HHS Public Access

Author manuscript

J Med Virol. Author manuscript; available in PMC 2019 November 01.

Published in final edited form as:

J Med Virol. 2018 November ; 90(11): 1765–1774. doi:10.1002/jmv.25268.

Intravaginal Practices and Genital Human Papillomavirus Infection among Female Sex Workers in Cambodia

Thanh Cong Bui[a], **Michael E. Scheurer**[b], **Vy Thi-Tuong Pham**[c], **Ly Thi-Hai Tran**[d], **Leng Bun Hor**[e], **Damon J. Vidrine**[a], **Michael W. Ross**[f], and **Christine M. Markham**[g]

[a]Stephenson Cancer Center, Department of Family and Preventive Medicine, The University of Oklahoma Health Sciences Center.

[b]Department of Pediatrics, Dan L. Duncan Cancer Center, Baylor College of Medicine.

[c]Department of Epidemiology, College for Public Health and Social Justice, Saint Louis University.

[d]Department of Epidemiology, School of Public Health, University of Texas Health Science Center at Houston.

[e]Cambodian National AIDS Authority.

[f]Department of Family Medicine and Community Health, Medical School, University of Minnesota.

[g]Department of Health Promotion and Behavioral Sciences, School of Public Health, University of Texas Health Science Center at Houston.

Abstract

Objectives: Intravaginal practices (IVPs) include washing, wiping, or inserting something inside the vagina. This study investigates the associations between IVPs and genital human papillomavirus (HPV) infection.

Methods: We conducted a cross-sectional study of 200 female sex workers aged 18–35 years in Phnom Penh, Cambodia from August–September 2014. Data on sociodemographic characteristics, IVPs, and other behaviors were collected through face-to-face interviews. Self-collected cervicovaginal specimens were tested for 37 HPV genotypes.

Results: Multivariable Poisson regression models showed that a lower number of infecting HPV genotypes were associated with intravaginal washing in the past 3 months (incident rate ratios [IRR] = 0.65, 95% confidence interval [CI]: 0.46–0.94) and often performing intravaginal washing

Corresponding author: Thanh Cong Bui, MD, DrPH; Assistant Professor of Research; Stephenson Cancer Center, Department of Family and Preventative Medicine, The University of Oklahoma Health Sciences Center, Oklahoma City, Oklahoma, 73104, United States of America; thanh-c-bui@ouhsc.edu, thanh.bui@aya.yale.edu. T: +1-405-271-8001 ext. 50559, F: +1-405-271-2808.

Authors' contributions: TCB, MWR, and CMM developed the research concept and applied for funding. TCB, VTP, LTT, and LBH collected data. MES provided advice for biological specimen collection and performed HPV genotyping tests. TCB and LTT analyzed and interpreted the data, with advice and support from DJV. TCB and DJV were major contributors in writing the manuscript. All authors contributed to reviewing and editing the manuscript. All authors read and approved the final manuscript.

Conflict of Interests: All authors declare that they have no conflict of interests.

Ethics approval and consent to participate:
The study was approved by institutional review boards of the Cambodian National AIDS Authority (No. 132 NAA, 2013) and of The University of Texas Health Science Center at Houston (HSC-SPH-13–0481). All women agreed to participate and provided written consent.

shortly after sex (IRR = 0.89, 95% CI: 0.81–0.99). Intravaginal washing before vaginal sex, intravaginal wiping, and intravaginal insertion were not associated with HPV infection.

Conclusion: These findings challenge the existing view that all types of vaginal cleansing are harmful. Specifically, intravaginal washing shortly after sex (mainly with water) may help prevent HPV infection in female sex workers, who have several partners and thus frequently expose to sources of HPV infection with different genotypes.

Keywords

human papillomavirus; intravaginal practices; douching; female sex workers; Cambodia

INTRODUCTION

Intravaginal practices (IVP) include intravaginal cleaning (e.g., douching or washing with liquids), intravaginal wiping (e.g., with a cloth or tissue), or intravaginal insertion of substances to dry or tighten the vagina for sexual pleasure.[1,2] IVP types, prevalence, products, frequency, and motivation vary markedly across countries.[3,4] Potential health outcomes of douching have been examined in several studies; yet, the effects of other types of IVPs have been underexplored. Most studies of douching, conducted primarily in United States (US) and African populations, suggested that douching was associated with adverse health outcomes,[5,6] including increased risk of bacterial vaginosis,[7–10] pelvic inflammatory disease,[11–14] bacterial sexually transmitted infections (STIs),[12,15–18] non-regression of low-grade squamous intraepithelial lesion,[19] and cervical cancer.[14] Nevertheless, some other studies reported no associations between douching and these outcomes, such as bacterial vaginosis,[20,21] pelvic inflammatory disease,[22,23] or bacterial STIs.[9,23,24] The discrepancies between these studies were explained as partially due to the types of products used for douching, frequency, and timing (e.g., in relation to sexual activity or menses).[6] Similarly, some studies of the various types of IVPs (i.e., not only douching) found no association between IVPs and bacterial vaginosis[25,26] or STIs.[27] Recent systematic review and meta-analyses showed that some IVPs (e.g., intravaginal cleaning with soap or intravaginal use of cloth or paper) were associated with HIV infection but other IVPs (e.g., intravaginal cleaning with water) were not.[1,28] Published studies on IVPs, however, differed widely in their definitions and measurement of IVPs; thus, the associations between a particular form of IVP and health outcomes might have been masked. In general, studies examining the effect of IVPs other than douching on vaginal health, particularly in Asian countries, are scarce.

