Analysis of TCT Combined with HPV Typing Detection in Cervical Lesions Screening of Female Medical Staff in Obstetrics and Gynecology Hospital

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ABSTRACT

Objective: This study was to analyze the physical examination and screening data of female medical staff in a maternity hospital in Shanghai, and to study the relationship between liquid-based cytology and human papillomavirus (HPV) infection.

Methods: TCT results of 1093 female medical staff in the hospital from 2015 to 2017 and HPV results of 1085 cases in 2017 were collected. SurePath liquid-based cell preparation was used for cytology and the segr-21 typing kit was used for HPV detection. The relationship between the pathological results of cervical colposcopy biopsy and HPV infection was studied by single factor and multiple factor regression analysis.

Result: The rate of HPV infection women, who were aged 22-90 years old in this research was 10.0% (108/1085) and increased with age. High-risk HPV infection accounted for 95.37% (103/108), and the major high-risk HPV infection was HPV 52, 53, 58, and 16 in descending order. The 3-year cytological positive rates were 1.5%, 1.6% and 4.0%, respectively. Sixty-six of the HPV and/or cytological positive cases underwent colposcopy biopsy, and the proportion of cytological positive, HPV positive, HPV16 positive, cytological negative in 2015 and 2016 was 41.1%, 19.1%, 60% and 29.7%, respectively. Multiple logistic regression showed that cytology was an independent risk factor, OR 7.87 (95%CI: 1.85-33.54).

Conclusion: The rate of HPV infection in medical staff was lower than that in the general population, but the composition ratio of high-risk HPV infection was very high. It was recommended that women should be carried out HPV test regularly from sexual life, and early detection of high-risk HPV infection could prevent the occurrence of cervical cancer. The sensitivity and specificity of cervical cytology screening were higher than HPV screening in people with low infection rate, and the cytology screening cycle should not exceed one year.

Introduction

Cervical cancer is a malignant tumor that seriously harms women’s health. According to the World Health Organization, about 70,000 new cases of cervical cancer were found worldwide in 2018, and more than 310,000 cases of death because of cervical cancer. The incidence rate of middle-income and low-income Asian and African countries was 76.2% [1]. Human papillomavirus (HPV) infection is recognized as an important cause of cervical cancer. At least 13 types of HPV high-risk infections may cause cancer. Among which, HPV16 and HPV18 high-risk subtype infections has a possibility as high as 70% to cause cervical cancer or cervical precancerous lesions [2]. There are generally different stages before an infection of HPV could become cervical cancer. The pathological changes of the cervical epithelium can be found by Papanicolaou test. Study shows that the incidence of cervical cancer has a younger trend. Since the 1950s, European and American countries have used Papanicolaou test to perform cervical cancer screening and has greatly reduced the incidence and mortality of cervical cancer [3].

Therefore, there is significant importance and meaning for women to take HPV test regularly, it will help detecting high-risk
HPV infection and thus be treated timely. The cervical liquid-based cytology method developed in the 1990s has gradually replaced the traditional Pap smear in many countries, further improving the detection rate of cervical lesions. The rise of HPV testing has led to debates about whether HPV testing can replace cytology as a first-line screening method for cervical lesions. In the 2016 study “The American College of Obstetricians and Gynecologists. Cervical Cancer Screening and Prevention”, it is recommended that women aged 3–65 years should give priority to the combination of cytology and HPV screening [4]. In recent years, studies and guidelines have proposed single HPV testing is a viable screening method for cervical cancer [5]. There has always been a widespread attention and debate on the choice of the right method to detect cervical cancer and that for specific groups.

In China, a number of studies have focused on HPV infection rates [6,7] and Pap smear test positive rates [8]. Medical staff is a special occupational group as they have higher awareness of HPV infection than the general population and have more medical expertise. However, staff at obstetrics and gynecology hospitals are of high-risk group as they’re expected to be exposed to HPV infection. There’s relatively small infection rate data of this certain group of people and whether the incidence of HPV infection and cervical lesions is higher than ordinary women are still unclear. By improving and analyzing the screening data of the hospital’s obstetricians and gynecologists, the basic data will be accumulated for the primary prevention and screening methods for high-risk groups in the future. This study analyzed the cytology and HPV characteristics of this specific group of medical staff from Shanghai. We also analyzed the effectiveness of various methods to prevent precancerous lesions.

