

Research Article

Concordance of Human Papillomavirus Type 16 and 18 in Cervical and Oral Specimen of Cervical Cancer Patients

Perbandingan Tingkat Kesesuaian Hasil Identifikasi Human Papilomavirus Tipe 16 dan 18 antara Spesimen Serviks dan Oral pada Penderita Kanker Serviks

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Abstract

Objective: To determine the conformity of HPV type 16 and 18 in cervical and oral/buccal specimens from cervical cancer patients.

Methods: A cross-sectional study was conducted in March - September 2016 at several hospitals in Makassar. HPV 16 and 18 genotyping in cervical and oral fluid of 77 patients with cervical cancer performed with PCR method.

Results: The prevalence of HPV type 18 infection both in the cervical and the oral fluid was higher than HPV type 16 [9(47.4%) vs 5(26.3%)]. The agreement of HPV type 18 infection ($r=0.328$; $p=0.000$) in the cervical-oral sites was higher than HPV type 16 ($r=0.194$; $p=0.042$).

Conclusions: HPV type 16 and 18 could infect both cervix and oral cavity although type-specific concordance is low.

Keywords: cervix, human papillomavirus, oral cavity,

Abstrak

Tujuan: Mengetahui tingkat kesesuaian hasil pemeriksaan HPV tipe 16 dan 18 antara spesimen serviks dan oral/buccal pada penderita kanker serviks

Metode: Penelitian potong lintang ini dilakukan pada Maret - September 2016 pada beberapa rumah sakit di Makassar. Pemeriksaan HPV 16 dan 18 pada cairan serviks dan oral dari 77 orang penderita kanker serviks menggunakan teknik PCR.

Hasil: Prevalensi infeksi bersama pada serviks dan oral HPV tipe 18 lebih tinggi dibandingkan HPV tipe 16 [9(47,4%) vs 5(26,3%)]. Tingkat kesesuaian antara HPV tipe 18 ($r=0,328$; $p=0,000$) pada serviks dan oral lebih tinggi dibandingkan tipe 16 ($r=0,194$; $p=0,042$).

Kesimpulan: HPV tipe 16 dan 18 dapat menginfeksi serviks dan oral meskipun tingkat kesesuaian kedua tipe ini lemah.

Kata kunci : human papillomavirus, kavum oral, serviks

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INTRODUCTION

Persistent infection of HPV high-risk types in the cervix uteri is a major cause of invasive cervical cancer and its precursor.¹ In addition to cervical cancer, HPV is also known to cause 0.7% of other cancer types including anal, vaginal, vulvar, penile and oropharyngeal cancer in women and men.^{2,3}

HPV16 and HPV18 are the cause of 50% and 20% of cervical cancer cases globally.⁴ HPV type 16 is also more common in oral carcinoma than other types (types 18, 31 and 33).⁵ A study shows

there is a strong correlation between high-risk HPV infection with oral squamous cell carcinoma.⁶⁻⁸ Another study on the oral mucosa of women with cervical lesions shows the progression of CIN associated with HPV16 infection in buccal.⁹ The prevalence of oral HPV infection is five times higher in women with cervical HPV infection compared with those without infection with HPV types although the agreement rate concordance rate between cervical and oral specimen was low.¹⁰ Conformity of HPV types both in oral and cervical allow exploring the differences and relationships between them.

METHODS

A cross-sectional study was conducted on cervical fluid and oral/buccal fluid from 90 cervical cancer patients from March to September 2016 in Wahidin Sudirohusodo hospital and affiliated hospitals at Department of Obstetrics and Gynecology, Faculty of Medicine, Universitas Hasanuddin. Cervical cancer diagnosis and HPV genotyping with PCR technique performed at the Laboratory of Pathology, Department of Pathology Faculty of Medicine, Universitas Hasanuddin. Data were analysed using SPSS and differences were considered to be significant if $p < 0.05$.

