

## Genital Human Papilloma Viruses (HPV) Infection in Men in Hong Kong

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### Abstract

**Aim:** To investigate the prevalence and typing of HPV infection in men in Hong Kong. **Materials and Methods:** Penile swabs from 32 consecutive patients attending a genital urinary clinic were tested after informed consents were obtained. DNA was extracted using FavorPrep™ Tissue Genomic DNA Extraction Kit (Favorgen Biotech Corp). HPV genotypes were detected using SNIPER HPV Genotyping Diagnosis Kit (Genetel Shenzhen). Twenty-nine HPV genotypes are covered including HPV 6, 11, 16, 18, 26, 31, 33, 35, 39, 40, 42, 43, 44, 45, 51, 52, 53, 54, 55, 56, 57, 58, 59, 66, 67, 68, 69, 73 and 82. The reaction was performed according to manufacturer's instructions. Briefly, L1 gene of HPV was amplified with biotinylated primers. The PCR products were added to the membranes for hybridization. The signals were developed and then scanned using GenoCam-9600 scanner. **Results:** A total of 17 penile swabs were HPV positive (53%). In this HPV-positive patient cohort, 12 penile swabs (70.6%) contained HPV high- risk types of 16, 33, 52, 58, 66, 68 and 73 were identified. There are 4 cases of HPV 58, 3 cases of HPV 68 and 2 cases of HPV 52, while one case for each of the remaining types, one sample with mixed infection of HPV 33 and 58. There were 7 cases with mixed HPV infection and 6 cases contained at least one high-risk type. The HPV infection age group in the present study was 38-74 years old. A high prevalence of high- risk HPV infection (37.5%) was determined in men with history of casual sex. **Conclusion:** HPV infection in men causes genital wart, penile, oral and other carcinomas. Most importantly, the high-risk HPV infection may be associated with cervical intraepithelial neoplasms in the female sexual partner of the patient. The two available HPV vaccines, Gardasil® and Cervarix® provide protection against 2 high-risk HPV types, HPV-16 and HPV-18. In this study, 7 different types of high- risk HPV were found including HPV-16, but HPV-58 and HPV-68 are not uncommon. In the future, when new HPV vaccine is to be developed, a wider range of high risk HPV should be covered in this region.

Key words: Human Paillomarvirus (HPV), Genital swab

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## Introduction

Genital warts are one of the common sexually transmitted diseases (STD) in the world. The symptom of the disease sometimes causes physical discomfort including pain, itchiness and bleeding.<sup>1</sup> Infected men may have psychosocial distress such as shame and embarrassment and affect the quality of life.<sup>2</sup> The major of the genital warts and benign epithelial mucosal tumors are related to Human papillomavirus (HPV) type 6 and type 11. Genital warts are not life-threatening and about 70% of infection clearing within 1 year.<sup>3</sup>

A previous survey of genital warts was conducted during 5-8 January 2010, where a total of 170 private doctors and all doctors working in the 5 sexually transmitted disease clinic in the public hospital participated. It was investigated that the incidence rate of genital warts was about 292.2 and 124.9 per 100,000 persons in males and females respectively. Two hundred and ninety-seven new cases of disease in men were found during the data collection period.<sup>4</sup>

Another recent study was investigated that male anogenital warts was not only associated with low risk type 6 and 11, but also co-infected with other types of HPV (26.2%). Other HPV types included HPV 39, 51, 52, 55, 59, 61, 62, 68, 58, 72, 81, 83, 84 and CP6108.<sup>5</sup>

The aim of our current study is to determine the scope of HPV infection in men in Hong Kong and to help planning strategy of health education in the prevention of genital HPV infection. With better understanding of the prevalence of high- risk HPV type infection, it may help to develop more effective HPV vaccine in the future, that would cover a wider range of HPV types while in the meantime to employ presently available HPV vaccine for HPV protection.

## Materials and Method

### *Patient Recruitment*

32 consecutive patients (Range: 38-74 year old and median: 52.5 year old) attending a private sexually transmitted diseases (STD) clinic for the check up or treatment and who had history of previous casual sex were recruited. Their participation in the project and result of their HPV test would not affect the current clinical management of their health condition. A written "Donor Information" sheet was issued to each participant and they were to sign an informed consent.

