Observational Study

The prevalence and genotype distribution of human papillomaviruses among women in Taizhou, China

Rongrong Jin, MD a, Hua Qian, MD b, Yongsheng Zhang, MD c, Donglan Yuan, MD b, Jingjing Bao, MD a, Huilin Zhou, MD a, Min Chen, MD d, Junxing Huang, MD d, Hong Yu, MD a,∗

Abstract

Human papillomavirus (HPV) infection is the leading cause of cervical cancer and precancerous lesions. Knowledge regarding the prevalence and genotype distribution of HPV in women is important to establish strategies for cervical cancer screening and HPV vaccination. This study aimed to evaluate the characteristics of HPV infection in Taizhou, China. HPV genotype of 10,733 women who visited Taizhou People’s Hospital from November 2016 to October 2018 was determined using a PCR and hybridization-based detection test. The prevalence of overall, high risk (HR), and low risk (LR) HPV infections was 34.58%, 29.92%, and 10.12%, respectively. Of HPV-positive cases, 2417 (65.13%) were infected with a single HPV genotype and 1294 (34.87%) were infected with multiple HPV genotypes. HPV-52 was the most prevalent genotype (6.21%), followed by HPV-16 (5.33%), HPV-53 (4.03%), HPV-58 (3.89%), and HPV-81 (3.75%). The highest prevalence of HPV infection was found in women aged ≥60 years (40.72%). Furthermore, the prevalence of HPV increased with the severity of cervical lesions. In conclusions, the prevalence and genotype distribution of HPV varied with age and cervical lesions. The findings might serve as a potential reference for guiding cervical cancer screening and vaccine-based HPV prevention in Taizhou.

Abbreviations: AGC = atypical glandular cells, ASC-H = atypical squamous cells-cannot exclude high-grade squamous intraepithelial neoplasia, ASCUS = atypical squamous cells of undetermined significance, CIN = cervical intraepithelial neoplasia, HPV = human papillomavirus, HR = high risk, HSIL = high-grade squamous intraepithelial neoplasia, IARC = International Agency for Research on Cancer, LR = low risk, LSIL = low-grade squamous intraepithelial neoplasia, NILM = negative for intraepithelial lesion or malignancy, SCC = squamous cell carcinoma.

Keywords: age, cervical cancer, distribution, human papillomavirus, prevalence

1. Introduction

Cervical cancer ranks fourth in both incidence and mortality in cancers diagnosed in women worldwide, accounting for 6.6% of all female cancer cases and 7.5% of all female cancer death.[1] Although great improvement in the health and sanitary conditions, both incidence and mortality rates of cervical cancer are still increasing in China.[2,3] It is deemed necessary to further strengthen cervical cancer screening program.

Human papillomavirus (HPV) infection is the cause of the majority of cervical neoplasia, as well as some other anogenital and oropharyngeal cancers,[4] accounting for approximately 5% of all cancers worldwide.[5] To date, >200 HPV subtypes have been identified, which are classified into high risk (HR) and low risk (LR) types on the basis of their correlation with malignant tumors, especially cervical cancer.[6–10] International Agency for Research on Cancer (IARC) has clearly classified HPV-16, HPV-18, HPV-31, HPV-33, HPV-35, HPV-39, HPV-45, HPV-51, HPV-52, HPV-56, HPV-58, and HPV-59 as HR-HPV which account for 95% of all cervical cancer.[11,12] On the other hand, LR-HPV subtypes such as 6, 11, can cause visible genital warts and early or low cervical epithelial cell changes. HPV vaccine has become a major progress in preventing cervical disease and cancer. The current vaccines were developed based on epidemiological data from Europe and the United States, especially for HPV-16 and -18, whereas the prevalence and genotype distribution of HR-HPV vary by race and country, and even the most common HPV genotypes may vary by the region in the same country.[13–15] As the 3 HPV vaccines have been approved by the Chinese government, it is necessary to understand the HPV genotype distribution in different regions of China to help improve cervical screening and vaccination. In the present study, we aimed to investigate the prevalence and
genotype distribution of HPV among women in Taizhou, Jiangsu Province, China.

