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Prevalence and distribution of HPV types in genital warts in Xi'an, China: a prospective study

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ABSTRACT

Objectives To characterise the prevalence and distribution of human papillomavirus (HPV) types in genital warts in Xi'an, China.

Methods This prospective study was conducted in Shaanxi Provincial Institute for Skin Disease and STD Control (SPISSC) between September 2014 and April 2017. Genital wart samples were obtained from 879 patients, including 512 men and 367 women. HPV genotyping was performed by using an automatic nucleic acid hybridisation system.

Results Of the 879 patients with genital warts, the detectable rates of low-risk, high-risk and total HPV types were 45.4%, 34.5% and 57.8%, respectively. The detectable rate of low-risk HPV types (45.4%) was significantly higher than that of high-risk HPV types (34.5%) (χ^2=21.85, p<0.01). The detectable rate of low-risk HPV types of men (52.3%) was significantly higher than that of women (35.7%) (χ^2=23.90, p<0.01). The detectable rates of one HPV type infection and two and three or more HPV type coinfections were 26.1%, 17.5% and 14.2%, respectively. HPV6 (24.9%), HPV11 (17.9%), HPV52 (9.9%) and HPV16 (7.3%) were the four most common HPV types.

Conclusions The results of this study suggest that low-risk HPV types are major pathogens of genital warts, but high-risk HPV type infections and multiple HPV type coinfections are also common in genital warts. HPV6, 11, 52 and 16 are the four most common HPV types in genital wart in Xi'an, China.

Strengths and limitations of this study

- This is the first study on the prevalence and distribution of HPV types in genital warts in Xi'an, China.
- Diagnosis of genital warts based on clinical manifestations might lead to bias, because infectious soft warts might be misdiagnosed as genital warts due to similar clinical manifestations.
- Some patients with genital warts did not participate because they feared this study might give away their privacy or they visited other convenient hospitals nearby.

INTRODUCTION

Genital warts are a kind of sexually transmitted diseases caused by certain types of human papillomavirus (HPV). The main clinical manifestation of genital warts is benign hyperplasia of the skin and mucous membrane in the genitalia, anus and perineum. Genital warts not only affect physiological function but also cause psychological stress. HPV is the most common sexually transmitted infection globally, and most people are infected at some point in their lives.[1] Genital HPV infections have a prevalence of 10%–20% in the USA, and 1% of sexually active adults have clinical manifestations.[2] In China, the prevalence of genital HPV infections is 16%–18%.[3]

To date, 201 different HPV types have been identified. More than 40 of these types can infect the mucous membrane of the anus and genitalia and have been classified as probably low-risk (HPV6, 11, 40, 42, 43, 44, 54, 61, 62, 71, 72, 81, 83, 84 and 89) or possibly high-risk (HPV26, 30, 34, 53, 66, 67, 68, 69, 70, 73, 82, 85 and 97) and high-risk (HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58 and 59), depending on their ability to lead to malignant progression.[4] A variety of HPV types can cause genital warts, but HPV6 and 11 together account for about 90% of all cases.[5] HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68, 73 and 82 are considered carcinogenic, but 70% of cervical cancers are caused by HPV16 and 18.[6] Three vaccines have been approved to prevent HPV infections. The vaccines consist of virus-like particles derived from HPV major capsid protein (L1). Cervarix is bivalent to protect against HPV16 and 18. Gardasil is a recombinant quadrivalent vaccine to protect against HPV6, 11, 16 and 18. Gardasil-9 is nonavalent to protect against HPV6, 11, 16, 18, 31, 33, 45, 52 and 58 and can prevent about 90% of cervical, vulvar, vaginal and anal cancers.[7] The reduction of genital wart incidence has been reported in countries where HPV vaccination programmes have been adopted.[8] In 2016, China Food and Drug Administration approved HPV vaccine to prevent cervical cancer and other HPV-associated diseases.