### *Selection Criteria*

The inclusion criteria for participants in this project were:

- (1) All male patients at or above age of 18 attending the clinic;

- (2) Asymptomatic or symptomatic patients
- (3) Patients did not receive any kind of treatment for genital HPV infection within one month of specimen collection.
- (4) All patients signed consent form after the content of informed consent was explained and was understood.

### *Investigation Methods*

DNA was extracted from the penile swabs using FavorPrep™ Tissue Genomic DNA Extraction Kit (Favorgen Biotech Corp). HPV genotypes were detected using SNIPER HPV Genotyping Diagnosis Kit (Genetel Shenzhen). Twenty-nine HPV genotypes are covered including HPV 6, 11, 16, 18, 26, 31, 33, 35, 39, 40, 42, 43, 44, 45, 51, 52, 53, 54, 55, 56, 57, 58, 59, 66, 67, 68, 69, 73 and 82. The reaction was performed according to manufacturer's instructions. Briefly, L1 gene of HPV was amplified with biotinylated primers at 50°C for 5 min, 95°C for 10 min and then followed by 45 cycles of denaturation at 95°C for 30 sec, annealing at 52°C for 45 sec and extension at 65°C for 30 sec. The reaction was further amplified at 65°C for 5 min. The PCR products were added to the membranes for hybridization. The signals were developed and then scanned using GenoCam-9600 scanner.

### **Results**

All samples indicated a sufficient amount of DNA concentration by beta-actin PCR. Regardless of the breakdown of the HPV types, a total of 17 patients were associated with HPV infection (53%; 17/32). In this

HPV-positive patient cohort, 12 penile swabs (70.6%; 12/17) contained HPV high-risk types of 16 (1), 33 (1), 52 (2), 58 (4), 66 (1), 68 (3) and 73 (1) with one case of mixed high-risk HPV types of 33 and 58.

Ten swabs (58.8%; 10/17) contained HPV low-risk types of 6 (2), 11 (3), 40 (2), 43 (4), 44 (2), 54 (1), 55 (2) with 4 cases of co-infection. There were 4 cases of mixed types (high-risk and low-risk) co-infection. A high prevalence of high-risk HPV infection (37.5%; 12/32) was determined in men with history of casual sex.

The samples from 2 patients with clinical genital wart and showed mixed subtype infection of lower-risk and high-risk HPV. One sample demonstrated mixed infection of lower-risk type 11, 40, 44 with high-risk type 73. The other case revealed mixed infection of lower-risk type 6, 55 with high-risk type 58.

Among the 32 patients participated in the study only 4 patients (12.5%; 4/32) did not have any clinical sign or diagnosis of STD. The most common STD diagnosed in this group was non-gonococcal urethritis, 13 cases out of 32 patients (40.6%; 13/32).

But with the 17 cases infected with HPV only 3 (17.6%; 3/17) cases of co-infection with Non Gonococcal Urethritis (NGU) were present.

### **Discussion**

Data regarding the prevalence of HPV

infection in men are few. Most studies have been concentrated on HPV infection relating to carcinoma of cervix and a lesser part to carcinoma of vulva and vagina. Understanding the relationship of high risk HPV infection (especially HPV-16 and HPV-18) to the development of cancer of cervix led to the development of HPV vaccine including the quadrivalent HPV vaccine.

Furthermore, better treatment and prevention of HPV infection in men may help to decrease the number of HPV infection and related development of cervical, penile and other cancer in the society.

The overall HPV infection in the present study was 53%, meaning 1 out of 2 patients attending the STD clinic with history of casual sex did have a high pick up rate of HPV infection of more than 50%. There is a high infection rate of high- risk HPV (70.5%; 12/17) among all the HPV positive samples. The 12 penile swabs contained HPV high-risk types of 16(1), 33(1), 52(2), 58(4), 61(1), 68(3) and 73(1) with 1 case of high-risk type co-infection. In the previous studies of prevalence of HPV infection in penile cancer, it has been shown a strong association between HPV infection and penile cancer.<sup>6</sup> and HPV-16 infection had the highest prevalence.<sup>7-11</sup> With the right age group and harboring the high risk HPV, it makes this group of patient at risk of developing penile cancer.

