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Concordance of Beta-papillomavirus across anogenital and oral anatomic sites of men: The HIM Study

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Summary

We evaluated the concordance between β-HPVs detected in external genital skin, anal canal, and oral cavity specimens collected simultaneously from 717 men that were participating in the multinational HIM Study. Viral genotyping was performed using the Luminex technology. Species- and type-specific concordance was measured using kappa statistics for agreement. Overall, concordance of β-HPVs across sites was low and mainly observed among paired genital/

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Ethical approval

Ethical approval was given by the University of South Florida (IRB# 102660), Ludwig Institute for Cancer Research, Centro de Referência e Treinamento em Doenças Sexualmente Transmissíveis e AIDS, and Instituto Nacional de Salud Pública de México.

Conflict of Interest

No conflicts of interest are declared for any of the remaining authors.

anal canal samples. When grouped by species, solely β−4 HPVs showed moderate concordance in genital/anal pairs (κ=0.457), which could be attributed to the substantial concordance of HPV-92 in men from Brazil and Mexico (κ>0.610). β-HPV type concordance was higher in Mexico, where HPV-19 was consistently concordant in all anatomic site combinations. Our analysis indicates that type-specific concordance across sites is limited to few viral types; however, these infections seem to occur more often than would be expected by chance, suggesting that although rare, there is agreement among sites.

Keywords

cutaneous human papillomavirus; males; HIM Study; concordance; anogenital; oral

Introduction

Most of over 200 human papillomavirus (HPV) types characterized to date cluster within the Alpha (α)-, Beta (β)-, or Gamma (γ)-HPV genus. Whereas α-HPVs are mainly mucosal types isolated from the anogenital epithelia, β- and γ-HPVs, originally classified as cutaneous types, are ubiquitously distributed throughout the body and may be an intrinsic part of the commensal flora (Antonsson et al., 2000, 2003; Bottalico et al., 2011; Hazard et al., 2007; Sichero et al., 2014, 2015). Additional research is crucial to better understand the pathological implications of the broad distribution of these viruses.

Several studies show that oral α-HPV infections are associated with immunosuppression and with sexual behavior resembling associations observed at the anogenital epithelia (Kreimer et al., 2004). In contrast, at the oral cavity and anogenital epithelia, β-HPVs seem to differ from that of α-HPVs, in the lack of association between β-HPV detection and sexual risk factors. This observation points toward other routes of transmission such as autoinoculation and non-penetrative sexual activities (Sichero et al., 2014, 2015; Nunes et al., 2016; Donà et al., 2015; Torres et al., 2015). Alternatively, detection of β-HPVs in one anatomic site may also represent deposition of virions shed from other anatomic sites (Sichero et al., 2014, 2015; Liu et al., 2015).

The few reports in which the oral and anogenital regions were analyzed concurrently indicate that simultaneous oral-cervical type-specific α-HPV infections are relatively rare (Fakhry et al., 2006; Smith et al., 2004; Steinau et al., 2017; Termine et al., 2011). Instead, specific β-HPV types are not only shared within family members, but are also commonly detected in different cutaneous anatomical sites within the same individual (Antonsson et al., 2000, 2003; Weissenborn et al., 2009). Nevertheless, the relation between oral and anogenital β-HPV infections remains poorly explored (Hampras et al., 2017; Moscicki et al., 2017). Therefore, the purpose of this study was to evaluate the concordance of 43 β-HPV types detected across samples simultaneously collected from the external genital skin, the anal canal, and the oral cavity among men enrolled in the multinational HPV Infection in Men (HIM) cohort study from three different countries.

Materials and Methods

Clinical samples and β-HPV genotyping

Biological specimens originated from the HIM Study cohort (HPV Infection in Men) conducted among over 4,000 men from São Paulo (Brazil), Cuernavaca (Mexico) and Tampa, Florida (US) (Nunes et al., 2016). Study methods and design are described in detail elsewhere (Giuliano et al., 2008, 2011; Kreimer et al., 2011; Nyitray et al., 2011). The study was approved by human research ethics committees at each study sites. Informed consent was obtained from all participants prior to enrollment.