Genital human papillomavirus (HPV) infection is the most common STIs in the world.[29] Twelve HPV types have been identified as carcinogenic, and 13 others are classified as probably or possibly carcinogenic to humans (i.e., Groups 2A and 2B), hereafter referred to as high-risk types.[30] These caused six types of cancer: cervix, penis, vulva, vagina, anus, and oropharynx.[29] Nevertheless, very little is known about the associations between IVPs and cervicovaginal HPV infection; the few studies that investigated this association generated contrary results. Some studies showed that douching was associated with increased risk of HPV positivity of any genotype,[31,32] HPV infection with multiple genotypes,[33] or HPV 16 redetection in follow-ups.[34] Meanwhile, other studies suggested that douching was

associated with a lower likelihood of genital warts,[15] HPV infection with types 6 or 11,[35] or HPV positivity of any type.[36] Vulnerability to HPV or to other viral infections has been hypothesized to result from physical abrasions or disruption of the vaginal stratified squamous epithelium, caused by equipment (e.g., a douching device), materials (e.g., cloth), or substances (e.g., herbs) used in IVPs.[5,37] Vaginal douching or wiping may also disturb local innate immunity or remove cervicovaginal mucus secretions that serve as a protective barrier against HPV.[38] Some chemicals used in intravaginal cleansing (e.g., soaps, detergents, or antiseptics) may cause epithelial damage, increase vaginal pH, facilitate bacterial vaginosis, and thus facilitate viral infections.[5] In contrast, intravaginal cleansing, particularly after sexual intercourse, may help to clear the transmitted HPV and reduce infection risk. Therefore, further evidence regarding the association between IVPs and HPV infection is needed. It is also important to investigate this association with specific to types of IVPs, solutions or substances used, and timing of the practice.

In Cambodia, cervical cancer ranks first of all cancers in women.[39] As of 2012, there were no national organized programs for cervical cancer screening or HPV vaccination.[39,40] Moreover, there is no nationally representative estimate of HPV prevalence in Cambodian women. In Cambodian female sex workers (FSWs), the cervical HPV prevalence was 41.1% for any-type HPV positivity, and 23.3% for infection with at least one oncogenic type.[41] Vaginal douching appears to be common in Cambodia. Among women who visited maternal and child health care clinics, 76.7% douched at least once a week.[42] In FSWs, 91.0% ever douched and 49.4% thought that douching could help prevent STIs including HIV.[43] There is no report on the prevalence of other forms of IVPs in Cambodia.

Aim

This study aims to investigate the associations between IVPs and HPV infection among FSWs in Phnom Penh, Cambodia. Findings from this study will help fill the gaps in knowledge regarding IVPs' effects on HPV infection, as described above.

MATERIALS AND METHODS

Study Design and Participants

We used a cross-sectional design. Participants were recruited through convenience and snow-ball sampling techniques. Recruitment sites consisted of three participating Voluntary Confidential Counseling and Testing (for HIV) sites in Phnom Penh and the Cambodian Prostitute Union office, which provides support and services to FSWs. All women who came to these recruitment sites in August–September 2014 were screened for eligibility. Eligibility criteria included (i) biological female, (ii) 18–35 years old, (iii) fluent in Khmer (the main language in Cambodia), and (iv) having at least one transactional sex act (sex in exchange for money, goods, services, or drugs) in the past 3 months. Eligible women were invited to participate in the study. Recruited participants were then asked to refer other eligible women. Six eligible women refused to participate during initial recruitment contacts; reasons for refusing were being busy and not wanting to participate. The process continued until we reached our desired total sample size of 200.

Procedures

All eligible participants were scheduled for a face-to-face interview and vaginal specimen collection at one of the recruitment sites. When scheduling, research staff asked participants to ensure that they would not be menstruating on the scheduled dates, or they would be rescheduled. Participants were also told not to douche or clean inside the vagina for at least 12 hours before the interview. The study was approved by institutional review boards of the Cambodian National AIDS Authority (No. 132 NAA, 2013) and of The University of Texas Health Science Center at Houston (HSC-SPH-13–0481). All women agreed to participate and provided written consent.

The interviews lasted 30–40 minutes, were conducted in Khmer by local female social workers, and were structured to collect participants' demographic and behavioral data. Interviewers were female social workers who were staff members at the Cambodian Women's Development Agency, a local nongovernmental organization. Participants' responses were recorded on paper and then manually entered into SPSS Statistics software for analysis. No identifiers were collected; each participant was assigned a unique identification number. Each participant received $9 USD in compensation.

After the interviews, participants were instructed on the use of a self-sampling technique to collect a cervicovaginal specimen, which technique has been shown to be as effective and sensitive as physician-obtained sampling for detecting HPV DNA.[44] In a private women's restroom, each participant was instructed to introduce a cytobroom (in a ThinPrep Pap Test kit) into her vagina until it met with resistance, rotate the broom 3–5 times, withdraw the broom, put the broom into a PreservCyt solution vial, push the broom into the bottom of the vial about 10 times, swirl the broom vigorously to further release material, discard the broom, tighten the vial's cap, and hand the vial to the research staff. Research staff recorded the participant's unique identification number on the vial. At the end of each day, all specimens were brought to the research office in Cambodia and were stored at 4^{o}C in a refrigerator.

Variables and Measurements

Main exposure variables are IVPs. Definitions of IVPs used in our study were based on the IVP classification developed by the World Health Organization's Gender, Sexuality and Vaginal Practices Study Group,[1–3] and our previous qualitative work.[45] Specifically, *intravaginal washing* was explained to participants as washing inside their vagina by using fingers and/or a device to introduce a stream of water or solution. *Intravaginal wiping* meant to wipe inside the vagina by using a material (e.g., cotton, cloth, tissues) with little or no water or solution. *Intravaginal insertion* was described to participants as placing or applying something inside the vagina with the intent to dry or tighten the vagina, excluding the use of condoms, barrier contraceptives, and products for absorption of menstrual blood (e.g., tampons). We asked participants whether they had ever performed each type of IVP, whether they performed it within the past 3 months, and what solutions or substances were used. For intravaginal washing and intravaginal wiping, we asked participants how often they performed it shortly before or after sex in their lifetime, with responses on a 5-point Likert scale ranging from *1 = Never* to *5 = Every time having vaginal sex*.

