The Frequency of HPV 16 and 18 in Cervical Discharge by PCR in Women with Abnormal Pap Smear or Biopsy

Majid Sharbatdaran¹, Sepideh Siadati¹, Mahtab Zeinalzadeh², Shahriar Shafaei¹, Zahra Basirat², Amir Esmi¹

1. Department of Pathology, Babol University of Medical Sciences, Babol, Iran
2. Department of Gynecology and Obstetrics, Babol University of Medical Sciences, Babol, Iran

ABSTRACT

Background & Objective: Cervical cancer is the second most common cancer in the world among women. Human papilloma virus (HPV) plays a major role in its development. The aim of this study was to determine the frequency of HPV type 16 and 18 in cervical discharge by polymerase chain reaction (PCR) method in women with atypical biopsy or Pap smear.

Materials and Methods: This case-control study was performed on women in Yahyanejad Hospital, Babol University of Medical Sciences, Babol, Iran during 2008-2009. Sixty women with normal Pap smear (group 1) and 30 with atypical Pap smear or biopsy (group 2) were enrolled in the study and their cervical discharge was assessed for HPV type 16 and 18. Data were analyzed with SPSS, Chi-Square, Fishers Exact test and t-test and P<0.05 was considered significant.

Results: HPV type 16 was found in 10% women of group 2 but not seen in group 1. HPV 18 was not detected. All women had one partner and none of them had alcohol consumption.

Conclusion: In comparison with other studies, the frequency of HPV infection was lower in our study. We considered this is strongly related to our culture and religious beliefs.

Keywords: Cervix, HPV 16, HPV 18, PCR

Introduction

Cervical cancer is the second most common cancer in the world, after breast carcinoma, among women (1, 2). It represents 9.8 % of all women cancers (3). Today, by epidemiologic and molecular studies, the association between human papillomavirus (HPV) and cervical cancer is established (2, 4, 5). 95-100% of cervical cancers are caused by this double-stranded DNA tumor virus (1, 6, 7). 10.4% of women are positive for cervical HPV
DNA and the prevalence in underdeveloped countries is higher than the developed countries, 15.5% vs. 10.0%. It has the highest prevalence (16.9%) in women under the age of 25 yr old (3).

In United States the most prevalence of HPV (44.8%) is in women between 20 to 24 years (6, 8). There are several types of HPV, and among them HPV 16, HPV18, HPV31, HPV 58, HPV 52 are the most common types. HPV 16 and HPV18 are considered the most important (3, 4, 9, 10). About 34% of infected women are positive HPV 16 or HPV18, or both. HPV 16 and HPV18 have a prevalence of 2.5 % and 5.9 %, respectively. HPV is responsible for 5.2% of all cancers around the world in both men and women (3).

As noted, the vast majority of women with positive Pap smears are HPV DNA positive, and because of the limitation of pap- stained cytological smears, screening protocols that use HPV DNA testing, solely or in combination with Pap smear may be more effective (11-13). Among the different methods, polymerase chain reaction (PCR) is the most sensitive method to detect HPV infection (14).

The aim of this study was to determine the frequency of HPV types 16 and 18 in cervical discharge by PCR method in women with atypical biopsy or Pap smear.

Material and Methods

This case- control study was performed on 60 Pap smears (group 1) and 30 atypical cervical biopsies or Pap smears (group2) of women attending the Gynecologic Clinic in Yahyanejad Hospital, Babol, Iran from 2008 to 2009. This study was approved by the Ethics Committee of Babol University of Medical Sciences, Babol, Iran.

For both groups, the patients data including age, marriage status, cigarette smoking, ethanol consumption, educational status, number of children, parity, abortion, frequency of intercourse, number of partners, age of the first sexual activity and the pathologic report of Pap smear or biopsy collected in questionnaire. Cervicovaginal fluid from all women (groups 1 & 2) were obtained by swabs inserted into the cervix.

HPV DNA were extracted using the DNA extraction kit (Roch, Germany), then with termocycler (Teeneh, England), DNA amplification was done. With agarose gel electrophoresis and staining with ethedium bromide, DNA bands analyzed using UV ray.

Statistical analysis was performed using SPSS 11.5 statistical software, Chi-Square, Fisher’s exact test and t-test. A P-value of <0.05 was considered statistically significant.

Results

The mean age of groups one and two were 26.19±10.81 and 29.31±11.28 years, respectively. There was no significant difference. All women in both groups were married. No one used nicotine or alcohol.

Furthermore, there was no significant difference between the educational status and the number of children, parity, abortion and frequency of intercourse. In group 2, thirty women including: ASCUS (17 cases), LSIL (2 cases) and HSIL (11 cases). HPV type 16 was detected in 3 cases of group 2 (10%) (LSIL: 1 case, HSIL: 2 cases) with PCR but was not seen in group 1 (P =0.035). The frequency of HPV 16 in women with abnormal Pap smear or biopsy was 10%. HPV type 18 was not seen at all.

The first intercourse for HPV infected women was under 18 years. All women in both groups had one partner.

Discussion

In our study, no one from group one (normal Pap smear or biopsy) had HPV 16 infection. In contrast, Kajar et al. showed 15.4% of cervical discharge in 1000 women with normal Pap smear were HPV DNA positive. In this study, teenagers who had ample sexual activity with multiple
partners were at increased risk than others at same age (15). Hagenesee et al. studied 2597 pregnant women with mean age 23.4±5.1 years, mean number of partner’s 3.3±2.2, and mean age of first intercourse 16.7±2.24 years. Totally, 28% of women were seropositive for HPV-16 (16). In their study, the mean number of partners (3.3±6.6) was very different from “one partner” in all women (groups 1& 2) in our study. We considered this difference significant, because multiple sexual partners are one of the important risk factors for cervical cancer. Some studies showed in women with more than one sexual partner that the HPV infection was significantly higher (17, 18).

Furthermore Ho et al. showed ethanol consumption was related to higher incidence of HPV (13). In our study, no one used ethanol.

6.7% HPV 16 and zero percent of HPV18 infection has been reported in Southern Iranian women with cervical carcinoma (19). In our study, the frequency of HPV 16 in women with abnormal Pap smear or biopsy was 10%. No one had HPV 18. These results are lower than the similar studies from other countries (20-23).

Hinchliffe et al. showed HPV DNA in cervical discharge in 27 young women with normal Pap smear. After 8 months, 43% of them were HPV DNA negative. They concluded that most HPV infection in young women was transient (24). Sweden reported the reduction of HPV prevalence from 21% to 3.2% in 2 years (25).

In addition, Cox showed that because of the transient infection with HPV in young women, HPV testing had no role in primary cervical screening. But for unequivocal Pap smear results follow-up is being recommended (26).

The difference between «one partner» in our study and the mean number of more than 3 partners in other studies is very important. Ethanol consumption was not seen in our study. This is due to cultural differences and religious beliefs.

Conclusion

According to the number of cases in our study, we recommend further studies and more patients to be involved.

Acknowledgment

We would like to thank the Vice Chancellery for Research of Babol University of Medical Sciences for the financial support. The authors declare that there is no conflict of interests.

References