The present analysis included 717 men (238 from the US, 241 from Brazil and 238 from Mexico) with adequate (β-globin positive and/or β-HPV positive) specimens collected from the oral cavity, anal canal and genital skin, collected at the same study visit. For HPV status assessment, PCRs were conducted using a mixture of specific biotinylated primers capable of amplifying a fragment of the *E7* gene of 43 β-HPV from five different species, followed by bead-based Luminex technology (Gheit et al., 2007; Nunes et al., 2016; Schmitt et al., 2006). Primers for the amplification of β-globin were also added to evaluate the quality of template DNA. For each probe, MFI (mean fluorescence intensity) values obtained when no PCR product was added to the hybridization mixture was considered the background values. Cutoffs were computed by adding 20 MFI to 1.1X the median background value.

Statistical analysis

Pearson chi-square test was used to compare the prevalence of any β-HPV at the anal canal or oral cavity among men positive or negative for any β-HPV at the genitals. Samples collected from the three anatomic sites were considered concordant if the same HPV species or genotype was detected at two or more anatomic sites simultaneously. HPV concordance between any two paired samples of the three different specimens (genital, anal canal, oral cavity) was calculated using kappa statistics (Cohen, 1960) which express the frequency of agreement beyond chance. Kappa-values less than 0.4 indicate poor agreement, a kappa estimate between 0.41 and 0.60 indicates moderate agreement, and a kappa estimate between 0.61 and 0.8 indicates substantial agreement (Landis and Koch, 1977). Precision, a measure of exactness, consists of the percentage of positive agreement between any two paired samples (either negative or positive for the same type specific β-HPV) among all cases analyzed.All analyses were conducted using SPSS 18.0 software (SPSS, Chicago, IL) to calculate kappa values and MedCalc (r) Version 16.8 (c) 1993–2016 MedCalc Software to calculate 95% confident interval [95%CI].

Results

We detected 3,302 single β-HPV infections across all anatomic sites among the 717 men included in this analysis. At least one β-HPV was detected in 557 (77.7%), 389 (54.3%) and 210 (29.3%) specimens of the external genital skin, anal canal, and oral cavity, respectively. Detection of multiple β-HPVs was more common at the genitals than in the anal canal or oral cavity. A detailed distribution of β-HPVs in this population and population characteristics have been previously described (Nunes et al., 2016).

Having β-HPV DNA (any type) at all three anatomical sites was observed in 127 men (17.7%). Detection of any β-HPV at the anal canal was more common among men with any β-HPV at the genitals (57.1%) than among genital β-HPV DNA negative men (44%) (p=0.004). Likewise, the prevalence of any β-HPV at the oral cavity was higher among men with any β-HPV at the genitals compared to men without genital β-HPV (32.5% versus 18%, p<0.001) (Table 1).

Independent of the sample geographical origin (US, Brazil, Mexico), when analyzed any β-HPV or grouped by species, kappa values revealed only slight agreement for all three anatomic pair combinations analyzed (Table 2). The only exception was β−4 HPV species for which agreement in genital/anal canal samples was moderate (κ=0.457). We next evaluated agreement strength for individual β-HPV types. Table 3 shows kappa values exclusively of viral types for which at least in one pair sample combination the agreement observed was moderate (κ=0.410–0.600) or substantial (κ=0.610–0.800). The highest kappa values were observed among Mexican men and among paired genital/anal canal samples. The majority of type-specific kappa values >0.410 were restricted to men from one single geographic region. Exceptions were HPV-92 (β−4 species) that showed substantial concordance in paired genital/anal canal specimens from Brazil and Mexico (κ>0.610), and HPV-93 (β−1 species) which presented moderate agreement in genital/anal canal and genital/oral pairs from Mexico (κ=0.434, both), and substantial agreement in oral/anal canal regions in the US. Furthermore, in Mexico, HPV-19 (β−1) was consistently concordant in all anatomical sites combinations analyzed. Nevertheless, it is important to highlight that when considering individual β-HPV types, analyses were based in very few cases (Table 4) which precluded the evaluation of socio-demographic and behavior variables that could be associated to the risk of detecting the same viral type in more than one anatomic site.

