Low Prevalence of Oral and Nasal Human Papillomavirus in Employees Performing CO2-laser Evaporation of Genital Warts or Loop Electrode Excision Procedure of Cervical Dysplasia

Kofoed, Kristian; Norrbom, Christina; Forslund, Ola; Moller, Charlotte; Froding, Ligita P.; Pedersen, Anders Elm; Markauskas, Algirdas; Blomberg, Maria; Baunigartner-Nielsen, Jane; Madsen, Jakob Torp; Strauss, Gitte; Madsen, Klaus G.; Sand, Carsten

Published in:
Acta Dermato-Venereologica

DOI:
10.2340/00015555-1912

2015

Acta Derm Venereol 2015; 95: 173–176

INVESTIGATIVE REPORT

Low Prevalence of Oral and Nasal Human Papillomavirus in Employees Performing CO_2-laser Evaporation of Genital Warts or Loop Electrode Excision Procedure of Cervical Dysplasia*

Kristian KOFOED[1], Christina NORRBOM[2], Ola FORSLUND[3], Charlotte MØLLER[4], Ligita P. FRØDING[5], Anders Elm PEDERSEN[6], Algirdas MARKAUSKAS[7], Maria BLOMBERG[8], Jane BAUMGARTNER-NIELSEN[9], Jakob Torp MADSEN[10], Gitte STRAUSS[11], Klaus G. MADSEN[12] and Carsten SAND[1]

[1]Department of Dermato-venereology, Bispebjerg Hospital, Copenhagen University Hospital, Copenhagen, [2]Department of Obstetrics and Gynecology, Hillerød Hospital, Copenhagen University Hospital, Hillerød, Denmark, [3]Department of Laboratory Medicine, Division of Medical Microbiology, Lund University, Skåne University Hospital, Malmö, Sweden, [4]Department of Gynecology and Obstetrics, Aarhus University Hospital, Aarhus, [5]Department of Gynecology, Herlev Hospital, Copenhagen University Hospital, Herlev, [6]Department of International Health, Immunology and Microbiology, University of Copenhagen, Copenhagen, Departments of [7]Gynecology and Obstetrics and [10]Dermato-venereology, Odense University Hospital, University of Southern Denmark, Odense, [8]Virus, Lifestyle and Genes, Danish Cancer Society Research Center, Copenhagen, [9]Department of Dermato-venereology, Aarhus University Hospital, Aarhus, [11]Department of Dermato-allergology, Gentofte Hospital, Copenhagen University Hospital, Hellerup and [12]Sanofi Pasteur MSD, Lyngby, Denmark

Risk of human papillomavirus (HPV) transmission during laser vaporisation of genital warts or loop electrode excision procedure is controversial. An oral rinse, a nasal swabs, history of HPV-related diseases and data on HPV exposure were collected from 287 employees at departments of dermato-venerology and gynaecology in Denmark. A mucosal HPV type was found among 5.8% of employees with experience of laser treatment of genital warts as compared to 1.7% of those with no experience ($p = 0.12$). HPV prevalence was not higher in employees participating in electrosurgical treatment or cryotherapy of genital warts, or loop electrode excision procedure compared with those who did not. HPV 6 or 11 were not detected in any samples. Hand warts after the age of 24 years was more common among dermatology than among non-dermatology personnel (18% vs. 8.0%, $p = 0.03$). Mucosal HPV types are infrequent in the oral and nasal cavity of health care personnel, however, employees at departments of dermato-venereology are at risk of acquiring hand warts. *Key words: human papillomavirus; HPV; plume; LEEP; carbon dioxide laser; genital warts; condyloma acuminatum; hand warts.*

Accepted Jun 10, 2014; Epub ahead of print Jun 18, 2014

Acta Derm Venereol 2015; 95: 173–176.