**Materials and Methods**

**Research Object**

Data was collected from the cervical cancer screening data of all active and retired female medical staff of Obstetrics and Gynecology Hospital of Fudan University from 2015 to 2017. The screening period is once a year, with an interval of approximately 12 months. Thin prep cytology test was done during 2015-16, and HPV screening was added in 2017. The total number of people undergoing physical examinations in 2017 has increased compared to the past as it includes new employees as well as employees from 2015 and 2016. The number of tested samples in three years was 660, 729, and 1049.

**Research Objects and Methods**

**HPV Test Method**

The 21 full-type HPV test kit from JIANGSU BIOPERFECTUS TECHNOLOGIES COMPANY, LTD was used for the test. Thirteen types of high-risk types HPV (16, -18, -31, -33, -35, -39, -45, -51, -52, -56, -58, -59 and -68), five potential high-risk types (26, -53, -66, -73 and -82), and three low-risk types (-6, -11, -81) were simultaneously detected by type-specific primers. The classification of the three HPVs is based on IARC’s carcinogenic potential of HPV, that is, the high-risk type is carcinogenic or highly likely to be carcinogenic; the potentially high-risk type is likely to be carcinogenic; the low-risk type is non-carcinogenic [9]. Intracellular single copy gene DNAtopoisomerase 3 was used as a relative quantitative internal reference gene. Both PCR system and amplification assay is operated according to the product instructions. The result is based on the threshold set by the device.

**TCT (Thinprep Cytology Test) Method**

TCT was collected and maintained using a Sure Path cervical sampler and preservation solution (Becton, Dickinson and Company, Franklin Lakes, USA). After mixing, 2 ml of the sample was taken for HPV detection, and the remaining samples were used for TCT test. The cell preparation was conducted with BD PrepStain automatic processor and with the dyeing machine from Becton, Dickinson and Company. The procedure was carried out according to the protocol provided by the company. The results for the TCT test is either atypical squamous cell of uncertain significance, ASC-US or positive, including ASC-US, low grade squamous intraepithelial lesion, LSIL and high grade squamous intraepithelial lesion, HSIL.

**Statistical Analysis**

Data analysis was conducted by using Excel 2007 and SPSS 20.0 (…software). The prescriptive analytics results were presented by mean, standard deviation, median and percentile. The HPV infection rate of different age groups was compared using Chi-Squared Test, the amount and percentage were presented as composition ratio (%). $\chi^2$ test was used for univariate analysis and logistic regression was used for multivariable analysis with a test level of $\alpha=0.05$.

**Result**

**Overall Summary**

The number of medical staff who participated in the TCT test in 2015 and 2016 was 670 and 741 respectively, while in 2017, the number of samples for TCT and HPV test were 1093 and 1085, 8 of them only participated in the TCT test. The age distribution ranged from 22 to 90 years, with an average age of 41.6 years (41.6 years ± 14.0 years) and a median age of 37 years, with upper and lower quantile of 30 and 49. According to 4.0% respectively, and the ASC-US rate were 0.9%, 0.9% and 3.1% (Table 1). The positive and ASC-US rate in 2017 were significantly higher than that in 2015 and 2016 Pearson $\chi^2=16.03, P<0.01$. Colposcopy and histological biopsy were performed to 66 patients with positive TCT tests (ASC-US and above) in all three years. The results showed that 51 patients (77.3%) were normal (negative for intraepithelial lesion or malignancy, NILM), 13 (19.7%) were LSIL and 2 were HSIL.
### Table 1: 2015-2017 TCT Test Results of Female Medical Staff in a First-class Hospital at Grade 3 in Shanghai.