Other variables collected through the interviews included demographic characteristics (age, education level), sex work characteristics (e.g., main venues to contact clients, number of different paying partners in the past 3 months, condom use in lifetime and in the past 3 months), other behavioral characteristics (cigarette smoking, alcohol use, drug use), and health status (HIV status, HPV vaccine completion). Condom use was measured on a 5-point Likert scale from *1 = Never* to *5 = Every time having vaginal sex*.

HPV infection was the main outcome. Collected cervicovaginal specimens were shipped to a laboratory at Baylor College of Medicine in US every 2 weeks. Specimens were tested for 37 HPV DNA genotypes by using Roche Linear Array HPV Genotyping. Of these 37 genotypes, 16 are classified as low-risk genotypes (6, 11, 40, 42, 54, 55, 61, 62, 64, 71, 72, 81, 82 subtype IS39, 83, 84, and 89) and 21 are classified as high-risk genotypes (16, 18, 26, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 67, 68, 69, 70, 73, and 82).[30] Although most HPV infections are asymptomatic,[46] HPV types 6 and 11 can cause genital warts and thus may result in more intravaginal cleansing to relieve symptoms. The inclusion of symptomatic HPV types 6 and 11 in the outcome may mask or confound the association between an IVP and other asymptomatic HPV infections. Therefore, we excluded HPV types 6 and 11 from our analyses. The main outcome variable was the number of all genital HPV DNA types detected, except types 6 and 11, which was a sum of positive results with all 35 genital HPV DNA types.

Statistical Analysis

To examine the associations between IVPs and the HPV outcome, we used Poisson regression models for count outcomes (i.e., number of all genital HPV DNA types detected) in both bivariate and multivariable analyses. Covariates included in multivariable models were selected based on a priori knowledge, criteria in the definitions of confounding effect or confounders,[47] and their actual associations with HPV outcome in bivariate analyses.

RESULTS

Table 1 displays participants' characteristics. Ninety percent of participants had ever performed intravaginal washing; of these 97.2% (175/180) had continued it in the past 3 months. Most participants (88.1%) started intravaginal washing before or within 2 years after engaging in sex work. Most participants (92.8%) used water and might add soap, salt, or lemon in intravaginal washing; about half of these also used commercial products sometimes. Twenty-nine percent had ever performed intravaginal wiping, and 5.5% had ever performed intravaginal insertion. Almost everyone (55/56, 98.2%) who had ever performed intravaginal wiping also had ever performed intravaginal washing. Participants mainly learned about intravaginal washing from health care professionals, including physicians and nurses (39.2%), other female sex workers (29.8%), and female relatives including mothers and sisters (29.8%). Only 17.2% (n = 30) reported that they had received information or consultation from health care professionals who had recommended that intravaginal washing or wiping not be performed. Detailed descriptive characteristics of IVPs can be found in Supplementary Table 1.

The overall prevalence of HPV infection with any type was 47.0%. The prevalence of HPV infection with one, two, three, and 4–8 types was 17.0%, 11.0%, 12.0%, and 7.0%, respectively. The most common types were HPV-62 (10.5%), HPV-16 (7.5%), HPV-18 (6.0%), HPV-52 (6.0%), HPV-53 (6.0%), and HPV-68 (6.0%); except for HPV-62, these were high-risk types. Among those who had any HPV type detected, 77.7% harbored at least one high-risk type. The prevalence of HPV infection with types 6 or 11, which were excluded in the main dependent outcome in the following analyses, was 2.1%. Ten percent had completed all required doses of HPV vaccines before data collection. Twelve percent reported being HIV-positive.

In bivariate analyses, being infected with a higher number of HPV types was associated with a higher number of years in sex work, a higher average number of drinks per week in the past 3 months, being HIV-positive, and a higher number of different partners in the past 3 months (Table 2). Completion of all required doses of the HPV vaccine was not associated with the number of types of HPV infection. Ever performing intravaginal washing, often performing intravaginal washing shortly after vaginal sex in lifetime, and a higher number of times performing intravaginal washing per week in the past 3 months were associated with a lower number of types of HPV infection; however, often performing intravaginal washing shortly before vaginal sex and types of solutions used were not. Intravaginal wiping and intravaginal insertion were not associated with HPV infection.

In multivariable Poisson regression models, performing intravaginal washing in the past 3 months and often performing intravaginal washing shortly after vaginal sex remained associated with a lower number of types of HPV infection, after adjusting for age, main venues to contact clients, self-reported HIV status, number of paying partners in the past 3 months, condom use with all paying partners in the past 3 months, and completion of all required doses of the HPV vaccine (Table 3). In a model similar to Model 2 in Table 3, intravaginal washing shortly *before* vaginal sex, in replacement of intravaginal washing shortly after sex, was not associated with the number of types of HPV infection ($P = .162$, data not shown). Similarly, when intravaginal washing was replaced by ever performed intravaginal wiping and ever performed intravaginal insertion, the number of types of HPV infection was not associated with these wiping and insertion practices ($P > .689$, data not shown). Also in models similar to Models 1 and 2 but in which the dependent variable was replaced by the number of all infecting HPV types *including* types 6 and 11, the effect of intravaginal washing on HPV outcome remained very similar, both in terms of statistical significance and point estimates. There was no interaction between intravaginal washing, HIV status, and completion of HPV vaccination.

DISCUSSION

Compared to Couture et al. (2012) study in a similar population,[41] our results show higher prevalence of HPV infection, including infection with any types (47.0% vs 41.1%), infection with multiple types (30.0% vs 16.4%), and infection with at least one high-risk type (77.7% vs 23.3%). These differences might be due to HPV tests used and high-risk HPV classifications. The HPV vaccination program in Cambodia is still in the pilot stage; thus, there is no current national HPV vaccination routine immunization program.[39,40] To our

knowledge, at the time of data collection for this study, access to HPV vaccines was limited and FSWs had to fully pay out-of-pocket for the vaccines. Some FSWs might have received a bivalent or quadrivalent HPV vaccine through their participation in previous health studies.