Kristian Kofoed, Department of Dermato-venereology, Copenhagen University Hospital, Bispebjerg, Bispebjerg Bakke 23, DK-2400 Copenhagen NV, Denmark. E-mail: kristian.kofoed@regionh.dk

Genital warts are caused by human papillomavirus (HPV), which is the most common sexually transmitted infection (STI) (1–3). At least 40 HPV types can infect the genital mucosa (4) and they are classified as oncogenic (high-risk) or non-oncogenic (low-risk) HPV types, on the basis of their association with malignant genital lesions (5). The majority of genital warts is caused by the low-risk HPV types 6 or 11 (6, 7). Molecular and epidemiological studies have demonstrated the role of genital high-risk HPV, in the aetiology of oro-pharyngeal cancers (8, 9) and HPV types 6 and 11 is the cause of both juvenile- and adult-onset laryngeal papillomas (10).

Genital HPV infections mainly spread by direct genital contact, especially in areas of epidermal barrier erosion or friction (11). Other routes of HPV transmission are incompletely understood. Oral HPV infection may be related to orogenital or oral-to-oral spread in adults (12, 13).

Occupational HPV transmission from patient to medical personnel during carbon dioxide (CO_2) laser vaporisation of genital warts or loop electrode excision procedure (LEEP) of cervical dysplasia is controversial. Studies have shown that HPV DNA is present in the plume generated during laser vaporisation of verrucae, laryngeal papillomas and genital warts (14–16). HPV 16 is the most frequently isolated HPV type from plume samples taken during LEEP (17). Two cases of laryngeal papillomatosis among operating personnel who used laser vaporisation to treat genital warts have been described (18, 19). This has resulted in increased use of personal protective equipment and smoke evacuators in Denmark during the last decade; which may be sufficient to protect medical personnel from acquiring HPV infection (20, 21).

To investigate the prevalence of mucosal HPV types in the nasal and oral cavity and history of HPV-related diseases in relation to work exposures, we collected oral rinses, nasal swabs and information on HPV related diseases from medical personnel at departments of gynaecology and dermato-venereology in Denmark. Results from the same cohort have been published in a previous paper (22). That paper focus on the presence

*Part of the data has been presented at the 28th International Papillomavirus Conference (Puerto Rico 2012), The 6th International Conference of HPV, Polyomavirus and Ultraviolet Radiation in Skin Cancer (Berlin 2012) and the 72nd annual meeting of the American Academy of Dermatology (Denver 2014).

 doi: 10.2340/00015555-1912
 ISSN 0001-5555

of HPV types from the *Betapapillomavirus* and *Gammapapillomavirus* genera in the nasal mucosa and does not contain results regarding the work-related exposition to HPV, work place or history of HPV-related diseases.

METHODS

Participants

Medical personnel employed at departments of gynaecology and dermato-venereology was invited to participate. The participating departments of dermato-venereology were located at Copenhagen University Hospital Gentofte and Bispebjerg, Odense University Hospital, and Aarhus University Hospital. The participating departments of gynaecology were located at Copenhagen University Hospital Hvidovre, Herlev and Hilleroed, Odense University Hospital, and Aarhus University Hospital. The total number of medical personnel at the participating departments of dermatology is approximately 290, and approximately 1,320 medical personnel are working at the participating departments of gynaecology. The number of employees with experience of treating patients with HPV-related diseases is not known. Additionally, employees at a department at the medical faculty at Copenhagen University were invited to participate. The study was performed between 2010 and 2011. Participants had to be at least 18 years of age and should not have treated patients with HPV-related diseases for 24 h before sampling. The Scientific Ethical Committee of the Capital Region approved sample collection based on written informed consent (H-D-2010-077). All patients were offered access to their study results and subsequent counselling if needed.

After sampling the participants answered a questionnaire concerning demographic data, previous and current work-related HPV exposure, and history of HPV-related diseases.

Sample collection

An oral rinse was obtained by means of a ≈30 s oral rinse and gargle with 7 ml isotonic saline collected in a 10 ml tube. Nasal samples were collected using a swab applicator with a nylon fibre flocked tip moistened in isotonic saline. The applicator was placed ≈1 cm inside the nostril, and with moderate pressure against the nasal septum rotated 3 times in each nostril. Subsequently the applicator was placed in a cryo tube containing 1.5 ml isotonic saline solution.