| NILM (%) | 2015      | 2016      | 2017      | Pearson $\chi^2$ | P     |
|----------|-----------|-----------|-----------|-------------------|-------|
|          | 660(98.5) | 729(98.4) | 1049(96.0)| 14.62             | 0.001 |
| ASC-US and above(%) | 10(1.5)  | 12(1.6)   | 44(4.0)   | 14.62             | 0.001 |
| ASC-US (%) | 6(0.9)   | 7(0.9)    | 34(3.1)   | 16.03             | <0.01 |
| LSIL (%) | 3(0.4)    | 3(0.4)    | 8(0.7)    | 0.59*             |       |
| HSIL (%) | 1(0.1)    | 2(0.3)    | 2(0.2)    | 1*                |       |
| Total (%) | 670(100)  | 741(100)  | 1093(100) |                   |       |

Note:* Fisher exact probability method:
ASC-US: Atypical Squamous Cell of Uncertain Significance; LSIL: Low Grade Squamous Intraepithelial Lesion; HSIL: High Grade Squamous Intraepithelial Lesion.

### HPV Infection Status of Personnel in 2017

#### Relationship Between Age and HPV Infection and TCT Test Result

The HPV infection rates of the age groups of 21 to 30 years, 31 to 50 years, and over 51 years were 9.6%, 8.6%, and 13.3%, respectively. The infection rate and the cytological positive rate increased as the age increases. (Table 2, Pearson $\chi^2 = 4.43, P = 0.11$, Table 1, Pearson $\chi^2 = 4.34, P = 0.11$).

### Table 2: 2017 HPV Test Result by Age Group.

| Age Group | HPV Results | TCT Result |
|-----------|-------------|------------|
|           | Negative (%)| Positive (%)| NILM (%) | ASC-US and above (%) |
| 21~30     | 246 (90.4)  | 26 (9.6)    | 266 (97.1)| 8 (2.9) |
| 31~50     | 510 (91.4)  | 48 (8.6)    | 541 (96.4)| 20 (3.6) |
| 51~       | 221 (86.7)  | 34 (13.3)   | 242 (93.8)| 16 (6.2) |
| Total     | 977 (90.0)  | 108 (10.0)  | 1049 (96.0)| 44 (4.0) |

Note: a $\chi^2$ test, Pearson $\chi^2 = 4.43, P = 0.11$

b $\chi^2$ test, Pearson $\chi^2 = 4.34, P = 0.11$

#### Different Subtypes of HPV Infection

Among the 108 HPV-positive medical staff, 82 cases (75.93%) were single infection, and 26 cases were mixed infection (24.07%). The HPV subtypes were divided into high-risk type, potential high-risk type and low-risk type. The common subtypes of high-risk HPV were HPV52, HPV58, HPV16, etc, the results showed that the number of infected people of high-risk HPV was 103 (73.04%). The common potential high-risk HPV subtypes include HPV53, etc, it has a number of infected people of 26 (18.44%). The common subtypes of low-risk HPV includes HPV81, and the results showed that the number of infected people was 12 (8.51%) (see Table 3).

### Table 3: 2017 HPV Infection Status of Screening Personnel in a First-class Hospital at Grade 3 in Shanghai.

| HPV Subtype          | # of People | Infection Rate (%) |
|----------------------|-------------|--------------------|
| High risk subtype    |             |                    |
| HPV52                | 31          | 21.99              |
| HPV58                | 15          | 10.64              |
| HPV16                | 13          | 9.22               |
| others (HPV56/59/39/68/18/33/31/51/55/45) | 44(8/8/7/7/4/2/1/1) | 31.21 |
| Potential high-risk subtype | | |
| HPV53                | 19          | 13.48              |
| HPV66                | 2           | 1.42               |
| HPV73                | 2           | 1.42               |
| HPV82                | 2           | 1.42               |
| HPV26                | 1           | 0.71               |
| Low risk subtype     |             |                    |
| HPV81                | 7           | 4.96               |
| HPV6                 | 3           | 2.13               |
| HPV11                | 2           | 1.42               |
Relationship Between Different Screenings Strategies and Cervical Biopsy