RESULTS

We examined HPV types 16 and 18 at the buccal and cervical fluid from 90 cervical cancer patients but only 77 patients enrolled in the study because 13 samples from 13 patients had lysis. HPV types 16 and 18 found in cervical fluid and oral/buccal. This study showed 38 (49.4%) of cervical cancer patients aged 40-49 years, 61 (79.2%) multiparous and 35 (45.5%) using contraception (22.1% using contraceptive injections/DMPA) (table 1).

HPV low-risk and high-risk found in both cervical fluid and oral/buccal. The prevalence of high-risk HPV more prevalent in the cervix compared to oral/buccal. The cervical fluid

contains only 1 (1.3%) low-risk HPV (type 53) whereas high-risk HPV were found type 16 (34.2%), 18 (40.8%), 33 (3.9%), 45 (9.2%) and 51 (10.5%). In the oral/buccal there are two low-risk types of HPV, which are type 11 (3.4%) and 42 (10.3%) whereas high-risk HPV type consists of type 16 (31%), 18 (37.9%) and 45 (17.2%). This study also showed that the prevalence of cervical and oral together with HPV types 18 is higher than HPV 16 (47.4% vs 26.3%). The prevalence of HPV types 18 higher than in other high-risk types of HPV both cervical and oral/buccal (Table 2). Level of concordance between HPV type 18 $r = 0.328$; $p = 0.000$ in cervical and oral higher than type 16 ($r = 0.194$; $p = 0.042$) (table 3).

Table 1. Sample Characteristic

| Characteristic | n | % |
|----------------------|----|------|
| Age (years) | | |
| 20-29 | 3 | 3.9 |
| 30-39 | 15 | 19.5 |
| 40-49 | 38 | 49.4 |
| >50 | 21 | 27.3 |
| Parity | | |
| Nulliparous | 16 | 20.8 |
| Multiparous | 61 | 79.2 |
| Contraception | | |
| No | 42 | 54.5 |
| Pil | 11 | 14.3 |
| Injection | 17 | 22.1 |
| Condom | 6 | 7.8 |
| Tubectomi | 1 | 1.3 |

Table 2. HPV Type in Cervical and Oral/Buccal Specimen

| HPV | Cervical n(%) | Oral n(%) | Cervical and Oral n(%) |
|------------------|------------------|--------------|---------------------------|
| High-risk | | | |
| 16 | 26(34.2) | 9(31) | 5(26.3) |
| 18 | 31(40.8) | 11(37.9) | 9(47.4) |
| 33 | 3(3.9) | - | - |
| 45 | 7(9.2) | 5(17.2) | 5(26.3) |
| 51 | 8(10.5) | - | - |
| Low-risk | | | |
| 11 | - | 1(3.4) | - |
| 42 | - | 3(10.3) | - |
| 53 | 1(1.3) | - | - |

Table 3. Correlation of HPV Types 16 and 18 between Cervical and Oral

| Cervical and oral of HPV type | Kappa | P-value |
|-------------------------------|-------|---------|
| 16 | 0.194 | 0.042 |
| 18 | 0.328 | 0.000 |

DISCUSSION

In the present study, we found the prevalence of HPV type 18 in the cervix and oral was higher than that of type 16. Evidence from virologic, molecular, clinical and epidemiological studies has identified HPV as a primary etiologic agent in cervical cancer.^{11,12} HPV types 16 and 18 are the cause of 70% of all invasive cervical cancers compared to HPV types 45, 31 and 33.^{1,13,14} However, this results are different from Syrjanen who obtained HPV type 16 is the most common found in oral carcinomas compared to other types (18, 31 and 33).⁵ Epidemiological studies by Vet in Jakarta, Tasikmalaya, and Bali on screening 2686 women obtained HPV type 16 with the highest prevalence (11.4%) through Pap smear and PCR techniques.¹⁵ High-risk HPV types, 16, 18, and 45 were found both in cervical and buccal while two other types (33 and 51) were only found in the cervix with higher prevalence than buccal. The present study shows the similar prevalence of types 16 and 18. This prevalence difference may be due to population differences and study characteristics. We did not assess whether oral HPV infection causes oral carcinoma and the presence or absence of lesions in the oral cavity.