The most important finding of this study is that intravaginal washing shortly after sex might reduce the risk of HPV infection, whereas intravaginal washing shortly before sex had no effect. Performing intravaginal washing in the past 3 months was associated with a reduction of 35% in the incident rate of HPV infection with an additional type, and every level increase in often performing intravaginal washing shortly after sex was associated with a reduction of 11% in the incident rate of HPV infection with an additional type. As explained above, the association between douching and HPV infection was conflicting in a few previous studies on this topic. Our findings in this study also contradict results from our recent analysis of the 2003–2004 US National Health and Nutrition Examination Survey (NHANES), which showed that douching in the past 6 months was significantly associated with a higher number of all genital HPV types (relative risk ratio [RRR] = 1.26, 95% confidence interval [CI], 1.03–1.54) and with a higher number of HPV high-risk types (RRR = 1.40, 95% CI, 1.09–1.80), independent from other risk factors for HPV infection such as younger age or having multiple sexual partners.[33]

These differing results may be explained by several factors. First, the NHANES and other US studies often examined douching in general without specific focus on timing of the practice. In this Cambodian FSW study, we investigated IVPs specifically with regard to types of practices (e.g., washing separately from wiping) and timing (e.g., before versus after vaginal sex). Douching regularly in general or in preparation for sex (e.g., cleaning vaginal odor before sex) may facilitate HPV acquisition because it may clear cervicovaginal secretions that can serve as a protective mucus barrier, or it may cause microepithelial abrasions that would serve as an entry portal for HPV. Intravaginal washing shortly after sex, in contrast, may help clear the transmitted HPV viral loads. This supposition was supported by an in vitro study showing that washing within 30 minutes after HPV exposure, the approximate amount of time needed for HPV to attach to cells, could prevent 90.0% of HPV infection.[38] Second, most Cambodian FSWs used their fingers for intravaginal washing; very few used a commercial douching device as did US women. Thus, microabrasions of the vaginal epithelium might have occurred less frequently. Third, the inconsistent associations between douching and other adverse health outcomes in the US were partially explained by differences in race.[6] Several studies in the US that had a predominant proportion of African Americans reported no associations. Our Cambodian study had a much more homogeneous sample and thus was less likely to be influenced by race or ethnicity. Finally, our study focused on a FSW population who have many different clients and are frequently exposed to various HPV strains. Thus, intravaginal washing after sex in FSWs may have a beneficial net effect compared with douching among women in the general population, who have a significantly lower number of sexual partners.

This result needs to be viewed with caution. On one hand, if intravaginal washing shortly after sex can actually reduce HPV infection in populations at risk for STIs, albeit its inconsistent effect in the general population, this practice can supplement available HPV prevention options such as vaccination. Specifically, intravaginal washing shortly after sex

may substantially benefit FSWs who cannot receive HPV vaccines (e.g., due to inaccessibility or unaffordability) or are not eligible for HPV vaccination (e.g., out of the recommended ages for HPV vaccines). In HIV-positive FSWs who frequently expose to various HPV types yet their immunogenesis is compromised, the practice also may be beneficial with regard to reducing the likelihood of HPV acquisition with a new genotype. Moreover, because IVPs may be deeply embedded in sociocultural norms of womanhood or may become a favored habit, harm reduction instead of complete elimination of IVPs may be more practical.[45,48,49] So, intravaginal washing at different times (e.g., before sex, during menses) should be discouraged, but intravaginal washing after sex may not.

On the other hand, even if the net effect of intravaginal washing shortly after sex is beneficial to HPV prevention, recommendations of the practice need to be carefully considered. As aforementioned, although some studies suggested no association between douching and adverse health outcomes, numerous studies have shown harmful effects from douching. If intravaginal washing after sex is as potentially harmful as douching, the practice should not be supported. However, as explained above, most previous U.S. studies examined douching in general without specific focus on timing of and devices used in the practice. A longitudinal study is needed to investigate the effects of intravaginal washing after sex, separated from other IVPs, on multiple vaginal health outcomes. Moreover, the adverse health outcomes, particularly long-term ones such as low-/high-grade squamous intraepithelial lesions, may not necessarily be caused by the practice but by the chemical solutions used. Thus, it is also important to investigate the practice's effects with a homogeneous solution (e.g., water only or water with soap) used by all participants, or to conduct other studies that aim to specifically examine different pharmacologic effects of different solutions (e.g., homemade vs commercial).

Intravaginal wiping was not associated with the number of HPV types. This lack of association may be true but may also be due to some biases. Except in one case, all FSWs who performed intravaginal wiping also performed intravaginal washing. Thus, the effect of intravaginal wiping on HPV outcome might have been masked or confounded by the more common practice of intravaginal washing. Alternatively, intravaginal wiping might have similar benefit and harm as intravaginal washing; yet, because it might be more harmful than intravaginal washing (e.g., causing more epithelial micro-breaks), the net effect became neutral. Intravaginal insertion with the intent to dry or tighten the vagina for sexual pleasure is potentially harmful as suggested in previous studies[1] and thus may increase the vulnerability to HPV infection. Nevertheless, the low proportion of FSWs performing intravaginal insertion in our study sample might have led to reduced statistical power to detect the associations.