Using the positive results of the 2017 biopsy (LSIL or HSIL lesions) as criteria, the TCT and HPV test result from 2015 to 2017 were analyzed. 3 cases of negative result were missed diagnosed (8.1%) in the subjects when cytology was used as single screening strategy and all of them were LSIL lesions. While using HPV as single screening strategy, 9 patients were diagnosed as positive (19.1%), including 7 with LSIL and 2 patients with HSIL. 6 patients with HPV-negative result was missed diagnosed (31.6%), all of which were LSIL lesions. HPV16, the most relevant subtype with cervical cancer, has a high positive biopsy coincidence rate of 60% (3/5). Due to the small number of biopsies, the relevance of screening is not statistically significant. The second test with higher positive rate of biopsy was single cytology test. The biopsy rate of cytology was 41.4%, with 10 cases of LSIL and 2 cases of HSIL. The positive rate for both TCT and HPV test was 35% (7/20), while the positive biopsy rate for at least one of the two tests was 25% (14/56). The missed diagnosis rate was 29.7% (11/37) for those patients with negative CTC test result in 2015 and 2016 (Table 4).

Table 4: Histological biopsy compliance rate for different screening strategies.

| Screening Strategy | NILM(%) | LSIL(%) | HSIL(%) | Total |
|--------------------|---------|---------|---------|-------|
| Single TCT negative | 34 (91.9) | 3 (8.1) | 0 (0.0) | 37    |
| Single TCT positive | 17 (58.6) | 10 (34.5) | 2 (6.9) | 29    |
| HPV negative | 13 (68.4) | 6 (31.6) | 0 (0.0) | 19    |
| HPV positive | 38 (80.9) | 7 (14.9) | 2 (4.3) | 47    |
| HPV16 positive | 2 (40.0) | 2 (40.0) | 1 (20.0) | 5     |
| TCT and HPV positive | 13 (65.0) | 5 (25.0) | 2 (10.0) | 20    |
| TCT or HPV positive | 42 (75.0) | 12 (21.4) | 2 (3.6) | 56    |
| Previous TCT negative | 26 (70.3) | 9 (24.3) | 2 (5.4) | 37    |

Risk Analysis of Positive Test Results

Dividing the histological biopsy results into 2 groups, negative and positive (including LSIL and HSIL), we then use them as dependent variables of the logistic regression model along with ASC-US negative samples. After correcting age-related factors, the analysis was designed to study the relationship between dependent variables with TCT results (negative and positive), HPV results (negative and positive), HPV type 16 results (negative and positive), and the TCT results (negative and positive) for the first 2 years. The TCT results and HPV16 infection results were correlated with histological results, with an odds ratio (OR) of 7.99 (95% CI: 1.97-32.36) (P=0.004) and of 10.3 (95% CI: 1.24-85.15) (P = 0.031), respectively (Table 5). The Odds ratio becomes 7.87% (95% CI:1.85-33.54) (P=0.005) for the TCT result after adding cytological and HPV 16 results as the two independent variables while the correlation between HPV 16 infection and biopsy results disappeared (P=0.06) (Tables 5 & 6).

Table 5: Single Logistic Regression Analysis of Biopsy Results of Screening Personnel in a First-class Hospital at Grade 3 in Shanghai by Risk Factors.

| Factors | β | x² | P Ratio | Ratio 95% Confidence Interval |
|---------|---|----|---------|-----------------------------|
| TCT positive | 2.078 | 8.468 | 0.004 | 7.985 (1.97-32.36) |
| HPV positive | -0.652 | 1.1 | 0.294 | 0.521 (0.154-1.761) |
| HPV16 positive | 2.328 | 4.647 | 0.031 | 10.256 (1.235-85.151) |
| TCT positive in previous 2 years | 0.125 | 0.009 | 0.923 | 1.134 (0.09-14.21) |

Table 6: Multiple Logistic Regression Analysis of Biopsy Results of Screening Personnel in a First-class Hospital at Grade 3 in Shanghai by Risk Factors.

