Human papillomavirus frequency of women at low risk of developing cervical cancer: a preliminary study from a Turkish university hospital

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Summary

Purpose: To investigate the frequency of human papillomavirus (HPV) infection among low-risk women for cervical cancer in our region.

Methods: In one year period, 230 consecutive women at low risk of developing cervical cancer were enrolled to the study. HPV DNA testing was performed by Hybrid Capture-I System (HC-I) and groups were constituted by HPV-positive and HPV-negative women. A comparison of the groups according to age, obstetric history and age at the beginning of sexual intercourse was made. Statistical analysis was performed.

Results: The frequency rate of HPV infection was demonstrated to be 6.1% (n = 14) in our study (5.9% in women ≤ 45 years and 7.7% in women > 45 years). Age-dependent differences were not observed between groups. There was no significant difference between HPV-positive and negative women regarding obstetric characteristics and mean age at first intercourse.

Conclusion: This study provided significant information on the frequency of HPV infection of low-risk women in our region. When considered with studies performed in other countries, our study may give some help on the natural history of HPV infection and cervical squamous lesions.

Key words: HPV frequency; Low-risk women; Middle East Anatolia.

Introduction

In women, cervical cancer is the second most common cancer and cause of cancer death worldwide; 80% of cases occur in developing countries [1]. There is a well-established association between high-risk malignant subtypes of human papillomavirus (HPV) infection and the development of cervical neoplastic changes [2, 3]. Although cervical cytologic evaluation is the best available screening method for a preinvasive condition, some problems have arisen from false negative results. Normal cytologic results in subclinic HPV infection is one of the reasons for these false negative results [4]. Since the rate of subclinic HPV infection is very high (70%) and HPV-positive women are more likely to develop squamous lesions than negative ones, alternative tests are mandatory for the detection of HPV infection [4, 5]. The Hybrid Capture (HC) system for the detection of HPV DNA is one of the most popular tests recently.

In the medical practice, the HC system is used in the triage of suspicious cytologic results, such as atypical squamous cells of undetermined significance (ASCUS), or low-grade and high-grade squamous intraepithelial lesions (LSIL and HSIL) [6, 7, 8]. On the other hand, knowledge about the prevalence of HPV infection in the cervical cytology of normal women is more limited [9]. In this study the aim was to determine HPV frequency and to do a one-year survey of women at low risk of developing cervical cancer in our region by the HC-I system.

Materials and Methods

This study was carried out at Erciyes University Medical Faculty, Department of Obstetrics and Gynecology, between June 2000 and May 2001. All women appropriate for our inclusion criteria were enrolled in the study. All of the 230 subjects had normal cervical cytology (negative for squamous intraepithelial lesions according to the Bethesda system), only one sexual partner, no habit such as smoking and no previous sexually transmitted disease history. For HPV testing, cells from the transformation zone of the cervix and the endocervical canal were collected with the Digene cervical brush placed in the Digene specimen transport medium and stored at –20°C until assayed.

HPV DNA was determined by the Hybrid Capture-I HPV DNA assay (Digene, Silver Springs, MD.) according to the manufacturer's instructions [10]. In this study only the high oncogenic risk HPV probe, which includes probes for HPV types 16, 18, 31, 33, 35, 45, 51, 52 and 56, was used. Samples were classified as positive for HPV DNA if the relative light unit reading obtained from the luminometer was equal to or greater than the mean of the positive control values.

According to the results of the HC-I test, subjects were divided into two groups – HPV-positive and HPV-negative. All of the HPV-positive subjects underwent colposcopic examina-

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tion. In colposcopy, standard procedures were used that consisted of examining the cervix and one-third of the upper vagina at x 15 and x 23 magnification after application of 5% acetic acid solution. Every six months cytology and colposcopy were performed and at the end of the first year the HC-I test was renewed.

The statistical analysis was performed using the SPSS 11.0 package for Windows. The average age was compared between the two groups using the Student's t-test. The chi-square test was used to determine the frequency of HPV according to age groups. Comparison of the obstetric characteristics were made with the Mann Whitney U test. Statistical significance was considered a p value < 0.05.

Results

In this study, 230 low-risk women with respect to cervical neoplasms were enrolled. HPV infection was determined in 14 (6.1%) of them and these subjects were grouped as the HPV-positive group (Table 1). The remaining 216 women formed the HPV-negative group. The age range was 18 to 68 years (35.79 ± 8.49) in the study group. The mean age was 34.64 ± 12.81 years and 35.86 ± 8.17 years in the HPV-positive and negative groups, respectively and no significant difference was found ($p > 0.05$). The age-related frequency of disease was also analyzed by using the upper reproductive age limit (45 years) and no differences were determined ($p > 0.05$). There were no significant differences between groups regarding mean age at first intercourse or obstetric characteristics ($p > 0.05$).