Analysis

After purification and amplification a Luminex-based HPV genotyping was used to identify HPV types as previously described (7, 23). The technique allows the detection of at least 39 HPV genotypes of which 15 are high-risk HPV genotypes: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68, 73 and 82; and 5 probable high-risk types genotypes: 26, 53, 66, 67 and 69, and 19 low-risk HPV genotypes: 6, 11, 30, 40, 42, 43, 54, 61, 62, 70, 74, 81, 83, 86, 87, 89, 90, 91 and 114. The Luminex assay also included 2 "universal" HPV probes and samples positive only for the "universal" probe were typed by DNA-sequencing. β-globin real-time PCR was included as a separate test of sample adequacy for PCR (7). Only participants with β-globin positive samples were included in the analysis.

Statistics

Qualitative variables were given as number (percentage) and were studied using Fisher's exact test. Quantitative data were expressed as mean (± standard deviation) or median (range) as appropriate.

Data analysis was conducted using SAS statistical software (SAS Institute Inc., Cary, NC, USA) and GraphPad Prism (GraphPad Software Inc., San Diego, CA, USA). p-values < 0.05 were considered to be statistically significant.

RESULTS

A total of 314 persons participated. Two did not fill out the questionnaire and were excluded and 25 participants from the medical faculty were excluded because they had not worked at a department of dermato-venereology or gynaecology. Characteristics of the 287 persons are given in Table SI[1]. As shown in Table SII[1] there was no difference in the history of HPV-related diseases, warts since the age of 25 years or warts at inclusion between physicians and non-physicians. Participation in CO_2 laser treatment of HPV-related diseases did not influence the reported history of HPV-related diseases. A significantly higher number of employees at a department of dermato-venerology reported having had an HPV related disease in their lifetime (OR 1.9; 95CI% 1.1-3.2).This was mainly due to significantly employees reporting hand warts after the age of 24 years (OR 2.2; 95%CI 1.1-4.5). Three persons, all of whom were employees at a department of dermato-venereology reported nasal or oro-pharyngeal warts after the age of 24 years.

Due to a β-globin negative oral rinse or a nasal swab 16 persons were not included in the analysis of oral or nasal HPV. HPV was isolated from 13 persons, types and sites are given in Table SIII[1]. The HPV type isolated from 2 participants was not regarded as a mucosal HPV type (HPV 10 and a putative subtype of a *Gammapapillomavirus* HPV isolate) and thus not included in the comparative analysis.

A mucosal HPV type was found among 5.8% of employees with experience of laser treatment of genital warts as compared to 1.7% of those with no experience ($p = 0.12$) (Table I). HPV 6 or 11 were not detected in any of the samples, while HPV 16 or 18 were found in 4.

Employees participating in CO_2 laser treatment of genital warts, had been doing the procedure for a median of 5 years (range 0–25). Laser personnel that treated patients with genital warts for at least 5 years had a significantly higher prevalence of mucosal HPV types in the nasal or oral cavity than employees that used CO_2 laser for less than 5 years or never (OR 6.7 (95% CI 1.7–26.0; $p = 0.004$)). The median age of employees with ≥ 5 years experience of CO_2 laser treatment was 49 years (range 36–68), while the median age of personnel with less than 5 years or no experience of laser treatment was 40 years (range 22–64). Persons performing cryotherapy or LEEP did not have a higher prevalence of mucosal HPV compared with those who did not. The prevalence of mucosal HPV was not significantly higher