HPV-16 and HPV- 18 accounted for about 70% of all cervical cancers.<sup>12</sup> A subset of

high- risk HPV types (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68, 73, 82) is associated with more than 99% of the cervical cancers. With high prevalence of high risk HPV infection in men in this study, they may pose great risk of transmitting HPV infections to their female sexual partners with an infective rate of more than 60%<sup>12</sup> and to the development of genital warts and cervical cancer.

Penile cancer is a rare malignancy occurring mostly in older man between 50-70 years.<sup>13</sup> The prevalence of overall HPV infection and high- risk HPV infection in men between age 41 to 60 years in this study was high, showing that this group of men was at risk of developing penile cancer. The prevalence of HPV infection in this study further supported to the close relationship between HPV infection and the development of penile cancer.

The prevalence of genital wart is highest among men aged between 25 to 29 years old.<sup>14</sup> There were 3 cases of clinical genital warts found in this study, 1 case each (5.8%) at age of 38, 46 and 52 years old. These cases showed one low-risk HPV 11 infection, and mixed HPV infection, HPV- 6, HPV- 55, HPV- 58 in the case of age 46 years old and HPV-11, 40, 44 and HPV-73 in the other case of age 52 years old. The HPV diagnosed in this study was by molecular method and not clinical diagnosis which may explain the difference of age difference from that described by Insinga et al.<sup>14</sup>

He et al. indicated that the dominant HPV

types were 16 (17.4%), 3 (16.4%), 57 (7.9%), 18 (7.2%), 87 (4.6%) and 58 (3.3%) in the genital swab samples in rural China where sequencing technique was applied.<sup>15</sup> In Brazil, Freire et al. showed that HPV 6 (17.7%) was most dominant type, followed by HPV 42 (13.8%), HPV 16 (11.8%) and HPV 51 (11.0%) respectively. 錯誤! 找不到參照來源。

The variation of HPV genotypes in different geographical area may be due to genetics disparity, traveling and immigration of people.

In a study of epidemiology of HPV infection in women in Hong Kong Chan et al., showed that the most common HPV infection were HPV-16 (18.3%), 52 (18.3%), 58 (17.7%) and 18 (9.1%).<sup>17</sup> In another study of the prevalence of HPV infection in cervical carcinoma, Chan et al., showed that the prevalence of HPV types were as follows, HPV-16 (59.6%), 18 (23.9%), 52 (13.2%) and 58 (8.8%).<sup>18</sup>

In Chan et al., (2009a) (2009b) studies the HPV types 16 and 18 were mostly associated with carcinoma of cervix followed by HPV-52 and 58.<sup>17,18</sup>

In the present study, the prevalence of high risk HPV present in penile swab samples represented HPV-58 (33.3%), HPV-68 (25%), HPV-52 (25%) and HPV-66, 33 and 16 (each 8.3%). So the prevalence of HPV-16, 52, 58 in this study coincides with the finding of Chan et al., (2009a, 2009b), although we did not find HPV 18 and also the prevalence of HPV 16 was lower than expected.<sup>17,18</sup>

Men in Hong Kong are encouraged to have HPV vaccination. Currently the quadrivalent HPV vaccine is recommended to men age between 9 to 26 years old in Hong Kong. In the future development of HPV vaccine a wider range of HPV high risk types should be covered in this region.

The patients who were demonstrated with HPV infection in this study should be further given three doses of bivalent vaccine and then recheck for clearance of HPV infection.

## Conclusion

In this study, a high incidence of HPV infection was present in men who had previous casual sex history and 7 different types of high risk HPV were found including HPV-16 but HPV-58 and HPV-68 are not uncommon. The available HPV vaccine, bivalent and quadrivalent vaccine provides protection against 2 high- risk HPV, HPV 16 and HPV 18.

In the future, when new HPV vaccine is to be developed, a wider range of high risk HPV should be covered in this region.

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