Discussion

We previously reported the presence of any β-HPV in 67.3% of normal skin swabs and 56.5% of eyebrow hairs within a subcohort of 209 US men enrolled to the HIM study (Hampras et al., 2014). Additionally, among 123 participants from the US, we observed that concordance of any β-HPV detection was greater (31.0%) across keratinized tissue sites (genital skin, eyebrow hairs, and forearm skin) than across mucosal sites (anal and oral mucosa, 6.9%) (Hampras et al., 2017). We now widen our analysis to access the agreement of β-HPV DNA detection across samples collected concurrently from the external genital skin, anal canal, and oral cavity in a HIM sub-cohort of 717 healthy men from three different countries (Brazil, US, Mexico). Despite the broad diversity of β-HPV types detected, and the fact that any β-HPV infection was higher at the anal canal and oral cavity of men in which any β-HPV was also detected in the genitals, species- and type-specific β-HPV agreement across two anatomic sites was a rare event.

Considering solely cutaneous anatomic regions (arms, forehead and thighs), high type-specific β-HPV agreement across sites has been described among healthy individuals (Antonsson et al., 2000, 2003; Weissenborn et al., 2009), and also among individuals affected with psoriasis (Cronin et al., 2008), epidermodisplasia verruciformis (Dell'Oste et al., 2009), actinic keratosis (Schneider et al., 2013), or squamous cell carcinoma (Plasmeijer

et al., 2010). In contrast, in our study the strength of agreement across genital, anal canal, and oral cavity was poor for most individual viral types analyzed. Nevertheless, one should consider that given the large number of samples and β-HPV types analyzed, it is unlikely that the same HPV type will be found at a distant skin site by chance. Furthermore, since the prevalence of HPV in the oral cavity was less than a half of that observed at the genitals, the low positivity agreement measured was not surprising.

The high type-specific HPV discordance observed by us could derive either from differences in HPV natural history at each of three anatomic sites or from separate exposures events. The natural history of α-HPV infections at anogenital epithelia and the oral cavity are associated with sexual behavior as these infections are mostly sexually transmitted. Despite a common mode of transmission, oral-cervical type-specific α-HPV agreement is low among women (Fakhry et al., 2006; Steinau et al., 2017; Termine et al., 2009, 2011), and in men α-HPV agreement, although rare, is higher between the penile and anal sites (7%) compared to anal-oral concordance (2%) (King et al., 2015; Tsikis et al., 2017; van Rijn et al., 2014). Interestingly, among men with genital warts, agreement of α-HPV types between the anal canal and genital wart was 78.1%, while concordance between oral and genital wart types was 60.9% (Kofoed et al., 2014). Taken together, these data indicate that the low type-specific α-HPV agreement reported possibly derives independent infection events. For β-HPVs, autoinoculation and non-penetrative sexual activities may be plausible forms of viral transmission (Donà et al., 2015; Nunes et al., 2016; Sichero et al., 2014, 2015; Torres et al., 2015). However, among healthy women from Brazil, we observed no association of β-HPV infections at the cervix and certain hygienic/sexual behaviors including hygienic tampon or menstrual cloth use, masturbation frequency, or vagina douching (Sichero et al., 2017). Additionally, it was recently reported that among heterosexual couples, the transmission rate of β-HPVs between anogenital sites was 15.9 per 100 person months from men-to-women and that risk for women-to-men transmission was similar, suggesting that these can be sexually transmitted (Moscicki et al., 2017). Unfortunately, the low type-specific agreement between any two anatomic sites precluded an analysis to assess factors associated with concordance at any two anatomical regions. Finally, one may not exclude the possibility of differences in the susceptibility of the oral and anogenital mucosa to genotype specific β-HPV infections.

We observed that β-HPV type-specific agreement was more common in the Mexican population as compared to Brazil and the US populations. In fact, among men from the HIM study, we also recently reported significant geographic differences in the natural history of α-HPV types 6 and 16 genital infection (Sudenga et al., 2017). Interestingly, we observed that β-HPV types for which strength of agreement for any two anatomical sites was moderate or substantial were not the most prevalent in the populations analyzed (Nunes et al., 2016). For instance, in Mexico, the most prevalent β-HPV types were HPV-21 (β−1), −22 (β−2), 24 (β−1), and −38 (β−2) independent of the anatomic region, whereas HPV-19 (β−3) showed substantial agreement between genital-oral and anal-oral specimens of the same individual. Thus, although small in number, concurrent infections of the same viral types occurred more often than would be expected by chance. Further studies are warranted to elucidate the biological basis of agreements observed.