However, the distribution of HPV types varies from country to country or region to region.[9] At present, the prevalence and distribution of HPV types in genital warts are not clear in Xi'an, China. We conducted this study before implementing an HPV vaccination programme. One objective is to characterise the prevalence of HPV infections and distribution of HPV types in Xi'an, China, in order to further evaluate the effectiveness of the vaccine, as data showed a decline in vaccine-related HPV types in regions covered by HPV vaccination.[10] Another objective is to assess whether commercially available HPV vaccines can cover the most common HPV types in order to select a suitable vaccine for Xi'an, China.

METHODS

Study design

This study was a prospective study conducted from September 2014 to April 2017. Newly diagnosed patients with genital warts in Shaanxi Provincial Institute for Skin Disease and STD Control (SPISSC) were invited to participate in the study. The specimens were collected from lesions of genital warts, and 23 HPV types were detected.

Diagnostic criteria of genital warts

Patients were diagnosed according to the Diagnostic Criteria of Genital Warts of China.[11] Briefly, genital warts were diagnosed based on epidemiological history and clinical manifestations. The epidemiological histories included unsafe sex, sexual partners with genital warts or multiple sexual partners. The clinical manifestations were papuloid lesions, which might gradually develop into papillary, coronal or cauliflower-like masses. When skin lesions were atypical, acetic acid white test was used to confirm the diagnosis.

Participants

Between September 2014 and April 2017, a total of 912 patients were diagnosed with genital warts in SPISSC. Twenty-two of these patients were excluded from the study because they were not residents of Xi'an. Eleven of the patients refused to participate because they feared the study might reveal their privacy. The remaining 879 patients, including 512 men and 367 women, participated in this study. Their ages ranged from 16 to 68 years, and no one had been vaccinated against HPV prior to enrolment.

Specimen collection

We used sampling brushes to collect specimens from lesions of genital warts. The specimens were eluted from the sampling brushes with preservation solution and were stored in the refrigerator at −20°C before detecting HPV types. The sampling brushes and preservation solutions were provided by Yaneng BIOscience (Shenzhen) Co.

HPV genotyping

We used an automatic nucleic acid hybridisation system manufactured by Yaneng BIOscience (Shenzhen) Co. to detect HPV types. All procedures were carried out following the manufacturer's instructions. This system used HPV DNA amplification and hybridisation to simultaneously detect low-risk HPV types, HPV6, 11, 42, 43 and 81, and high-risk HPV types, HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 68, 73, 82 and 85. Quality control measures were implemented during HPV detection.

Statistical analysis

All data were analysed using SPSS software V.16.0. R×Cχ^2 test was used for comparing differences among groups, with $\alpha \leq 0.05$ considered statistically significant. A correlation coefficient was calculated to quantify the statistical relationship between two values, with $\alpha \leq 0.05$ considered significant.

Patient and public involvement

Specimens of patients were collected from lesions of genital warts. Patients were not further involved in the implementation of this study.

RESULTS

Prevalence of low-risk and high-risk HPV types in genital warts

Specimens of 879 patients with genital warts, including 512 men and 367 women, were tested for low-risk HPV type (HPV6, 11, 42, 43 and 81) and high-risk HPV type (HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 68, 73, 82 and 85). As shown in table 1, of the 512 men, 268 were infected by low-risk HPV types (52.3%), 166 by high-risk HPV types (32.4%) and 308 by total HPV types (60.2%). Of the 367 women, 131 were infected by low-risk HPV types (35.7%), 137 by high-risk HPV types (37.3%) and 200 by total HPV types (54.5%). The detectable rate of low-risk HPV types of men (52.3%) was significantly

Table 1 Prevalence of low-risk and high-risk HPV types in genital warts in Xi'an (n=879)

Sex	n	Low-risk HPV (%)	High-risk HPV (%)	Total (%)
Men	512	268 (52.3)	166 (32.4)	308 (60.2)
Women	367	131 (35.7)	137 (37.3)	200 (54.5)
Total	879	399 (45.4)	303 (34.5)	508 (57.8)

HPV, human papillomavirus.