During the one-year follow-up period no cytologic or colposcopic abnormality was detected in the HPV-positive group. Spontaneous regression of the HPV infection was determined in four of the 14 HPV-positive women at the end of the year.

Discussion

The major goal in cervical cancer screening is to decrease morbidity and mortality from cervical cancer. To this end the pap smear, a widespread conventional screening test, has been combined with other screening methods such as HPV testing or colposcopy to increase its utility. Although some combinations have been used in the triage of squamous lesions, the combination of cytology and HPV testing has proven to be a better screening tool than either alone, generally reaching a sensitivity of over 95% for important cervical diseases [7, 8, 11]. Another problem in this area is whether women with cytologically normal or at low risk of developing cervical cancer are investigated for HPV infection. Before resolving this dilemma, determination of the prevalence of HPV infection among low-risk women is mandatory.

Table 1. — Statistical comparison of patient characteristics between HPV-positive and HPV-negative groups.

	High-risk HPV (+) (n = 14)	High-risk HPV (–) (n = 216)	p value
Age (years)*	34.64 ± 12.81	35.86 ± 8.17	
≤ 45 years (n %)	2 (7.7)	24 (92.3)	> 0.05
> 45 years (n %)	12 (5.9)	192 (94.1)	
Gravidity¶	3 (0-6)	3 (0-10)	> 0.05
Parity¶	2 (0-5)	2 (0-9)	> 0.05
Abortion¶	0.5 (0-3)	1 (0-5)	> 0.05
Age at first intercourse (years)*	18.35 ± 2.9	19.46 ± 3.03	> 0.05

*mean± standard deviation (SD)
¶ median (min-max)

In our study, we tried to select low-risk patients for cervical neoplasia using strict criteria. Conseguently the number of the patients enrolled to the study was very limited. Since the study population consisted of women seen at a gynecologic outpatient department for a wide spectrum of gynecologic complaints, this study determined only the frequency of HPV infection in a low-risk group. When we compare our study results with some previous studies we did not determine any significant differences between age groups with respect to HPV infection [12, 13]. In a study by Bauer *et al.* [12], HPV prevalence was found in 17.7% of cytologically normal women and proportional decline with age in HPV prevalence in this study group was demonstrated. Maehama *et al.* [13] reported that the prevalence rates of HPV by age were 20.6% in the group of ≤ 29 years and 9.0-10.9% in the groups ≥ 30 years in the Okinawan population. This rate was found to be 6.1% (5.9% in the group ≤ 45 years and 7.7% in the group > 45 years) in our study ($p > 0.05$). The incidence rate of cervical cancer in Okinawa was reported to be 30.8 per 100,000 women while this rate was 1.17 in our region (Cancer Registry Report of Turkey 1993-1994). In our opinion, the differences in HPV-infection rates between studies could have arisen from these different incidences of cervical cancer by regions.

The hypothesis that HPV infection is the primary cause of cervical neoplasia is a largely acceptable idea [5]. In light of this hypothesis and the rates of HPV infection in cytologically normal women, alternative screening algorithms are mandatory in the evaluation of this population. In the results of a case-control study, Liaw *et al.* [5] found that, in comparison with initially HPV-negative women, women who tested positive for HPV DNA at enrollment were 3.8 times more likely to have LSIL subsequently diagnosed for the first time during a ten-year follow-up and 12.7 times more likely to develop HSIL. Since the follow-up period was very short in our study, trying to interpret subsequent neoplastic changes is impossible for now. However, we consider that increased knowledge about this issue will make the triage of women at low risk for cervical cancer more important.

With the development of reliable and sensitive techniques for HPV detection, epidemiological studies for this infection have become possible. However, we have no information on the prevalence or frequency rate of HPV infection in women at low risk of developing cervical cancer in Turkey. In this first study we wanted to demonstrate the frequency of HPV infection among this population in the eastern Central Anatolia Region.

In conclusion, this study, when considered with similar studies, will provide significant information for the evaluation of cytologically normal women with regard to risk of cervical neoplasia. Our study is a preliminary one and we are planning to increase the number of low-risk women. We think that it will be useful to compare the long-term follow-up results with high-risk women. Thus the triage of low-risk women may be changed in the future.

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