The study has some limitations. First, because the study design was cross-sectional, temporal relationships between intravaginal washing and HPV infection cannot be guaranteed. However, about 90% of HPV infections are cleared within 2–3 years,[50] so HPV infection detected in the study was recent or current. Meanwhile, most (88%) of FSWs started intravaginal washing before or within 2 years after engaging in sex work; and among those who started intravaginal washing, 97.2% continued it within the past 3 months. These statistics suggest that intravaginal washing had been long maintained and was unlikely

consequent to current HPV infection. Moreover, the infection of HPV types other than 6 and 11 is often asymptomatic and thus unlikely resulted in intravaginal washing. Nevertheless, the association between intravaginal washing and HPV infection may be indirect, e.g., those who had better medical knowledge might perform intravaginal washing and had other preventive behaviors that reduce the likelihood of HPV infection. Second, our study cannot distinguish between new HPV infection and HPV persistence. A longitudinal study is needed to address both of these limitations: examine the preconditions (i.e., at baseline) and investigate the effects of these, including IVPs, on new HPV infection versus redetection or persistence. Third, IVPs and variables collected through interviews might not be accurately reported. Finally, the moderate sample size might have reduced statistical power to detect some associations or interactions in multivariable models, and limited the ability for multiple comparisons (e.g., examining the effect of frequency of, timing of, and solutions used in intravaginal washing together).

CONCLUSION

Our findings suggest that intravaginal washing shortly after sex may help protect FSWs, who have a lot of different sexual partners, against HPV infection. Intravaginal washing shortly before sex and intravaginal wiping had no association with HPV infection. The effect of intravaginal insertion on HPV outcomes was inconclusive. Although IVP in general should be discouraged due to their harmful effects, intravaginal washing *after sex in sex workers* may not necessarily be so, particularly when it has culturally been a favored habit and cannot be completely eliminated. However, this needs to be carefully considered because of other potential adverse health outcomes of intravaginal washing. Future longitudinal studies are needed to comprehensively investigate the effects of IVP on multiple health outcomes, particularly long-term ones such as cervical intraepithelial neoplasia.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgement:

TCB was supported by a UTHealth Innovation for Cancer Prevention Research postdoctoral fellowship, grant RP101503 from the Cancer Prevention and Research Institute of Texas, and a faculty fellowship from The University of Texas MD Anderson Cancer Center's Duncan Family Institute for Cancer Prevention and Risk Assessment. TCB, DJV, and the preparation of this manuscript is also supported in part by a grant from the Oklahoma Tobacco Settlement Endowment Trust, 092–016-0002 (PI: Vidrine). We thank Cambodian National AIDS Authority, Coalition to Address Sexual Exploitation of Children in Cambodia (COSECAM), and Cambodian Women's Development Agency for their support with data collection and fieldwork supervision.

Funding: This work was supported by a 2013 Developmental Grant from the Baylor-UTHouston Center for AIDS Research (CFAR), an NIH-funded program (AI036211).

List of abbreviations

IVP	intravaginal practices
FSW	female sex workers
HPV	human papillomavirus

HIV	human immunodeficiency virus
STI	sexually transmitted infections
IRR	incident rate ratio

References

1. Low N, Chersich MF, Schmidlin K, et al. Intravaginal practices, bacterial vaginosis, and HIV infection in women: Individual participant data meta-analysis. PLoS Medicine. 2011;8(2):e1000416. [PubMed: 21358808]
2. Hilber AM, Chersich MF, van de Wijgert JHHM, Rees H, Temmerman M Vaginal practices, microbicides and HIV: What do we need to know? Sex Transm Infect 2007;83(7).
3. Hull T, Hilber AM, Chersich MF, et al. Prevalence, motivations, and adverse effects of vaginal practices in Africa and Asia: Findings from a multicountry household survey. J Womens Health 2011;20(7):1097–1109.
4. Hilber AM, Hull TH, Preston-Whyte E, et al. A cross cultural study of vaginal practices and sexuality: Implications for sexual health. Soc Sci Med 2010;70(3):392–400. [PubMed: 19906477]
5. Cottrell BH. An updated review of evidence to discourage douching. MCN Am J Matern Child Nurs 2010;35(2):102–107. [PubMed: 20215951]
6. DiClemente RJ, Young AM, Painter JL, Wingood GM, Rose E, Sales JM. Prevalence and correlates of recent vaginal douching among African American adolescent females. J Pediatr Adolesc Gynecol 2012;25(1):48–53. [PubMed: 22051790]
7. Klebanoff MA, Nansel TR, Brotman RM, et al. Personal hygienic behaviors and bacterial vaginosis. Sex Transm Dis 2010;37(2):94–99. [PubMed: 19823112]
8. Koumans EH, Sternberg M, Bruce C, et al. The prevalence of bacterial vaginosis in the United States, 2001–2004; associations with symptoms, sexual behaviors, and reproductive health. Sex Transm Dis 2007;34(11):864–869. [PubMed: 17621244]
9. Ness RB, Hillier SL, Richter HE, et al. Douching in relation to bacterial vaginosis, lactobacilli, and facultative bacteria in the vagina. Obstet Gynecol 2002;100(4):765–772. [PubMed: 12383547]
10. Rajamanoharan S, Low N, Jones SB, Pozniak AL. Bacterial vaginosis, ethnicity, and the use of genital cleaning agents: a case control study. Sex Transm Dis 1999;26(7):404–409. [PubMed: 10458635]
11. Critchlow CW, Wolner-Hanssen P, Eschenbach DA, et al. Determinants of cervical ectopia and of cervicitis: age, oral contraception, specific cervical infection, smoking, and douching. Am J Obstet Gynecol 1995;173(2).
12. Scholes D, Stergachis A, Ichikawa LE, Heidrich FE, Holmes KK, Stamm WE. Vaginal douching as a risk factor for cervical Chlamydia trachomatis infection. Obstetric & Gynecology. 1998;91(6): 993–997.
13. Wolner-Hanssen P, Eschenbach DA, Paavonen J, et al. Association between vaginal douching and acute pelvic inflammatory disease. JAMA. 1990;263(14):1936–1941. [PubMed: 2313870]
14. Zhang J, Thomas AG, Leybovich E. Vaginal douching and adverse health effects: a meta-analysis. Am J Public Health. 1997;87(7):1207–1211. [PubMed: 9240115]
15. La Ruche G, Messou N, Ali-Napo L, et al. Vaginal douching: Association with lower genital tract infections in African pregnant women. Sex Transm Dis 1999;26(4):191–196. [PubMed: 10225584]
16. Tsai CS, Shepherd BE, Vermund SH. Does douching increase risk for sexually transmitted infections? A prospective study in high-risk adolescents. Am J Obstet Gynecol 2009;200(1): 38.e31–38.e38. [PubMed: 18667177]
17. Peters SE, Beck-Sague CM, Farshy CE, et al. Behaviors associated with Neisseria gonorrhoeae and Chlamydia trachomatis: cervical infection among young women attending adolescent clinics. Clin Pediatr 2000;39(3):173–177.
18. Thurman AR, Holden AE, Shain RN, Perdue S, Piper JM. Preventing recurrent sexually transmitted diseases in minority adolescents: a randomized controlled trial. Obstet Gynecol 2008;111(6):1417–1425. [PubMed: 18515527]