2. Materials and methods

2.1. Subjects

This retrospective study was designed to analyze the results of cervical HPV test of 10,733 women who visited Taizhou People’s Hospital from November 2016 to October 2018. The patients were aged from 16 to 85 years and the mean age was 40 years. Patients who had history of cervical intraepithelial neoplasia (CIN), cervical cancer, and hysterectomy were excluded. No gynecological examination, vaginal drug administration, and sex life were performed within 3 days before the examination. Of 10,733 women, 4865 cases simultaneously or subsequently underwent cervical cytology. Cytological specimens were blinded and evaluated by experienced pathologists according to the Bethesda System. The samples were classified as: negative for intraepithelial lesion or malignancy (NILM), atypical squamous cells of undetermined significance (ASCUS), atypical squamous cells-cannot exclude high-grade squamous intraepithelial neoplasia (ASC-H), low-grade squamous intraepithelial neoplasia (LSIL), high-grade squamous intraepithelial neoplasia (HSIL), atypical glandular cells (AGC), or squamous cell carcinoma (SCC). Furthermore, 343 patients were diagnosed as cervical intraepithelial neoplasia or cervical cancer by cervical biopsy or conization. This study was approved by the Ethics Committee of Taizhou people’s Hospital.

2.2. HPV genotyping

HPV isolates were genotyped using the PCR-RDB HPV genotyping for 23 types kit (Yaneng Biotech, Shenzhen, China), which can identify 17 HR-HPV types (16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 68, 73, and 82) and 6 LR-HPV types (6, 11, 42, 43, 81, and 83). HPV DNA was extracted, amplified, and genotyped according to the manufacturers’ protocol. Furthermore, sterile water and specimens with known HPV genotypes were used as the negative and positive controls, respectively.

2.3. Statistical analyses

All statistical analyses were performed by using SPSS V22.0 (SPSS, Chicago, IL). Differences between groups were examined using the chi-square test or Fisher exact probability test. A P value of <.05 was considered statistically significant.

3. Results

3.1. General characteristics

A total of 10,733 women who received medical treatment and physical examination in Taizhou People’s Hospital from November 2016 to October 2018 were retrospectively analyzed. The characteristics of the study population were summarized in Table 1. Among 4865 women who simultaneously or subsequently underwent cervical cytology, 4294 had normal cytology, 407 had ASCUS, 75 had LSIL, and 85 had ASC-H/HSIL. Furthermore, 343 women were diagnosed with CIN or cervical cancer, 180 with CIN1, 145 with CIN2/3, and 18 with SCC.

### Table 1

| Characteristics                  | Number | %   |
|----------------------------------|--------|-----|
| Total                            | 10,733 |     |
| Age (y, range)                   | 40.00 (16–85) | 45.33 |
| Cervical cytology                | 4,865  | 88.26 |
| ASCUS                            | 4,294  | 88.26 |
| LSIL                             | 75     | 1.54 |
| ASC-H/HSIL                       | 85     | 1.75 |
| AGC                              | 4      | 0.08 |
| Cervical histological types       | 343    | 2.02 |
| CIN1                             | 180    | 52.48 |
| CIN2/3                           | 145    | 42.27 |
| SCC                              | 18     | 5.25 |

ASCUS = atypical squamous cells of undetermined significance. CIN = cervical intraepithelial neoplasia, LSIL = low-grade squamous intraepithelial neoplasia, NILM = negative for intraepithelial lesion or malignancy, SCC = squamous cell carcinoma.

3.2. Prevalence of HPV types

Of 10,733 subjects, 3711 were positive for HPV infection, with the infection rate of 34.58%. The prevalence of HR-HPV and LR-HPV infections was 29.92% (3211/10,733) and 10.12% (1086/10,733), respectively. HPV-52 was the most prevalent genotype (6.21%), followed by HPV-16 (5.33%), HPV-53 (4.03%), HPV-58 (3.89%), and HPV-81 (3.75%) (Fig. 1). The most common HR-HPV types after HPV-52, HPV-16, HPV-53, and HPV-58 were HPV-51 (3.25%), HPV-68 (2.59%), HPV-59 (2.47%), and HPV-56 (2.33%). HPV-18 was found in 2.09% of women (n=224), ranking the 15th most common HPV genotype. The most common LR-HPV types after HPV-81 were HPV-42 (2.17%), and HPV-43 (2.16%).

The infection rates of single and multiple HPV genotypes were 22.52% (2417/10,733) and 12.06% (1294/10,733), respectively. The prevalence of single HPV genotype in both LR-HPV and HR-HPV groups were higher than that of multiple HPV genotypes. For the single HPV infection, the most common genotype of LR-HPV was HPV-52 (3.11%), followed by HPV-16 (2.68%), HPV-58 (1.82%), HPV-53 (1.55%), and HPV-51 (1.46%). For multiple HPV infection, HPV-52 (3.09%) and HPV-16 (2.65%) were also the first and second leading common genotypes, respectively, followed by HPV-53 (2.49%), HPV-58 (2.07%), and HPV-51 (1.79%). As for the LR-HPV genotypes, HPV-81 was the most common genotypes, followed by HPV-42 or HPV-43 in both single and multiple HPV infections. HPV-83, HPV-82, and HPV-73 were consistently the third least frequent HPV genotypes in single and multiple HPV-infected subjects.