Table 2 Prevalence of low-risk and high-risk HPVs in age groups in Xi'an (n=879)

Age (years)	n (%)	Low-risk HPV (%)	High-risk HPV (%)	Total (%)
15-19	17 (1.9)	10 (58.8)	6 (35.3)	11 (64.7)
20-24	152 (17.3)	66 (43.4)	51 (33.6)	84 (55.3)
25-29	232 (26.4)	102 (44.0)	81 (34.9)	127 (54.7)
30-34	147 (16.7)	62 (42.2)	45 (30.6)	77 (52.4)
35-39	101 (11.5)	46 (45.5)	39 (38.6)	63 (62.4)
40-44	88 (10.0)	33 (37.5)	28 (31.8)	48 (54.5)
45-49	73 (8.3)	37 (50.7)	25 (34.2)	48 (65.8)
50-54	43 (4.9)	25 (58.1)	18 (41.9)	31 (72.1)
55-59	15 (1.7)	11 (73.3)	7 (46.7)	12 (80.0)
≥ 60	11 (1.2)	7 (63.6)	3 (27.3)	7 (63.6)
Total	879 (100.0)	399 (45.4)	303 (34.5)	508 (57.8)

HPV, human papillomavirus.

higher than that of women (35.7%) (χ^2=23.90, p<0.01), while there was no significant difference in the detection rate of high-risk HPV types between men (32.4%) and women (37.3%) (χ^2=2.28, p=0.13). The total detectable rate of low-risk, high-risk and total HPV types were 45.4%, 34.5% and 57.8%, respectively. The total detectable rate of low-risk HPV types (45.4%) was significantly higher than that of high-risk HPV types (34.5%) (χ^2=21.85, p<0.01).

Prevalence of low-risk and high-risk HPV types in age groups

As shown in table 2, we divided patients with genital warts into 10 age groups. The detectable rates of low-risk HPV types peaked in the ages of 15–19 and 55–59 years, while those of high-risk HPV types peaked in the age of 55–59 years.

Prevalence of each HPV type in genital warts

As shown in table 3, the detectable rates of HPV6 (24.9%), HPV11 (17.9%), HPV52 (9.9%) and HPV16 (7.3%) ranked the top 4 among the 23 HPV types detected. The detectable rate of HPV6 (24.9%) was significantly higher than that of HPV11 (17.9%) (χ^2=13.00, p<0.01), while that of HPV11 (17.9%) was significantly higher than that of the HPV52 (9.9%) (χ^2=23.32, p<0.01), and that of HPV52 (9.9%) was significantly higher than that of HPV16 (7.3%) (χ^2=3.84, p=0.05). So, HPV6, 11, 52 and 16 were the four most common HPV types in genital warts in Xi'an, China.

Multiple HPV type coinfections in genital warts

As shown in table 4, the detectable rate of one HPV type infections was 26.1%, while those of two and three or more HPV type coinfections were 17.5% and 14.2%, respectively. The detectable rate of one HPV type infection (26.1%) was significantly higher than that of two HPV type coinfections (17.5%) (χ^2=18.77, p<0.01) and that of three or more HPV type coinfections (14.2%) (χ^2=38.26, p<0.01). The number of HPV types in genital warts varied from 1 to 13. The number of HPV types had a negative correlation with the detectable rate (r=−0.7258, p=0.05), but there was no correlation between the number of HPV types and age (r=−0.0086 , p=0.50).

DISCUSSION

Significances of studying the prevalence and distribution of HPV types

HPV, as one of the seven notorious carcinogenic viruses, has been studied worldwide. Genital warts are the most common clinical manifestation of HPV infections. Although not life-threatening, genital warts carry a huge psychological and economic burden. Patients with genital warts are disturbed by shame and embarrassment, and fear of recurrence and transmission. Coinfections with multiple HPV types are possible and may combine both low-risk and high-risk types.[12] In recent years, significant progress has been made in the control and prevention of HPV infections, and HPV vaccines have been used successfully to prevent HPV-associated cancers and genital warts.[13] However, there are 201 HPV types and their distribution varies from region to region.[14 15] While the vaccines protect people in some areas, they cannot protect people in others because of regional differences in the distribution of HPV types. Therefore, it is important to study the prevalence and distribution of HPV types in specific regions before implementing the vaccination programme.