19. Chu TY, Hsiung CA, Chen CA, et al. Post-coital vaginal douching is risky for non-regression of low-grade squamous intraepithelial lesion of the cervix. Gynecol Oncol 2011;120(3):449–453. [PubMed: 21145098]
20. Uscher-Pines L, Hanlon AL, Nelson DB. Racial differences in bacterial vaginosis among pregnant women: the relationship between demographic and behavioral predictors and individual BV-related microorganism levels. Matern Child Health J 2009;13(4):512–519. [PubMed: 18543090]
21. Vermund SH, Sarr M, Murphy DA, et al. Douching practices among HIV infected and uninfected adolescents in the United States. J Adolesc Health. 2001;29(3).
22. Rothman KJ, Funch DP, Alfredson T, Brady J, Dreyer NA. Randomized field trial of vaginal douching, pelvic inflammatory disease and pregnancy. Epidemiology. 2003;14(3):340–348. [PubMed: 12859036]
23. Ness RB, Hillier SL, Kip KE, et al. Douching, pelvic inflammatory disease, and incident gonococcal and chlamydial genital infection in a cohort of high-risk women. Am J Epidemiol 2005;161(Generic):186–195. [PubMed: 15632269]
24. Fonck K, Kaul R, Keli F, et al. Sexually transmitted infections and vaginal douching in a population of female sex workers in Nairobi, Kenya. Sex Transm Infect 2001;77(4):271–275. [PubMed: 11463927]
25. Esber A, Moyo P, Munjoma M, et al. Cessation of intravaginal practices to prevent bacterial vaginosis: a pilot intervention in Zimbabwean women. Sex Transm Infect 2015;91(3):183–188. [PubMed: 25355772]
26. Masese L, McClelland RS, Gitau R, et al. A pilot study of the feasibility of a vaginal washing cessation intervention among Kenyan female sex workers. Sex Transm Infect 2013;89(3):217–222. [PubMed: 23002190]
27. Myer L, Denny L, De Souza M, Barone MA, Wright TC, Kuhn L, Jr. Intravaginal practices, HIV and other sexually transmitted diseases among South African women. Sex Transm Dis 2004;31(3): 174–179. [PubMed: 15076931]
28. Hilber AM, Francis SC, Chersich M, et al. Intravaginal Practices, Vaginal Infections and HIV Acquisition: Systematic Review and Meta-Analysis. PLoS ONE 2010;5(2):e9119. [PubMed: 20161749]
29. Forman D, de Martel C, Lacey CJ, et al. Global burden of human papillomavirus and related diseases. Vaccine. 2012;30, Supplement 5(0):F12–F23. [PubMed: 23199955]
30. International Agency for Research on Cancer. IARC monographs on the evaluation of carcinogenic risks to humans: Biological agents - Volume 100B. Vol 100B. Lyon, France: International Agency for Research on Cancer; 2012.
31. Sun C-A, Hsiung CA, Lai C-H, et al. Epidemiologic correlates of cervical human papillomavirus prevalence in women with abnormal Pap smear tests: A Taiwan cooperative oncology group (TCOG) study. J Med Virol 2005;77(2):273–281. [PubMed: 16121376]
32. Tarkowski TA, Koumans EH, Sawyer M, et al. Epidemiology of human papillomavirus infection and abnormal cytologic test results in an urban adolescent population. J Infect Dis 2004;189(1): 46–50. [PubMed: 14702152]
33. Bui TC, Thai TN, Tran LT, Shete SS, Ramondetta LM, Basen-Engquist KM. Association between vaginal douching and genital human papillomavirus infection among women in the United States. J Infect Dis 2016;214(9):1370–1375. [PubMed: 27553042]
34. Moscicki A-B, Ma Y, Farhat S, et al. Redetection of cervical human papillomavirus type 16 (HPV16) in women with a history of HPV16. J Infect Dis 2013;208(3):403–412. [PubMed: 23599313]
35. Low A, Didelot-Rousseau MN, Nagot N, et al. Cervical infection with human papillomavirus (HPV) 6 or 11 in high-risk women in Burkina Faso. Sex Transm Infect 2010;86(5):342–344. [PubMed: 20410079]
36. Lee H, Lee D-H, Song Y-M, Lee K, Sung J, Ko G. Risk factors associated with human papillomavirus infection status in a Korean cohort. Epidemiol Infect 2014;142(08):1579–1589. [PubMed: 24139383]