3.3. Age-specific HPV prevalence

All women were divided into 9 age groups: ≤24 years old, 25 to 29 years old, 30 to 34 years old, 35 to 39 years old, 40 to 44 years old, 45 to 49 years old, 50 to 54 years old, 55 to 59 years old, and ≥60 years old. As shown in Table 2, the HPV infection prevalence in women ≤24 years old, 25 to 29 years old, 30 to 34 years old, 35 to 39 years old, 40 to 44 years old, 45 to 49 years old, 50 to 54 years old, 55 to 59 years old, and ≥60 years old was 36.19%, 12.07%, 1.53%, 10.21%, 9.31%, 9.78%, 15.50%, 46.39%, and 58.77%, respectively. A different age specific prevalence was observed in both overall and HR-HPV infection (P for trend <.001). The distribution of both overall and HR-HPV infection types were compared using chi-square test or Fisher exact probability test. A P value of <.05 was considered statistically significant.
infection showed a bimodal pattern in different age groups. The fourth highest infection rate was observed in women aged 20–24 years old, which declined to its lowest point in women aged 25 to 29 years old and increased gradually and reached its highest point in women aged ≥60 years old. For LR-HPV infection, there was a weak but significant increase trend with age (P for trend = .030).

In addition, HPV-16 and HPV-52 were consistently the 2 most common HPV genotypes, whereas HPV 83 was the least common HPV genotype in all 9 age groups. HPV-56 prevalence was increased with age (P for trend < .001), which was the fifth and fourth most common genotypes in women aged 55 to 59 years old (4.64%) and ≥60 years old (6.82%), respectively.

3.4. HPV prevalence according to cervical cytology

Among 4865 women with concurrent cytological results, the prevalence of overall HPV, HR-HPV, and LR-HPV were 24.73%, 20.05%, and 7.82% in women with NILM, respectively.

**Table 2**

The prevalence and genotype distribution and of HPV according to age.

| HPV Genotype | Age groups, y | Prevalence (%) |
|--------------|---------------|----------------|
| Any HPV      | 25–29 (%)     | 477 (31.42)    |
|             | 30–34 (%)     | 472 (31.35)    |
|             | 35–39 (%)     | 472 (31.35)    |
|             | 40–44 (%)     | 472 (31.35)    |
|             | 45–49 (%)     | 472 (31.35)    |
|             | 50–54 (%)     | 472 (31.35)    |
|             | 55–59 (%)     | 472 (31.35)    |
|             | ≥60 (%)       | 472 (31.35)    |

**Figure 1.** The prevalence and genotype distribution of HPV among 10,733 women. HPV = human papillomavirus.

HPV = human papillomavirus.
vitively, which were significantly lower than those in women with ASCUS, LSIL, and ASC-H/HSIL (P < .001, Table 3). Furthermore, the prevalence of overall HPV and HR-HPV increased with the severity of cervical cytology (P < .001). Women with LSIL had the highest prevalence of overall HPV (96.00%) and LR-HPV (37.33%), whereas those with ASC-H/HSIL had the highest prevalence of HR-HPV (85.33%). HPV-52 was the most common genotype in women with NILM (3.96%) and ASCUS (16.22%) but ranked the second and third most common genotype in those with LSIL (18.67%) and ASC-H/HSIL (17.65%), respectively. HPV-53 and HPV-16 were the most common genotype in women with NILM (3.96%) and ASCUS (16.22%) but ranked the second and third most common genotype in those with LSIL (18.67%) and ASC-H/HSIL (17.65%), respectively. HPV-53 and HPV-16 were the most common genotype in women with LSIL (29.33%) and ASC-H/HSIL (45.53%), respectively. As for 4 AGC cases, only 1 was infected with HPV.

3.5. HPV prevalence according to cervical lesions

Among the 343 women with cervical intraepithelial neoplasia or SCC, the prevalence of overall HPV, HR-HPV, and LR-HPV infections was 85.56%, 83.33%, and 22.78% in CIN1 cases, 93.10%, 92.41%, and 14.48 in CIN2+ cases, and 94.44%, 94.44%, and 22.22% in SCC cases, respectively. The prevalence of overall HPV and HR-HPV increased with the severity of cervical lesions (P for trend = .028 and .011, respectively). The top 5 most prevalent HPV types in CIN1 and CIN2+ cases were HR-HPV, but LR-HPV-81 was the second common type in SCC cases (Fig. 2). The prevalence of HPV-16 increased with the severity of cervical lesions (P for trend < .001) and reached its highest point in SCC with the infection rate of 61.11%.