A limitation of our study is that we examined only 43 of the 51 β-HPV types characterized to date, and the current analysis was not extended to γ-HPV genomes. Assuming new HPV cutaneous types are continually identified, our study likely underestimated the prevalence of additional viral genotypes. Nevertheless, because the methodology used to access β-HPV DNA is a high-sensitive DNA-based assay one may be over detecting viral DNA that not necessarily represent true infection.

In conclusion, our data point towards a high prevalence of different β-HPV types at the external genital skin, as compared to the anal canal and oral cavity of men from different countries. Although type-specific concordance across sites was limited to few viral types, our analysis indicates that such infections are unlikely to be independent of one another. Further studies may allow a more detailed investigation of the biological relationship between any two anatomic sites, but also of risk factors of viral acquisition and modes of transmission in different populations.

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Abbreviations:

(HPV)	Human papillomavirus
(HIM Study)	HPV Infection in Men Study
(SS)	skin swabs
(EB)	eyebrow hairs
(NMSC)	non-melanoma skin cancer

References

Antonsson A, Forslund O, Ekberg H, Sterner G, Hansson BG. The ubiquity and impressive genomic diversity of human skin papillomaviruses suggest a commensalic nature of these viruses. J Virol 74(24):11636–41,2000. [PubMed: 11090162]

Antonsson A, Karanfilovska S, Lindqvist PG, Hansson BG. General acquisition of human papillomavirus infections of skin occurs in early infancy. J Clin Microbiol 41(6):2509–14,2003. [PubMed: 12791874]

Bottalico D, Chen Z, Dunne A, Ostoloza J, McKinney S, Sun C, Schlecht NF, Fatahzadeh M, Herrero R, Schiffman M, Burk RD. The oral cavity contains abundant known and novel human papillomaviruses from the Betapapillomavirus and Gammapapillomavirus genera. J Infect Dis 204(5):787–92,2011. [PubMed: 21844305]

Cohen J A Coefficient of Agreement for Nominal Scales. Educ Psychol Meas 20(1):37–46,1960.

Cronin JG, Mesher D, Purdie K, Evans H, Breuer J, Harwood CA, McGregor JM, Proby CM. Beta-papillomaviruses and psoriasis: an intra-patient comparison of human papillomavirus carriage in skin and hair. Br J Dermatol 159(1):113–9,2008. [PubMed: 18510676]

Dell'Oste V, Azzimonti B, De Andrea M, Mondini M, Zavattaro E, Leigheb G, Weissenborn SJ, Pfister H, Michael KM, Waterboer T, Pawlita M, Amantea A, Landolfo S, Gariglio M. High beta-HPV DNA loads and strong seroreactivity are present in epidermodysplasia verruciformis. J Invest Dermatol 129(4):1026–34,2009. [PubMed: 18923444]

Donà MG, Gheit T, Latini A, Benevolo M, Torres M, Smelov V, McKay-Chopin S, Giglio A, Cristaudo A, Zaccarelli M, Tommasino M, Giuliani M. Alpha, beta and gamma Human Papillomaviruses in the anal canal of HIV-infected and uninfected men who have sex with men. J Infect 71(1):74–84,2015. [PubMed: 25698067]

Fakhry C, D'souza G, Sugar E, Weber K, Goshu E, Minkoff H, Wright R, Seaberg E, Gillison M. Relationship between prevalent oral and cervical human papillomavirus infections in human immunodeficiency virus-positive and -negative women. J Clin Microbiol 44(12):4479–85,2006. [PubMed: 17021055]

Gheit T, Billoud G, de Koning MN, Gemignani F, Forslund O, Sylla BS, Vaccarella S, Franceschi S, Landi S, Quint WG, Canzian F, Tommasino M. Development of a sensitive and specific multiplex PCR method combined with DNA microarray primer extension to detect Betapapillomavirus types. J Clin Microbiol 45(8):2537–44,2007. [PubMed: 17581938]