37. Kilmarx PH, Limpakarnjanarat K, Supawitkul S, et al. Mucosal disruption due to use of a widely-distributed commercial vaginal product: potential to facilitate HIV transmission. AIDS. 1998;12(7):767–773. [PubMed: 9619809]
38. Chu TY, Chang YC, Ding DC. Cervicovaginal secretions protect from human papillomavirus infection: effects of vaginal douching. Taiwan J Obstet Gynecol 2013;52(2):241–245. [PubMed: 23915858]
39. Bruni L, Barrionuevo-Rosas L, Albero G, et al. Human papillomavirus and related diseases report: Cambodia. ICO Information Centre on HPV and Cancer (HPV Information Centre); 12 23rd, 2015 2015.
40. Eav S, Schraub S, Dufour P, Taisant D, Ra C, Bunda P. Oncology in Cambodia. Oncology. 2012;82(5):269–274. [PubMed: 22538443]
41. Couture MC, Page K, Stein E, et al. Cervical human papillomavirus infection among young women engaged in sex work in Phnom Penh, Cambodia: Prevalence, genotypes, risk factors and association with HIV infection. BMC Infect Dis 2012;12(1):166. [PubMed: 22839728]
42. Heng LS, Yatsuya H, Morita S, Sakamoto J. Vaginal douching in Cambodian women: Its prevalence and association with vaginal candidiasis. J Epidemiol 2010;20(1):70–76. [PubMed: 20009371]
43. Bui TC, Tran LT, Ross MW, Markham CM. Douching practices among female sex workers in Phnom Penh, Cambodia. Int J STD AIDS. 2015;26(4):238–242. [PubMed: 24810219]
44. Petignat P, Faltin DL, Bruchim I, Tramer MR, Franco EL, Coutlee F. Are self-collected samples comparable to physician-collected cervical specimens for human papillomavirus DNA testing? A systematic review and meta-analysis. Gynecol Oncol 2007;105(2):530–535. [PubMed: 17335880]
45. Bui TC, Tran LT, Hor LB, Scheurer ME, Vidrine DJ, Markham CM. Intravaginal practices in female sex workers in Cambodia: A qualitative study. Arch Sex Behav 2016;45(4):935–943. [PubMed: 26742508]
46. Doorbar J, Quint W, Banks L, et al. The biology and life-cycle of human papillomaviruses. Vaccine. 2012;30, Supplement 5(0):F55–F70. [PubMed: 23199966]
47. Greenland S, Pearl J, Robins JM. Causal diagrams for epidemiologic research. Epidemiology. 1999;10(1):37–48. [PubMed: 9888278]
48. Lees S, Zalwango F, Andrew B, et al. Understanding motives for intravaginal practices amongst Tanzanian and Ugandan women at high risk of HIV infection: The embodiment of social and cultural norms and well-being. Soc Sci Med 2014;102(0):165–173. [PubMed: 24565154]
49. Hilber AM, Kenter E, Redmond S, et al. Vaginal practices as women's agency in Sub-Saharan Africa: A synthesis of meaning and motivation through meta-ethnography. Soc Sci Med 2012;74(9):1311–1323. [PubMed: 22401645]
50. Moscicki A-B, Schiffman M, Burchell A, et al. Updating the natural history of human papillomavirus and anogenital cancers. Vaccine. 2012;30, Supplement 5(0):F24–F33. [PubMed: 23199964]

Table 1.

Descriptive Statistics of Participants' Demographic and Behavioral Characteristics

Characteristics	N	%
Age (years) (mean [SD])	26.7 [4.5]	
Ethnicity		
Khmer	196	98.0
Chinese	1	0.5
Vietnamese	3	1.5
Religious beliefs		
Buddhism	194	97.0
Catholics	1	0.5
Other	1	0.5
No religion	4	2.0
Education level		
Did not attend school	51	25.5
Some elementary school	64	32.0
Completed elementary school or secondary school	71	35.5
Completed secondary school or higher	14	7.0
Age at which first engaged in traded sex, (mean [SD])	20.9 [3.8]	
Years in sex work (mean [SD])	6.0 [4.3]	
Average income per month in the past year from all types of sex work in USD (median [interquartile range])	200 [150]	
Main venue to contact clients		
Establishment-based (e.g., brothels, massage parlors, karaoke parlors)	59	29.5
Freelancers (e.g., at bars, beer gardens)	62	31.0
Street-based	79	39.5
Cigarette smoking		
Never	162	81.0
Former	1	0.5
Current (smoked in the past 3 months)	24	12.0
Alcohol use in past 3 months, average no. drinks/week		
0	37	18.5
<3	48	24.0
3 to <7	17	8.5
7 to <14	66	33.0
≥14	32	16.0
Ever used drugs		
No	117	58.5
Yes, but never injected	81	40.5
Yes, ever injected	2	1.0
Self-reported HIV status		
Negative	176	88.0
Positive	23	11.5

Characteristics	N	%
Completed all required doses of HPV vaccines		
No	179	89.5
Yes	21	10.5
Had a non-paying regular partner (e.g., husband, boyfriend) in the past 3 months		
Yes	38	19.0
No	153	76.5
No. of different paying partners in the past 3 months		
≤30	63	31.5
31–60	63	31.5
61–90	24	12.0
91–180	30	15.0
>180	19	99.5
Condom use with all partners in lifetime		
Never	0	0.0
Less than 1/2 the times having vaginal sex	2	1.0
About 1/2 the times having vaginal sex	7	4.5
More than 1/2 the times having vaginal sex	103	51.5
Every time having vaginal sex	87	43.5
Condom use with all paying partners in the past 3 months		
Never	48	24.0
Less than 1/2 the times having vaginal sex	6	3.0
About 1/2 the times having vaginal sex	5	2.5
More than 1/2 the times having vaginal sex	27	13.5
Every time having vaginal sex	113	56.5

Table 2.