| Variable | β | x² | P | Ratio 95% Confidence Interval |
|----------|---|----|---|-----------------------------|
| Age | 0.035 | 1.632 | 0.201 | 1.035 (0.982-1.092) |
| TCT result | 2.063 | 7.773 | 0.005 | 7.867 (1.845-33.54) |
| HPV16 result | 2.331 | 3.551 | 0.060 | 10.288 (0.911-116.219) |
| Constant | -6.365 | 11.175 | 0.001 | 0.002 |
Conclusion

The HPV infection rate of the respondents in this study was 10.0%, while there have been many studies about the correlation between HPV infection and cervical cancer and precancerous lesions. The results of those studies vary as the region and test subject changes. According to the HPV test result of a First-class Hospital at Grade 3 in Hunan, the HPV infection rate of female who visited STD clinic and Urology clinic was 45.33%. The peak age of infection was 26-35 years old, but the difference in infection rates among different age groups was not statistically significant [10].

The HPV infection rate from the medical examination of women in a First-class Hospital at Grade 3 in Beijing was 12.84%. According to the result, infection rate was the highest among women under 26 years old and decreases as age grows [11]. The HPV positive rate of women under 39 years old in Shenzhen was significantly higher than those of 40 to 49-year-old and of over 50 years old [12]. In Laos, the percentage of HPV infection for women in age of 30-54 years old is 47.7% [13]. The HPV infection rate of medical staff in our study was 10.84% and was lower than that of hospital specialist outpatients and general medical examination. The author believes that it may be related to the target population of this study. Medical staff is a special occupational group with a higher awareness of HPV infection than the general population and more medical expertise. They pay more attention to personal protection in daily life and have better disease detection as well as treatment. These can all be some of the reasons why the HPV infection rate is lower for themedical staff than the general population.

On the other hand, the HPV subtypes of infection in this study vary a lot. In this study, the most prevalent infection subtype is HPV52, 53, 58 and 16. While in the past, most researches are concentrated in types of HPV16, 52, 58 and other high-risk subtypes [14,15]. HPV16 is classified as a high-risk HPV and have been confirmed by studies that its persistent infection is significantly associated with advanced cervical cancer [16]. The correlation was found in our study. However, the correlation disappears in multiple logistic regression analysis. The possible reasons for that could be the limitation of number of samples or other factors.

The rise of HPV testing has led to debates about whether HPV testing can replace cytology as a first-line screening method for cervical infections. The results of this study suggest that the cytology test result is dependent on risk factors for cervical lesions, and its specificity and sensitivity of cytological detection is higher than that of HPV detection. The positive rate of biopsy positive for both cytology and HPV is not higher than that of cytology screening alone, thus this study supports cytology as a first-line screening tool. However, the effectiveness of cytology highly depends on qualified condition, and the cytological quality of our hospital is within the range of reference values published by the American Association of Pathologists (CAP) [17]. Since then, previously reported ASC-US rate is at the lower limit of the CAP range, the ASC-US rate in the 2017 results has increased significantly from the previous two years and is conducive to improving cytological sensitivity. There is a controversy about the reasonable screening cycle. According to this study, a 1-year screening cycle may be necessary if cytology is used as a single screening method, there is still a 29.7% or more of the risks of LSIL given a negative cytology result for the previous 2 years. HPV typing detection, especially HPV16 and 18, has obvious advantages over untyped HPV testing by improving the accuracy of positive prediction. This is also the reason for the immediate referral of colposcopy in HPV16 and/or HPV18 positive in the American Society of Colposcopy and Cervical Pathology (ASCCP) guidelines.

This study was carried out in a group of medical staff in a First-class Hospital at Grade 3 in Shanghai. It analyzed the surveyed population’s HPV infection rate and HPV infection subtypes and analyzed the positive prediction effects of HPV detection and cytology screening. Due to the small sample number, no correlation with histology was found except for HPV16 in this study. Therefore, in future research, we can expand the sample size of the surveyed population, conduct research on the correlation between highly pathogenic HPV type and histology, and even explore the highly pathogenic HPV type of the Chinese population for further independent detection. That will effectively increase the specificity of HPV as a screening for cervical lesions. This provides independent test a significant meaning especially while using as primary screening in remote areas with scarce cytological diagnostics by reducing medical costs and improving screening rate.