[1]http://www.medicaljournals.se/acta/content/?doi=10.2340/00015555-1912

Table I. *Mucosal human papillomavirus type in relation to exposure*

Work exposure and position	HPV+ *n* (%)	HPV– *n*	*p*[a]
Treating human papillomavirus (HPV) related diseases			
Yes	11 (4.9)	213	
No	0 (0.0)	47	0.22
CO_2-laser treatment of genital warts			
Yes	9 (5.8)	147	
No	2 (1.7)	113	0.12
Cryotherapy of genital warts			
Yes	4 (4.1)	93	
No	7 (4.0)	167	1.00
Electrosurgical treatment of genital warts			
Yes	6 (6.5)	87	
No	5 (2.8)	173	0.20
Loop electrosurgical excision procedure			
Yes	5 (4.7)	101	
No	6 (4.6)	159	0.76
Occupation			
Physician	8 (5.3)	143	
Non-physician	3 (2.5)	117	0.36
Work place			
Dermato-venerology	6 (6.3)	90	
Gynaecology	5 (2.9)	170	0.18

[a]Fisher's exact test.

in employees participating in electrosurgical treatment of genital warts (Table I).

All persons currently involved in CO_2 laser treatment, reported taking some protective measures against HPV infection. All except 2 (93%) used examination gloves, 88% used smoke evacuators and 79% laser plume masks. The majority (64%) of medical personnel who stopped using CO_2 laser in the treatment of genital warts $\geq$5 years before the study was conducted took no protective measures against HPV infection or wore only examination gloves.

DISCUSSION

In our study we found that participating in cryotherapy, CO_2 laser or electrosurgical evaporation of genital warts or LEEP of cervical dysplasia did not significantly increase the prevalence of nasal or oral HPV.

In a recent study of medical personnel treating laryngeal papillomas and urethral warts using CO_2 laser it was found that after treatment of urethral warts, HPV DNA corresponding to patient tissue specimens was present in all samples obtained from the gloves of the surgeons (21). Oral mucosa samples from all 18 different employees tested HPV negative, as did the surgical mask specimens. Similar results have been found by others (20, 24). Thus, it seems that wearing a laser plume mask or even a surgical mask protects from upper airway HPV infection. Or it may be that regardless of protective measures the risk of HPV infection by inhalation of laser plume is very small. Bellina et al. (24) showed that only a few morphologically intact cells are present in the plume collected during CO_2 laser treatment of genital warts and incubation of the cellular debris resulted in no metabolic activity, replication or transcription of HPV. On the other hand, Garden et al. (25) treated bovine papillomavirus (BPV)-induced cutaneous fibropapillomas on calves with CO_2 laser. The laser plume was collected and then re-inoculated onto the skin of other calves. Fibropapillomas developed at laser plume-inoculated sites.

We did not detect HPV 6 or 11, which is by far most frequently encountered types in genital warts, in a single employee (6, 7). However, we did find an elevated prevalence of nasal or oral HPV in employees that had participated in CO_2 laser for at least 5 years. This may simply be due to an increasing number of nasopharyngeal HPV infections with age, which have previously been shown (13, 26). Only 13 (4.4%) employees had a mucosal HPV positive sample. In another study collected oral mucosa specimens from 18 health care professionals all tested HPV negative (21). The reported prevalence of HPV in oral rinse samples in the United States among men and women aged 14–69 years is 6.9% (13). In that study the prevalence of HPV 16 and 18 were 1% and 0.25%, respectively, which is approximately the same as detected in our study. A similarly low prevalence of high-risk HPV types (0.63%) was found in mouthwash from Swedish healthy controls (27). However, among normal oral cavity scrapings taken with a brush Rautava et al. (28), reported HPV DNA prevalence of 17% in Finnish pregnant women.

We found that employees at a department of dermato-venerology have a greater prevalence of hand warts and possibly nasal or oral warts, than employees at departments of gynaecology. In a study based on a self-administered questionnaire, 4 out of 570 U.S. CO_2 laser surgeons reported a history with nasopharyngeal warts. This small number was significantly higher than the number observed in matched control subjects (29). However, when warts were grouped together without specification of anatomic site the reported prevalence of warts among the laser surgeons was not significantly different from the prevalence in the control group (29). Lobraico et al. (30) conducted a survey by a questionnaire sent to laser users, and discovered a significantly higher prevalence of acquired hand warts among the dermatologists compared to other specialties. However, genital warts and warts of the skin are induced by an entirely different set of HPV.