Bivariate Associations Between Participants' Characteristics, Intravaginal Practices, and HPV Infection

Characteristics	Number of infecting HPV genotypes, excluding HPV 6&11	
	IRR (95% CI)	**p-values (p-trend)**
Age	0.98 (0.950–1.01)	.218
Education level		(.390)
Did not attend school	1	
Some elementary school	0.89 (0.62–1.23)	.532
Completed elementary school or some secondary school	1.10 (0.78–1.53)	.591
Completed secondary school or higher	0.69 (0.36–1.31)	.258
Years in sex work	0.95 (0.92–0.98)	.004
Main venue to contact clients		
Establishment-based (e.g., brothels, massage parlors, karaoke parlors)	1	
Freelancers (e.g., at bars, beer gardens)	0.69 (0.47–1.00)	.050
Street-based	1.14 (0.84–1.56)	.394
Cigarette smoking		
Never	1	
Former/current	0.86 (0.56–1.31)	.855
Alcohol use in past 3 months, average no. drinks/week		(<.001)
0	1	
<3	1.24 (0.87–1.77)	.235
3 to <7	0.18 (0.06–0.51)	.001
7 to <14	0.67 (0.45–0.97)	.036
≥14	0.59 (0.36–0.96)	.033
Ever used drugs		
No	1	
Yes	1.02 (0.77–1.33)	.916
Self-reported HIV status		
Negative	1	
Positive	1.58 (1.11–2.24)	.011

Characteristics	Number of infecting HPV genotypes, excluding HPV 6&11	
	IRR (95% CI)	p-values (p-trend)
Completed all required doses of HPV vaccines		
No	1	
Yes	0.65 (0.38–1.10)	.106
No. of different paying partners in the past 3 months		(.001)
≤30	1	
31–60	1.78 (1.27–2.47)	.001
61–90	1.01 (0.61–1.67)	.961
91–180	0.81 (0.50–1.33)	.413
>180	1.23 (0.74–2.06)	.424
Condom use with all partners in lifetime[a]	0.98 (0.79–1.22)	.878
Condom use with all paying partners in the past 3 months[a]	1.03 (0.95–1.12)	.469
Ever performed intravaginal washing		
No	1	
Yes	0.59 (0.41–0.85)	.005
Often performed intravaginal washing shortly before vaginal sex in lifetime[a]	0.92 (0.85–1.01)	.086
Often performed intravaginal washing shortly after vaginal sex in lifetime[a]	0.88 (0.81–0.97)	.007
Solutions used in intravaginal washing[b]		
Water only	1	
Water with soap only	1.03 (0.60–1.79)	.904
Water with lemon or lemon only	1.00 (0.30–3.34)	.999
Water with salt only	1.39 (0.74–2.58)	.306
Commercial solutions only	1.14 (0.51–2.57)	.746
Ever used ≥2 different solutions	1.00 (0.63–1.58)	.999
Average number of times performed intravaginal washing per week in the past 3 months		(.003)
0	1	
1–2 times	0.78 (0.52–1.16)	.212
3–4 times	0.53 (0.33–0.86)	.010
5–7 times	0.49 (0.28–0.87)	.014

J Med Virol. Author manuscript; available in PMC 2019 November 01.

Characteristics	Number of infecting HPV genotypes, excluding HPV 6&11	
	IRR (95% CI)	p-values (p-trend)
8–10 times	0.39 (0.23–0.67)	<.001
>10 times	0.86 (0.55–1.33)	.489
Ever performed intravaginal wiping		
No	1	
Yes	0.82 (0.60–1.12)	.208
Often performed intravaginal wiping shortly before vaginal sex in lifetime[a]	0.99 (0.83–1.19)	.963
Often performed intravaginal wiping shortly after vaginal sex in lifetime[a]	0.94 (0.79–1.12)	.474
Average number of times performed intravaginal wiping per week in the past 3 months		(.352)
0	1	
1–2 times	0.85 (0.60–1.22)	.376
3–4 times	0.51 (0.23–1.15)	.106
5 times or more	1.04 (0.57–1.92)	.900
Ever performed intravaginal insertion		
No	1	
Yes	0.81 (0.43–1.52)	.508

Note: IRR: incident rate ratios

[a] Values ranged from *1 = Never* to *5 = Every time having vaginal sex*

[b] Among those who performed intravaginal washing, n = 180

J Med Virol. Author manuscript; available in PMC 2019 November 01.

Table 3.

Multivariable Associations Between Intravaginal Washing and Number of Infecting HPV Genotypes (N=191)

Variables	Model 1 [a]		Model 2 [a]	
	IRR (95% CI)	p-values (p-trend)	IRR (95% CI)	p-values (p-trend)
Age	0.96 (0.93–0.99)	.023	0.96 (0.93–0.99)	.026
Main venue to contact clients				
Establishment-based (e.g., brothels, massage parlors, karaoke parlors)	1		1	
Freelancers (e.g., at bars, beer gardens)	0.71 (0.48–1.05)	.089	0.67 (0.45–1.00)	.051
Street-based	1.31 (0.94–1.83)	.115	1.31 (0.94–1.82)	.117
Self-reported HIV status				
Negative	1		1	
Positive	1.65 (1.11–2.47)	.014	1.50 (0.98–2.28)	.060
No. of different paying partners in the past 3 months		(.144)		(.207)
≤30	1		1	
31–60	1.68 (1.20–2.37)	.002	1.65 (1.18–2.31)	.004
61–90	1.03 (0.62–1.72)	.909	1.10 (0.65–1.84)	.743
91–180	0.66 (0.39–1.11)	.117	0.65 (0.38–1.10)	.105
>180	1.07 (0.63–1.80)	.813	1.13 (0.67–1.91)	.638
Condom use with all paying partners in the past 3 months [b]	1.06 (0.98–1.15)	.117	1.06 (0.97–1.15)	.196
Completed all required doses of HPV vaccines				
No	1		1	
Yes	0.63 (0.37–1.08)	.093	0.63 (0.37–1.08)	.093
Performed intravaginal washing in the past 3 months				
No	1			
Yes	0.65 (0.46–0.94)	.020		
Often performed intravaginal washing shortly after vaginal sex [b]			0.89 (0.81–0.99)	.034

Note: IRR: incident rate ratios

[a] Dependent variable = Number of infecting HPV genotypes, excluding HPV 6&11.

[b] Values ranged from *1 = Never* to *5 = Every time having vaginal sex*