4. Discussion

The persistent infection of HR-HPV is the key and necessary pathogenic factor for cervical cancer.\(^9,10\) Therefore, HPV testing reduces the incidence and mortality of cervical cancer.\(^16\) However, it is difficult to establish a comprehensive database of the prevalence and genotype distribution of HPV in China due to the large geographical area and the largest population in the world, which is important to determine the best prevention approach for cervical cancer and assess the effect of HPV vaccines. There is still lack of comprehensive data on the prevalence and genotype distribution of HPV in Taizhou. In the present study, we investigated the prevalence and genotype distribution of HPV in women who sought a medical examination and treatment. The overall prevalence of HPV (34.58%) in our study was higher than that in other regions in China, such as Shandong (28.4%),\(^17\) Zhejiang (22.8%),\(^18\) Shanghai (31.84%),\(^19\) Nanhai area of Foshan (13.5%),\(^20\) Jiangxi (22.49%),\(^21\) and Jiangsu (15.5%).\(^22\) The prevalence of HR-HPV (29.92%) was also higher than that in most regions in China but lower than that in Haikou (31.94%) and Shanghai (34.68%).\(^19\) The findings reveal that Taizhou has a higher prevalence of HPV infection than other regions in China. High infection rate of HR-HPV in our study implies the inadequacy of routine screening for cervical cancer in Taizhou.

Table 3

| HPV     | NILM (%) | ASCUS (%) | LSIL (%) | ASC-H/HSIL (%) |
|---------|----------|-----------|----------|----------------|
| (n = 4294) | (n = 407) | (n = 75)  | (n = 85)  |
| Any HPV | 1062 (24.73) | 299 (73.46) | 72 (96.00) | 79 (92.94) |
| HR-HPV  | 861 (20.05)  | 280 (68.80) | 64 (85.33) | 79 (92.94) |
| 16      | 108 (2.52)   | 59 (14.50)  | 9 (12.00)  | 37 (43.53) |
| 18      | 56 (1.30)    | 28 (6.88)   | 5 (6.67)   | 3 (3.53)   |
| 31      | 44 (1.02)    | 10 (2.56)   | 6 (8.00)   | 9 (10.59)  |
| 33      | 43 (1.00)    | 27 (6.63)   | 2 (2.67)   | 6 (7.06)   |
| 35      | 37 (0.86)    | 8 (1.97)    | 2 (2.67)   | 5 (5.88)   |
| 39      | 26 (0.61)    | 11 (2.70)   | 4 (5.33)   | 2 (2.35)   |
| 45      | 9 (0.21)     | 3 (0.74)    | 0 (0.00)   | 0 (0.00)   |
| 51      | 121 (2.82)   | 34 (8.35)   | 9 (12.00)  | 2 (2.35)   |
| 52      | 170 (3.96)   | 66 (16.22)  | 14 (18.67) | 15 (17.05) |
| 53      | 106 (2.47)   | 36 (8.85)   | 22 (29.33) | 2 (2.35)   |
| 56      | 58 (1.35)    | 23 (5.65)   | 11 (14.67) | 2 (2.35)   |
| 58      | 94 (2.19)    | 45 (11.06)  | 5 (6.67)   | 18 (21.18) |
| 59      | 83 (1.93)    | 20 (4.91)   | 1 (1.33)   | 1 (1.18)   |
| 66      | 54 (1.26)    | 18 (4.42)   | 10 (13.33) | 1 (1.18)   |
| 68      | 85 (1.98)    | 22 (5.41)   | 5 (6.67)   | 7 (8.24)   |
| 73      | 7 (0.16)     | 0 (0.00)    | 1 (1.33)   | 1 (1.18)   |
| 82      | 2 (0.05)     | 0 (0.00)    | 0 (0.00)   | 1 (1.18)   |
| LR-HPV  | 336 (7.82)   | 67 (16.46)  | 28 (37.33) | 11 (12.94) |
| 6       | 53 (1.23)    | 23 (5.65)   | 8 (10.67)  | 2 (2.35)   |
| 11      | 40 (0.93)    | 7 (1.72)    | 3 (4.00)   | 1 (1.18)   |
| 42      | 68 (1.56)    | 11 (2.70)   | 10 (13.33) | 1 (1.18)   |
| 43      | 74 (1.72)    | 12 (2.95)   | 3 (4.00)   | 3 (3.53)   |
| 81      | 121 (2.82)   | 21 (5.16)   | 7 (9.33)   | 4 (4.71)   |
| 83      | 6 (0.14)     | 1 (0.25)    | 3 (4.00)   | 0 (0.00)   |

ASCUS = atypical squamous cells of undetermined significance, HPV = human papillomavirus, LSIL = low-grade squamous intraepithelial neoplasia, NILM = intraepithelial lesion or malignancy.