Giuliano AR, Lazcano-Ponce E, Villa LL, Flores R, Salmeron J, Lee JH, Papenfuss MR, Abrahamsen M, Jolles E, Nielson CM, Baggio ML, Silva R, Quiterio M. The human papillomavirus infection in men study: human papillomavirus prevalence and type distribution among men residing in Brazil, Mexico, and the United States. Cancer Epidemiol Biomarkers Prev 17(8):2036–43,2008. [PubMed: 18708396]

Giuliano AR, Lee JH, Fulp W, Villa LL, Lazcano E, Papenfuss MR, Abrahamsen M, Salmeron J, Anic GM, Rollison DE, Smith D. Incidence and clearance of genital human papillomavirus infection in men (HIM): a cohort study. Lancet 377 (9769):932–40,2011. [PubMed: 21367446]

Hampras SS, Giuliano AR, Lin HY, Fisher KJ, Abrahamsen ME, Sirak BA, Iannacone MR, Gheit T, Tommasino M, Rollison DE. Natural History of Cutaneous Human Papillomavirus (HPV) Infection in Men: The HIM Study. PLoS One 9(9):e104843,2014.

Hampras SS, Rollison DE, Giuliano AR, McKay-Chopin S, Minoni L, Sereday K, Gheit T, Tommasino M. Prevalence and concordance of cutaneous beta human papillomavirus infection at mucosal and cutaneous sites. J Infect Dis 2017. [Epub ahead of print].

Hazard K, Karlsson A, Andersson K, Ekberg H, Dillner J, Forslund O. Cutaneous human papillomaviruses persist on healthy skin. J Invest Dermatol 127(1):116–9,2007. [PubMed: 17024097]

King EM, Gilson R, Beddows S, Soldan K, Panwar K, Young C, Jit M, Edmunds WJ, Sonnenberg P. Oral human papillomavirus (HPV) infection in men who have sex with men: prevalence and lack of anogenital concordance. Sex Transm Infect 91(4):284–6,2015. [PubMed: 25887283]

Kreimer AR, Alberg AJ, Daniel R, Gravitt PE, Viscidi R, Garrett ES, Shah KV, Gillison ML. Oral human papillomavirus infection in adults is associated with sexual behavior and HIV serostatus. J Infect Dis 189(4):686–98,2004. [PubMed: 14767823]

Kreimer AR, Villa A, Nyitray AG, Abrahamsen M, Papenfuss M, Smith D, Hildesheim A, Villa LL, Lazcano-Ponce E, Giuliano AR. The epidemiology of oral HPV infection among a multinational sample of healthy men. Cancer Epidemiol Biomarkers Prev 20(1):172–82,2011. [PubMed: 21148755]

Kofoed K, Sand C, Forslund O, Madsen K. Prevalence of human papillomavirus in anal and oral sites among patients with genital warts. Acta Derm Venereol 94(2):207–11,2014. [PubMed: 24096893]

Landis JR, Koch GG. The measurement of observer agreement for categorical data. Biometrics 33(1): 159–74,1997.

Liu Z, Rashid T, Nyitray AG. Penises not required: a systematic review of the potential for human papillomavirus horizontal transmission that is non-sexual or does not include penile penetration. Sex Health 13(1):10–21,2015.

Moscicki AB, Ma Y, Gheit T, McKay-Chopin S, Farhat S, Widdice LE, Tommasino M. Prevalence and Transmission of Beta and Gamma Human Papillomavirus in Heterosexual Couples. Open Forum Infect Dis 4(1):ofw216, 2017.

Nyitray AG, da Silva Carvalho RJ, Baggio ML, Lu B, Smith D, Abrahamsen M, Papenfuss M, Villa LL, Lazcano-Ponce E, Giuliano AR. Age-specific prevalence of and risk factors for anal human papillomavirus (HPV) among men who have sex with women and men who have sex with men: the HPV in men (HIM) study. J Infect Dis 203(1):49–57,2011. [PubMed: 21148496]

Nunes EM, Sudenga SL, Gheit T, Tommasino M, Baggio ML, Ferreira S, Galan L, Silva RC, Pierce Campbell CM, Lazcano-Ponce E, Giuliano AR, Villa LL, Sichero L; HIM Study group. Diversity of beta-papillomavirus at anogenital and oral anatomic sites of men: The HIM Study. Virology 495:33–41,2016. [PubMed: 27161202]