Our study has some limitations. Detection of HPV is dependent on sampling procedure and assay sensitivity and specificity. In the assay and sampling procedures we used may not have been able to detected low productive infections e.g. in the tonsillar crypts. When asked in hindsight every specialised health care professional may have a stronger recall of diseases, which belong to their own area of experience than the ones that do not. Even though this study is by far the largest on the prevalence of nasopharyngeal HPV in medical personnel it might still be too small to detect important

infrequent associations between nasopharyngeal HPV prevalence and work exposure. Most HPV infections are transient and thus the prevalence deduced by point testing with swap and rinses are not able to measure the transmission rate over a period of years.

ACKNOWLEDGEMENTS

We thank Aline Marshall and the laboratory staff at Medical Microbiology, Malmö, for careful performance of the HPV analysis, Helle Wium Sørensen who provided excellent and enthusiastic assistance in planning sample collection and study logistics, and statistician Andreas Habicht, Significans Aps for data handling and analysis.

Conflicts of interest. KK has received fees as a speaker and obtained research grants form Sanofi Pasteur MSD. CS has obtained research grants and received fees as a speaker and for Sanofi Pasteur MSD. KGM is the former managing director at Sanofi Pasteur MSD Denmark. The study was supported by a research grant from Sanofi Pasteur MSD, Denmark.