Figure 2. HPV prevalence according to cervical lesions. HPV = human papillomavirus.
Previous studies have indicated that the 5 most common HPV types were HPV-16, HPV-18, HPV-31, HPV-33, and HPV-58 in Europe, HPV-16, HPV-53, HPV-52, HPV-18, and HPV-39 in northern America, HPV-16, HPV-52, HPV-18, HPV-58, and HPV-31 in Africa, and HPV-16, HPV-52, HPV-58, HPV-18, and HPV-56 in Asia.[23] HPV-16 and HPV-18 play a predominant role in above regions. In the present study, the 5 most common types of HPV in Taizhou were HPV-52, HPV-16, HPV-53, HPV-58, and HPV-51, whereas HPV-18 had a relatively low prevalence, which was in agreement with that in Shandong Province[11] but was slightly different from previous studies in other regions of China, such as Zhejiang Province.[18] Shanghai[19] and even Jiangsu Province.[22] The reason for the inconsistencies may be that hospital population-based surveys are opportunistic screenings with a selection bias and the levels of economic development in different regions of Jiangsu Province varies greatly. On the other hand, HPV-16 plays a predominant role in all regions while other subtypes vary in different regions in China. Taken together, HPV-52, HPV-16, HPV-53, and HPV-58 seem to be the most common HPV genotypes in most regions in China.

Age is an important factor associated with HPV infection. The prevalence and genotype distribution of HPV and HR-HPV in different age groups are varied. The highest prevalence of overall HPV, HR-HPV, and LR-HPV was observed in women aged ≥ 60 years old. Furthermore, overall HPV and HR-HPV infection showed higher rates in younger women but lower rates in middle-age women. Previous studies have revealed 2 peaks of age-specific prevalence of HPV, which varied in different studies.[17,20–22] However, some studies showed that HPV prevalence decreased with the increasing age in other regions worldwide.[24–25] The peak age of HPV infection may be affected by the immature immune system of young women and the physiological and immunologic disorders caused by hormone fluctuations of older women.

HPV infection, especially HR-HPV, is responsive for the majority of CIN lesions. Our findings revealed that the prevalence of HR-HPV positivity increased with the severity of cervical lesion, which was in agreement with those found in other regions or countries.[26,27] A worldwide meta-analysis indicated that HPV-16 and HPV-18 were the most prevalent types of HPV associated with cervical lesions worldwide, while HPV-52 and HPV-58 were more prevalent in East Asian women.[28] Previous studies have suggested that HPV-52 and HPV-58 are 2 important HPV types associated with cervical cancer in China.[29] Our study showed that HPV-16, HPV-52, HPV-58, and HPV-18 were 4 of the 5 most common HPV genotypes in CIN1 and CIN2a cases. Furthermore, HPV-53 was the fourth, fifth, and third common genotypes in CIN1, NILM, and SCC cases, respectively. The result is required to validated in larger sample studies due to small size of SCC. A recent study by Qian et al[30] showed that HPV-53 was one of the most common genotypes in different cervical diseases including cervical cancer. These findings indicate that HPV-53 may be the major risk factor for cervical cancer in China. HPV vaccines against HPV including HPV-53 should be developed for Chinese women specifically.

In summary, this study represents the most comprehensive assessment of the prevalence and genotype distribution of HPV in women in Taizhou. The findings reveal a high HPV prevalence and implied a high incidence of cervical cancer in Taizhou. HPV-52, HPV-16, HPV-53, HPV-58 were the 4 most common HPV genotypes, which was consistent with those in other regions of China but was obviously different from those found in developed countries. It is necessary to develop the next-generation HPV vaccines targeting a broader spectrum of HR-HPV types such as HPV-53 for Chinese women.

Author contributions: Rongrong Jin, Hong Yu.

Data curation: Rongrong Jin.

Formal analysis: Hua Qian, Yongsheng Zhang, Jingjing Bao, Junxing Huang.

Funding acquisition: Hua Qian, Hong Yu.

Investigation: Hong Yu.

Methodology: Rongrong Jin, Yongsheng Zhang, Donglan Yuan, Jingjing Bao, Huilin Zhou.

Resources: Rongrong Jin, Donglan Yuan, Min Chen.

Writing – original draft: Rongrong Jin, Hua Qian, Hong Yu.

Writing – review & editing: Hua Qian, Junxing Huang, Hong Yu.

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