Plasmeijer EI, Neale RE, Buettner PG, de Koning MN, Ter Schegget J, Quint WG, Green AC, Feltkamp MC. Betapapillomavirus infection profiles in tissue sets from cutaneous squamous cell-carcinoma patients. Int J Cancer 126(11):2614–21,2010. [PubMed: 19856311]

Schmitt M, Bravo IG, Snijders PJ, Gissmann L, Pawlita M, Waterboer T. Bead-based multiplex genotyping of human papillomaviruses. J Clin Microbiol 44(2):504–12,2006. [PubMed: 16455905]

Schneider I, Lehmann MD, Kogosov V, Stockfleth E, Nindl I. Eyebrow hairs from actinic keratosis patients harbor the highest number of cutaneous human papillomaviruses. BMC Infect Dis 13:186,2013. [PubMed: 23618013]

Sichero L, Pierce Campbell CM, Fulp W, Ferreira S, Sobrinho JS, Baggio ML, Galan L, Silva RC, Lazcano-Ponce E, Giuliano AR, Villa LL. High genital prevalence of cutaneous human papillomavirus DNA on male genital skin: the HPV Infection in Men Study. BMC Infect Dis 14:667,2014. [PubMed: 25515915]

Sichero L, Nyitray AG, Nunes EM, Nepal B, Ferreira S, Sobrinho JS, Baggio ML, Galan L, Silva RC, Lazcano-Ponce E, Giuliano AR, Villa LL. Diversity of human papillomavirus in the anal canal of men: The HIM study. Clin Microbiol Infect 21(5):502–9,2015. [PubMed: 25698660]

Sichero L, El-Zein M, Nunes EM, Ferreira S, Franco EL, Villa LL. Cervical Infection with Cutaneous Beta and Mucosal Alpha Papillomaviruses. Cancer Epidemiol Biomarkers Prev 2017. [Epub ahead of print].

Smith EM, Ritchie JM, Yankowitz J, Wang D, Turek LP, Haugen TH. HPV prevalence and concordance in the cervix and oral cavity of pregnant women. Infect Dis Obstet Gynecol 12(2):45–56,2004. [PubMed: 15739817]

Steinau M, Gorbach P, Gratzer B, Braxton J, Kerndt PR, Crosby RA, Unger ER, Markowitz LE, Meites E. Concordance between Anal and Oral Human Papillomavirus (HPV) Infections Among Young Men Who Have Sex with Men. J Infect Dis 2017 [Epub ahead of print].

Sudenga SL, Torres BN, Silva R, Villa LL, Lazcano-Ponce E, Abrahamsen M, Baggio ML, Salmeron J, Quiterio M, Giuliano AR. Comparison of the natural history of genital HPV infection among men by country: Brazil, Mexico, and the United States. Cancer Epidemiol Biomarkers Prev 2017 [Epub ahead of print].

Termine N, Giovannelli L, Matranga D, Perino A, Panzarella V, Ammatuna P, D'Angelo M, Campisi G. Low rate of oral human papillomavirus (HPV) infection in women screened for cervical HPV infection in Southern Italy: A cross-sectional study of 140 immunocompetent subjects. J Med Virol 81(8):1438–43,2009. [PubMed: 19551817]

Termine N, Giovannelli L, Matranga D, Caleca MP, Bellavia C, Perino A, Campisi G. Oral human papillomavirus infection in women with cervical HPV infection: new data from an Italian cohort and a metanalysis of the literature. Oral Oncol 47(4):244–50,2011. [PubMed: 21429788]

Torres M, Gheit T, McKay-Chopin S, Rodríguez C, Romero JD, Filotico R, Doná MG, Ortiz M, Tommasino M. Prevalence of beta and gamma human papillomaviruses in the anal canal of men who have sex with men is influenced by HIV status. J Clin Virol 67:47–51,2015. [PubMed: 25959158]

Tsikis S, Hoefer L, Bethimoutis G, Nicolaidou E, Paparizos V, Antoniou C, Chardalias L, Stavropoulos GE, Sharma S, Long BC, Mujacic I, Kadri S, Segal JP, Furtado LV, Schneider J, Charnot-Katsikas A. Risk factors, prevalence, and site concordance of human papillomavirus in high-risk Greek men. Eur J Cancer Prev 2017. [Epub ahead of print].

van Rijn VM, Mooij SH, Mollers M1, Snijders PJ, Speksnijder AG, King AJ, de Vries HJ, van Eeden A, van der Klis FR, de Melker HE, van der Sande MA, van der Loeff MF. Anal, penile, and oral high-risk HPV infections and HPV seropositivity in HIV-positive and HIV-negative men who have sex with men. PLoS One 20;9(3):e92208,2014.

