**1.38 (1.08 – 1.77)**	**1.35 (1.05 – 1.73)**	0.94 (0.71 – 1.25)	0.93 (0.98 – 1.23)	0.201
Oncogenic HPV by Number of female partners							
Never smokers	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	
Former smokers	1.40 (0.62 – 3.11)	1.51 (0.65 – 3.48)	1.14 (0.84 – 1.55)	1.14 (0.84 – 1.55)	0.93 (0.71 – 1.22)	0.91 (0.69 – 1.20)	0.505
Current smoker	1.31 (0.72 – 2.39)	1.34 (0.72 – 2.47)	**1.46 (1.10 – 1.92)**	**1.46 (1.11 – 1.92)**	1.05 (0.82 – 1.35)	1.04 (0.81 – 1.34)	0.292
Only non-oncogenic HPV							
Never smokers	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	*1.0 (ref)*	
Former smokers	1.10 (0.48 – 2.70)	1.14 (0.45 – 2.84)	0.81 (0.58 – 1.14)	0.80 (0.57 – 1.12)	0.89 (0.67 – 1.22)	0.89 (0.65 – 1.21)	0.801
Current smoker	0.77 (0.37 – 1.64)	0.80 (0.38 – 1.70)	1.14 (0.84 – 1.54)	1.11 (0.82 – 1.51)	1.00 (0.76 – 1.33)	1.00 (0.76 – 1.33)	0.684

[a] mOR = multivariable odds ratio adjusted for age and country

[b] mOR = multivariable odds ratio adjusted for age, race, ethnicity, country, and circumcision