Age is a major factor in cervical cancer due to HPV infection. The prevalence of women in their 20's is infected with high and low-risk HPV between 20%-40%.¹⁶ Our study also shows that the highest HPV type 16 infection occurs at age 25-40 years compared to HPV type 18 for the same age group following the risk of infection decreases with age.^{17,18} The number of cervical cancer patients in this study increased from age 20 to 40 years and then decreased after the age of >50 years. Cervical cancer risk factors also increase due to the use of hormonal contraceptives. Use of oral contraceptives (progestogen-only pill) and injections (DMPA) for more than 5 years increase the risk of cervical cancer.¹⁹

Women infected with genital HPV had a higher frequency of oral HPV infection, according to a study by Giraldo et al. Cytological and Pap smear examination showed the presence of HPV in the oropharynx epithelium of women infected with genital HPV.²⁰ A study on the oral mucosa of women with cervical lesions indicates the progression of CIN is associated with HPV 16 infection in buccal.⁹ High prevalence of oral HPV

is reported in women with cervical and oral HPV infection.²¹⁻²⁴ Oral infection with HPV type 16 is a strong risk factor for oropharyngeal cancer. The prevalence of oral HPV is lower than that of the cervix² while another study shows the same prevalence.³ The present study did not examine the sexual behaviour of the patients. Several factors affecting cervical HPV infection against oral HPV infection include differences in the prevalence of HPV based on anatomical sites (cervix and oral), and the distribution of HPV types was not fully understood.²¹

We also found high-risk HPV infection (16 and 18) in oral lower than the cervix. The prevalence of genital HPV infection is much higher than that of oral HPV infection. This is may be due to the production of saliva that can dilute the HPV signal. Some studies suggested that saliva can be protective against HPV because of antimicrobial agents such as lysozyme, lactoferrin and immunoglobulin A against the viral infections.²⁴

The prevalence of HPV in normal oral mucosa varies depending on the different types of samples, mode of retrieval, virus detection methods, sensitivity levels, PCR primers used and PCR inhibitor.^{7,11,21} Oral sampling technique requires special attention as important in accurately detecting the presence of HPV such oral sampling technique should be able to obtain high-quality DNA. Methods of collecting samples in the oral cavity and oropharynx to obtain viral DNA virus usually performed with brushing and mouth rinse method or rinse.^{22,25,26} Garcia-Clocas et al. study that compared the method of mouth rinse and brushing to obtain the DNA genome concluded one mouthwash sample obtain more DNA than the two brushing samples, but both methods were adequate in isolating the DNA required for PCR examination. Mouthwash is the preferred method for knowing the epidemiology of oral HPV DNA. However, the possibility of obtaining a large positive sample may be used in combination of several concurrent oral sampling methods.²⁵ HPV detection methods in malignancy with PCR have better sensitivity and specificity compared to the standard methods (Southern blot technique, in situ hybridisation, immunohistochemistry).²⁷

The present study obtained low concordance rates infection of HPV types 16 and 18. Prevalence

of oral HPV infection in saliva samples by 24% in women with HPV cervical lesions.²⁸ Another study has shown a low risk of high-risk HPV infection in the oral cavity of women with cervical cancer.²⁹ Although co-infection with low-risk HPV types, cervical and oral HPV infections is not independent so that cervical HPV infection may be a risk factor for oral HPV infection.¹⁰ The prevalence of oral HPV and cervical infection in 221 women infected with HPV was 16.7%.²¹ Oral infection is more common in women with cervical HPV infection compared to women without cervical infection (25.5% vs 7.9%). A study by Canadas found the prevalence of infection with cervical and oral HPV was lower than Fakhry et al. study (12.7% vs 5%).³⁰ Further study has shown that the prevalence of oral HPV infection is 5-fold higher in women with cervical HPV infection than without infection (7.0% vs 1.4%) and among 3% of women infected with oral HPV or cervical studies. The suitability rate of 6.6% indicating there is a relationship between the two sites although the agreement rate was low.¹⁰

CONCLUSION

In conclusion, HPV types 16 and 18 can infect the cervix and oral although the agreement of these two types HPV is weak.

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