Weissenborn SJ, De Koning MN, Wieland U, Quint WG, Pfister HJ. Intrafamilial transmission and family-specific spectra of cutaneous betapapillomaviruses. J Virol 83(2):811–6,2009. [PubMed: 18987132]

Table 1.

Any β-HPV DNA detection at the genital region, anal canal, and oral cavity in paired samples from the same HIM participant.

	any β-HPV - anal canal[1]		any β-HPV - oral cavity[2]	
	positive	**negative**	**positive**	**negative**
any β-HPV positive - genital (n=557)	318	239	181	376
any β-HPV negative - genital (n=160)	71	89	29	131
total	389	328	210	507

Pearson chi-square test was used to compare the prevalence of any β-HPV at the anal canal or oral cavity among men positive or negative for any β-HPV at the genitals.

[1] p=0.004

[2] p<0.001.

Table 2.

Kappa values and precision (by n of pairs) of Beta-HPV species agreement across anatomical regions among 717 men participating in the HPV Infection in Men Study.

Beta-HPV	Genital/Anal canal (95%CI)[1]	Precision (%)[2]	Genital/Oral (95%CI)	Precision (%)	Oral/Anal canal (95%CI)	Precision (%)
Any β-HPV	0.093 (0.028; 0.157)	407 (56.76)	0.086 (0.044; 0.127)	314 (43.79)	0.173 (0.110; 0.235)	410 (57.18)
Beta-1	0.112 (0.047; 0.177)	388 (54.11)	0.135 (0.084; 0.185)	388 (54.11)	0.200 (0.126; 0.273)	497 (69.32)
Beta-2	0.161 (0.100; 0.221)	394 (54.95)	0.086 (0.047; 0.125)	323 (45.05)	0.141 (0.076; 0.207)	462 (64.44)
Beta-3	0.285 (0.204; 0.365)	562 (78.38)	0.119 (0.056; 0.182)	551 (76.85)	0.110 (0.017; 0.203)	624 (87.03)
Beta-4	**0.457 (0.119; 0.795)**	710 (99.02)	NE	709 (98.88)	NE	712 (99.30)
Beta-5	−0.008 (−0.012; −0.003)	706 (98.47)	NE	711 (99.16)	NE	712 (99.30)

[1] kappa coefficient of concordance; 95%CI- 95% confidence intervals; NE- could not be evaluated. Strength of agreement is poor when of value of kappa is <0.000; slight when kappa among 0.000–0.200; fair when 0.210–0.400; moderate when 0.410–0.600; substantial when 0.610–0.800. Moderate or substantial agreements are in bold.

[2] precision is percentage of agreement (number DNA negative or positive for the same type specific β-HPV in any two paired samples)/(total number of any two paired samples).

Table 3.

Value of kappa and precision of Beta-HPV types in agreement of anatomical sites from 717 men participating in the HPV Infection in Men Study.