Prevalence of low-risk and high-risk HPV types in genital warts

Prevalence of low-risk and high-risk HPV types in genital warts varies from report to report. A previous multinational study in men reported that the detectable rates of low-risk and high-risk HPV types in genital warts were 73.2% and 15.6%, respectively.[16] A study in women from Australia found that HPV was detected in 90.8% of genital warts, with HPV6/11 in 86% and high-risk HPV types in 31%.[17] Another study in men from Hong Kong reported that HPV6/11 was found in 63.1% and HPV16/18 was

Table 3 Distribution of each HPV types in genital warts in Xi'an (n=879, including 512 men and 367 women)

Type	Men (%)	Women (%)	Total (%)	Rankings
Low-risk HPV				
6	150 (29.3)	69 (18.8)	219 (24.9)	1
11	110 (21.5)	47 (12.8)	157 (17.9)	2
42	27 (5.3)	20 (5.4)	47 (5.3)	5
43	33 (6.4)	14 (3.8)	47 (5.3)	5
81	30 (5.9)	16 (4.4)	46 (5.2)	6
High-risk HPV				
52	54 (10.5)	33 (9.0)	87 (9.9)	3
16	33 (6.4)	31 (8.4)	64 (7.3)	4
58	24 (4.7)	22 (6.0)	46 (5.2)	6
68	30 (5.9)	9 (2.5)	39 (4.4)	7
51	24 (4.7)	14 (3.8)	38 (4.3)	8
56	22 (4.3)	16 (4.4)	38 (4.3)	8
53	18 (3.5)	19 (5.2)	37 (4.2)	9
18	22 (4.3)	13 (3.5)	35 (4.0)	10
66	19 (3.7)	12 (3.3)	31 (3.5)	11
59	17 (3.3)	13 (3.5)	30 (3.4)	12
39	10 (2.0)	9 (2.5)	19 (2.2)	13
33	5 (1.0)	11 (3.0)	16 (1.8)	14
31	8 (1.6)	6 (1.6)	14 (1.6)	15
73	9 (1.8)	4 (1.1)	13 (1.5)	16
35	7 (1.4)	5 (1.4)	12 (1.4)	17
85	4 (0.9)	3 (0.8)	7 (0.8)	18
45	5 (1.0)	1 (0.3)	6 (0.7)	19
82	1 (0.2)	0 (0.0)	1 (0.1)	20
Total	308 (60.2)	200 (54.5)	508 (57.8)	–

HPV, human papillomavirus.

found in 9.2% of genital warts.[18] Our study found that the detectable rates of low-risk and high-risk HPV types were 45.4% and 34.5% in genital warts in Xi'an, China. The above reports and our study indicate that low-risk HPV types are major pathogens of genital warts, but high-risk HPV types in genital warts are also common and can act as reservoirs of cancer-related HPV types to threaten local populations.

Table 4 Prevalence of one and multiple HPV type coinfections in genital warts in Xi'an (n=879)

HPV type (n)	n (%)	Age:$\bar{x}\pm s$
1	229 (26.1)	34.9±10.8
2	154 (17.5)	32.5±10.5
3	65 (7.4)	34.2±10.8
4	20 (2.3)	33.5±10.2
5	20 (2.3)	30.5±6.8
6	10 (1.1)	31.3±14.5
7	8 (0.9)	32.0±13.0
8	1 (0.1)	34.0±0.0
13	1 (0.1)	22.0±0.0
Total	508 (57.8)	33.1±10.0

HPV, human papillomavirus.