Conflict of Interest

The author declares that this article has no actual or potential conflict of interest.

References

1. (2018) World Health Organization / Cancer Fact:Cervical uteri Source Globocan 2018 [EB/OL].
2. Ebisch RM, Siebers AG, Bosgraaf RP, Leon Fag Massuger, Ruud Lm Bekkers, et al. (2016) Triage of high-risk HPV positive women in cervical cancerscreening [J]. Expert Rev Anticancer Ther16(10): 1073-1085.
3. Austin RM (2010) Exhortations to abandon the Pap test as a routine initial cervical screening test are still premature and carry significant risks[J]. Diagn Cytopathol38(11): 783-787.
4. (2016) The American College of Obstetricians and Gynecologists. Cervical Cancer Screening and Prevention[J]. Obstet Gynecol 127(1): e1-e20.
5. Shijie Hu, Yujing Yang, Xia Zhao (2018) Value of HPV test. liquid-based cytologic test (TCT) and combination of the two methods in screening cervical disease 34(5): 371-376.
6. Long W, Yang Z, Li X, Ming Chen, Jie Liu, et al. (2018) HPV-16, HPV-58, and HPV-53 are the most carcinogenic HPV genotypes in southwestern China and their viral loads are associated with severity of premalignant lesions in the cervix[J]. Virol J15(1): 94.
7. Li M, Du X, Lu M, Weiyi Zhang, Zhihui Sun, et al. (2018) Prevalence characteristics of single and multiple HPV infections in women with cervical cancer and precancerous lesions in Beijing, China[J]. J Med Virol.
8. Pan QJ, Hu SY, Guo HQ, Wen-Hua Zhang, Xun Zhang, et al. (2014) Liquid-based cytology and human papillomavirus testing: a pooled analysis using the data from 13 population-based cervical cancer screening studies from China[J]. Gynecol Oncol 133(2): 172-179.

9. Stoler M, Kim KR, Bergeron C, et al. (2014) Squamous cell tumours and precursors, in Kurman RJ, Carcangiu ML, Herrington CS, et al. WHO Classification of Tumours of Female Reproductive Organs, 4th Edn., IARC, Lyon: pp. 172-182.

10. Miao Liu, Jianrong Ye, Shishan Yuan (2018) Gene Types Analysis of human papillomavirus in out-patients and healthy cases 15(2): 39-41.

11. Limin Zhang, Fang Fang, Kelin Chen (2018) Detection and analysis on gene subtypes of cervical human papillomavirus infection in women with pregnancy and physical examination 22(2): 265-268.

12. Jinjin Xu, Yuxia Zhang (2017) Screening and prevention strategies of two cancers of suitable age women in Guangming New District of Shenzhen City from 2011 to 2015 32(10): 2168-2171.

13. Takamatsu R, Nabandith V, Pholsena V, Phouthisone Mouthisone, Katsu Nakasone, et al. (2017) Cervical cytology and human papillomavirus among asymptomatic healthy volunteers in Vientiane, Lao PDR [J]. BMC Cancer 17(1): 872.

14. Jiayan Li, Yuwu Luo, Chao Bi (2014) Analysis of 1786 cases of HPV genetic subtype 28(6): 595-597.

15. Zheng HS, Zheng DN, Li JM, et al. (2017) The survey of human papillomavirus infection and subtype in 2663 women [J]. Contemporary Medicine 7(21): 20-21.

16. KarbalaieNiya MH, Keyvani H, SafarnezhadTameshkel F, Mostafa Salehi-Vaziri, SedighehTeaghinezhad-S, et al. (2018) Human Papillomavirus Type 16 Integration Analysis by Real-time PCR Assay in Associated Cancers [J]. Transl Oncol 11(3): 593-598.

17. Tao X, Austin RM, Zhang H, et al. (2015) Pap Test Reporting Rates for Conventional Smear and Liquid-Based Cervical Cytology from the Largest Academic Women’s Hospital in China: Analysis of 1,248,785 Pap Test Reports [J]. Acta Cytol 59(6): 445-451.