REFERENCES

1. Trottier H, Franco EL. The epidemiology of genital human papillomavirus infection. Vaccine 2006; 24 Suppl 1: S1–15.
2. Nielsen A, Kjaer SK, Munk C, Iftner T. Type-specific HPV infection and multiple HPV types: prevalence and risk factor profile in nearly 12,000 younger and older Danish women. Sex Transm Dis 2008; 35: 276–282.
3. Skaaby S, Kofoed K. Anogenital warts in Danish men who have sex with men. Int J STD AIDS 2011; 22: 214–217.
4. Lacey CJ, Lowndes CM, Shah KV. Chapter 4: Burden and management of non-cancerous HPV-related conditions: HPV-6/11 disease. Vaccine 2006; 24 Suppl 3: S3–35–S3/41.
5. Lorincz AT, Reid R, Jenson AB, Greenberg MD, Lancaster W, Kurman RJ. Human papillomavirus infection of the cervix: relative risk associations of 15 common anogenital types. Obstet Gynecol 1992; 79: 328–337.
6. Garland SM, Steben M, Sings HL, James M, Lu S, Railkar R, et al. Natural history of genital warts: analysis of the placebo arm of 2 randomized phase III trials of a quadrivalent human papillomavirus (types 6, 11, 16, and 18) vaccine. J Infect Dis 2009; 199: 805–814.
7. Sturegard E, Johansson H, Ekstrom J, Hansson BG, Johnsson A, Gustafsson E, et al. Human papillomavirus typing in reporting of condyloma. Sex Transm Dis 2013; 40: 123–129.
8. Gillison ML. Human papillomavirus-associated head and neck cancer is a distinct epidemiologic, clinical, and molecular entity. Semin Oncol 2004; 31: 744–754.
9. Schwartz SM, Daling JR, Doody DR, Wipf GC, Carter JJ, Madeleine MM, et al. Oral cancer risk in relation to sexual history and evidence of human papillomavirus infection. J Natl Cancer Inst 1998; 90: 1626–1636.
10. Mounts P, Shah KV, Kashima H. Viral etiology of juvenile- and adult-onset squamous papilloma of the larynx. Proc Natl Acad Sci U S A 1982; 79: 5425–5429.
11. Oriel JD. Natural history of genital warts. Br J Vener Dis 1971; 47: 1–13.
12. Rintala M, Grenman S, Puranen M, Syrjanen S. Natural history of oral papillomavirus infections in spouses: a prospective Finnish HPV family study. J Clin Virol 2006; 35: 89–94.
13. Gillison ML, Broutian T, Pickard RK, Tong ZY, Xiao W, Kahle L, et al. Prevalence of oral HPV infection in the United States, 2009–2010. JAMA 2012; 307: 693–703.
14. Ferenczy A, Bergeron C, Richart RM. Human papillomavirus DNA in CO2 laser-generated plume of smoke and its consequences to the surgeon. Obstet Gynecol 1990; 75: 114–118.
15. Kashima HK, Kessis T, Mounts P, Shah K. Polymerase chain reaction identification of human papillomavirus DNA in CO2 laser plume from recurrent respiratory papillomatosis. Otolaryngol Head Neck Surg 1991; 104: 191–195.
16. Garden JM, O'Banion MK, Shelnitz LS, Pinski KS, Bakus AD, Reichmann ME, et al. Papillomavirus in the vapor of carbon dioxide laser-treated verrucae. JAMA 1988; 259: 1199–1202.
17. Sood AK, Bahrani-Mostafavi Z, Stoerker J, Stone IK Human papillomavirus DNA in LEEP plume. Infect Dis Obstet Gynecol 1994; 2: 167–170.
18. Hallmo P, Naess O. Laryngeal papillomatosis with human papillomavirus DNA contracted by a laser surgeon. Eur Arch Otorhinolaryngol 1991; 248: 425–427.
19. Calero L, Brusis T. Larynxpapillomatose – erstmalige Anerkennung als Berufskrankheit bei einer OP-Schwester. Laryngorhinootologie 2003; 82: 790–793.
20. Weyandt GH, Tollmann F, Kristen P, Weissbrich B. Low risk of contamination with human papilloma virus during treatment of condylomata acuminata with multilayer argon plasma coagulation and CO(2) laser ablation. Arch Dermatol Res 2011; 303: 141–144.
21. Ilmarinen T, Auvinen E, Hiltunen-Back E, Ranki A, Aaltonen LM, Pitkaranta A. Transmission of human papillomavirus DNA from patient to surgical masks, gloves and oral mucosa of medical personnel during treatment of laryngeal papillomas and genital warts. Eur Arch Otorhinolaryngol 2012; 269: 2367–2371.
22. Forslund O, Johansson H, Madsen GK, Kofoed K. The nasal mucosa contains a large spectrum of human papillomavirus types from the Betapapillomavirus and Gammapapillomavirus genera. J Infect Dis 2013; 208: 1335–1341.
23. Söderlund-Strand A, Carlson J, Dillner J. Modified general primer PCR system for sensitive detection of multiple types of oncogenic human papillomavirus. J Clin Microbiol 2009; 47: 541–546.
24. Bellina JH, Stjernholm RL, Kurpel JE. Analysis of plume emissions after papovavirus irradiation with the carbon dioxide laser. J Reprod Med 1982; 27: 268–270.
25. Garden JM, O'Banion MK, Bakus AD, Olson C. Viral disease transmitted by laser-generated plume (aerosol). Arch Dermatol 2002; 138: 1303–1307.
26. Forslund O, Johansson H, Madsen KG, Kofoed K. The nasal mucosa contains a large spectrum of human papillomavirus types from the betapapillomavirus and gammapapillomavirus genera. J Infect Dis 2013; 208: 1335–1341.
27. Hansson BG, Rosenquist K, Antonsson A, Wennerberg J, Schildt EB, Bladstrom A, et al. Strong association between infection with human papillomavirus and oral and oropharyngeal squamous cell carcinoma: a population-based case-control study in southern Sweden. Acta Otolaryngol 2005; 125: 1337–1344.
28. Rautava J, Willberg J, Louvanto K, Wideman L, Syrjanen K, Grenman S, et al. Prevalence, genotype distribution and persistence of human papillomavirus in oral mucosa of women: a six-year follow-up study. PLoS One 2012; 7: e42171.
29. Gloster HM, Jr., Roenigk RK. Risk of acquiring human papillomavirus from the plume produced by the carbon dioxide laser in the treatment of warts. J Am Acad Dermatol 1995; 32: 436–441.
30. Lobraico RV, Schifano MJ, Brader KR. Acquired HPV lesions compared in laser and nonlaser users. J Gynecol Surg 1989; 5: 77–85.