	US		Brazil		Mexico	
	Kappa[1] (95%CI)	Precision[2] (%)	Kappa (95%CI)	Precision (%)	Kappa (95%CI)	Precision (%)
genital/anal canal						
Beta-1						
HPV-19	−0.011 (−0.023; −0.0002)	97.48	−0.007 (−0.021; 0.006)	96.27	**0.565 (0.123; 1)**	98.74
HPV-93	0.277 (−0.162; 0.716)	97.90	NE	99.59	**0.434 (0.024; 0.844)**	97.90
HPV-105	**0.415 (0.088; 0.741)**	96.64	0.146 (−0.061; 0.353)	92.12	0.396 (0.160; 0.632)	93.28
Beta-2						
HPV-22	0.163 (0.009; 0.318)	81.09	0.169 (0.010; 0.328)	85.48	**0.469 (0.327; 0.610)**	84.45
HPV-104	NE	97.90	−0.007 (−0.017; 0.004)	97.93	**0.663 (0.225; 1)**	99.16
Beta-3						
HPV-49	**0.454 (0.159; 0.750)**	96.22	0.229 (0.006; 0.451)	92.53	0.102 (0.087; 0.478)	88.66
HPV-76	0.244 (0.054; 0.434)	88.24	**0.525 (0.365; 0.685)**	90.04	0.236 (0.049; 0.422)	87.82
Beta-4						
HPV-92	−0.008 (−0.017; −0.0002)	98.32	**0.663 (0.225; 1)**	99.17	**0.665 (0.047; 1)**	99.58
genital/oral cavity						
Beta-1						
HPV-19	−0.007 (−0.018; 0.004)	97.90	0.216 (−0.139; 0.572)	97.10	**0.798 (0.411; 1)**	99.58
HPV-36	NE	100	−0.008 (−0.017; −0.0002)	98.34	**0.497 (−0.103; 1)**	99.16
HPV-93	0.277 (−0.162; 0.716)	97.90	NE	99.17	**0.434 (0.024; 0.844)**	97.90
Beta-2						
HPV-15	0.176 (−0.123; 0.475)	96.22	0.231 (−0.049; 0.511)	95.02	**0.566 (0.127; 1)**	98.74
oral cavity/anal canal						
Beta-1						
HPV-14	**0.665 (0.047; 1)**	99.58	0.189 (−0.141; 0.519)	96.68	NE	100
HPV-19	−0.006 (−0.014; 0.002)	98.74	−0.004 (−0.010; 0.002)	99.17	**0.663 (0.225; 1)**	99.16
HPV-47	**0.565 (0.123; 1)**	98.74	−0.006 (−0.016; 0.003)	98.34	0.111 (−0.124; 0.347)	94.54
HPV-93	−0.008 (−0.017; −0.0002)	98.32	**0.665 (0.047; 1)**	99.59	0.237 (−0.164; 0.638)	97.48
Beta-2						
HPV-75	**0.497 (−0.103; 1)**	99.16	−0.006 (−0.013; 0.002)	98.75	NE	98.74

[1] Kappa-kappa coefficient of concordance; 95%CI- 95% confidence intervals; NE-could not be evaluable. Strength of agreement is poor when of value of kappa is <0.000; slight when kappa among 0.000–0.200; fair when 0.210–0.400; moderate when 0.410–0.600; substantial when 0.610–0.800. Moderate or substantial agreements are in bold.

[2] precision is percentage of agreement (number DNA negative or positive for the same type specific β-HPV in any two paired samples)/(total number of any two paired samples).

Table 4.

Detection of specific Beta-HPV types in different anatomical region among 717 men for which agreement of concordance was moderate or substantial[1].

	US			Brazil			Mexico			All		
	Genital	**Anal**	**Oral**	**Genital**	**Anal**	**Oral**	**Genital**	**Anal**	**Oral**	**Genital**	**Anal**	**Oral**
Beta-1												
HPV-14	6	2	1	23	8	2	7	0	0	36	10	3
HPV-19	4	2	1	2	1	1	3	4	2	9	7	4
HPV-36	0	3	0	2	0	2	1	0	3	3	3	5
HPV-47	6	3	4	19	1	3	24	11	4	49	15	11
HPV-93	5	2	2	0	1	2	5	4	4	10	7	8
HPV-105	10	4	0	18	5	1	18	10	1	46	19	2
Beta-2												
HPV-15	10	4	1	12	2	4	5	1	2	27	7	7
HPV-22	39	22	4	35	10	2	53	30	15	127	62	21
HPV-75	3	3	1	13	2	1	8	3	0	24	8	2
HPV-104	5	0	0	4	1	0	4	2	0	13	3	0
Beta-3												
HPV-49	12	5	0	20	4	3	25	16	5	57	25	8
HPV-76	27	13	6	39	17	4	28	13	9	94	43	19
Beta-4												
HPV-92	2	2	0	4	2	0	2	1	0	8	5	0

[1] number of men in which the specific β-HPV type was detected at the anatomical site indicated. Each infection is counted as an individual event in multiple infections.