The most common HPV types in different regions

One study from the USA found that HPV6 (43.8%), HPV11 (10.7%) and HPV16 (9.8%) were the most common types detected in genital warts in men.[19] Another study from Colombia found that HPV6 was the most common type in both women (62%) and men (56%), followed by HPV11 (20%). HPV16 ranked third in prevalence, where 16% of patients tested positive in genital warts.[20] A study from seven regions of China reported that the most common types were HPV6 (41.3%), HPV11 (37.6%) and HPV16 (10.4%) in genital warts.[21] Our study found that HPV6 (24.9%), HPV11 (17.9%), HPV52 (9.9%) and HPV16 (7.3%) were the most common types in genital warts in Xi'an, China. Consistent with our study, a study from Guangdong, China, found that the most common types

were HPV6 (42.2%), HPV11 (39.3%), HPV52 (7.7%) and HPV16 (7.56%) in genital warts.[22] The above reports and our study indicate that HPV6, 11 and 16 are the most common types in genital warts in most parts of the world. In addition, HPV52 is one of the most common types in Xi'an and Guangdong, China. Cervarix against HPV16 and 18 does not cover HPV6, 11 and 52, while Gardasil against HPV6, 11, 16 and 18 does not cover HPV52. Compared with Cervarix and Gardasil, Gardasil-9 against HPV6, 11, 16, 18, 31, 33, 45, 52, 58, covering the four most common HPV types (HPV6, 11, 52 and 16), may be more suitable for Xi'an, China.

Sex differences in HPV infection

In our study, the detectable rate of low-risk HPV types of men was significantly higher than that of women (52.3% vs 35.7%, p<0.01). However, there was no significant difference in the detectable rate of high-risk HPV types between men and women (32.4% vs 37.3%, p=0.13). Consistent with our study, the prevalence of low-risk HPV types in South Africa was higher for men (33.2%) than for women (14.0%), but the prevalence of high-risk HPV types was similar for men (22.4%) and women (22.7%).[23] In contrast, a study in Liuzhou, China, found no significant difference in the prevalence of low-risk HPV types between men and women (1.2% vs 1.4%, p=0.68), while the prevalence of high-risk HPV types of men was lower than that of women (9.4% vs 18.7%, p<0.01).[24] Widdice *et al* suggested that women may have a higher incidence rate or lower clearance rate of HPV infections than men due to their different biological structures between the sexes.[25] The report from Guangdong, China, suggested that infected men can transmit HPV to women, and high-risk HPV infections may be less likely to persist in men than in women.[26]

Prevalence of low-risk and high-risk HPV types in age groups

Our study found that the detectable rates of low-risk HPV types peaked in the ages of 15–19 and 55–59 years, which can be explained by the fact that patients of these two age groups are either too young or too old to have strong immunity to HPV infection. Our study also found that the detectable rates of high-risk HPV types peaked in the age of 55–59 years. Consistent with our study, Peng *et al* also observed the rate of HPV infections peaked at the age of ≤20 years (60%) and 55–59 years (50.70%). Most patients of the former age group had low-risk HPV types, whereas most patients of the latter age group had high-risk HPV types.[27]

Multiple HPV type coinfections in genital warts

In this study, we found that multiple HPV type coinfections in genital warts were common. Brown *et al* also found multiple HPV type coinfections in genital warts.[28] A study from the USA showed that 19.0% of HPV-positive samples were positive for two or more HPV types in women.[29] Data from Italy showed that multiple HPV type coinfections occurred by chance and that no particular types of HPV were more likely to occur in coinfections than others.[30] In our study, there was no correlation between number of HPV types and age, suggesting that HPV types do not accumulate with age.

CONCLUSIONS

The present study characterised the prevalence and distribution of HPV types in genital warts in Xi'an, China. The results suggest that low-risk HPV types are major pathogens of genital warts, but high-risk HPV type infections and multiple HPV type coinfections are also common in genital warts. HPV6, 11, 52 and 16 are the four most common HPV types in genital wart in Xi'an, China, and nonavalent vaccine against HPV6, 11, 16, 18, 31, 33, 45, 52 and 58, covering the four most common HPV types, may be more suitable for Xi'an, China.

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Contributors CZ and YW designed this study and analyzed data. WM collected specimens and detected HPV types. HZ and JM performed clinical diagnosis of genital warts. CZ drafted the paper. All authors approved the final version of the article.

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Competing interests None declared.

Patient consent for publication Obtained.

Ethics approval The Ethics Committee of SPISSC approved the collection of specimens of genital warts and testing of the HPV types in the specimens (approval number: SXEDC2017-001).

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