Prevalence of Human Papilloma Virus Types in Turkish and Albanian Women

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Abstract

**Background:** Human papilloma virus (HPV) infection is the major etiologic agent of cervical carcinoma. The aim of this study was to determine the prevalence of HPV infection and genotype distribution in cervical swabs from 2,234 Turkish and 357 Albanian women with similar lifestyles from two different countries. **Materials and Methods:** HPV detection and typing were performed by type specific multiplex fluorescent PCR and fragments were directly genotyped by high resolution fluorescence capillary electrophoresis. **Results:** The most common type was HPV 16 and the second one was HPV 6 for both country. The third common type was 39 and 18 for Turkish and Albanian women, respectively. **Conclusions:** When we compare our results with other studies, there are differences between the frequency and order of the HPV genotypes detected at the second and subsequent frequencies. This may due to differences in the quality and type of samples analyzed, as well as the HPV detection methods.

**Keywords:** Albania, HPV prevalence, Turkey

**INTRODUCTION**

Cervical cancer is the second most common cancer among women worldwide. Over 90% of women with cervical cancer are human papilloma virus (HPV) positive. Persistence of the HPV infection as well as viral DNA insertion in epithelial cells are the main factors leading to high-grade dysplasia with the potential of progression to carcinoma in situ and invasive cancer. HPVs are small viruses composed of a nonenveloped capsid and a circular double stranded DNA genome with sizes close to 8 kb. Their genome is composed of three domains: a noncoding upstream regulatory region of approximately 1 kb, an early coding region (E genes), and a late coding region (L genes). Different epidemiological studies reveal that HPV prevalence depends on age and sexual practices and show major differences across geographic areas. HPV prevalence has been described to be higher in Latin America and sub-Saharan Africa and lower in Asia and Europe. Although more than 100 HPV types have been identified and approximately 40 of them are known to infect humans, only about 15 of them cause cervical cancer and its precursor lesions. HPV types 16 and 18 are responsible for about 70% of all cervical cancer cases and most of the female genital warts are caused by HPV types 6 and 11 in many developed countries. Fifteen mucosal HR-HPVs (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68, 73, 82) are reported to be associated with cervical intraepithelial neoplasia (CIN) and cervical cancer. HPV 16 is the most common HPV type detected in squamous cervical cancer (SCC, 55%), followed by HPV 18 (12.8%), and together these two types cause around 70% or more of SCC irrespective of geographical locale. The next most frequently detected HPVs worldwide are HPV 31, 33, 45, which contribution does not exceed 4% each.

**MATERIALS AND METHODS**

Our study included 2,234 Turkish and 357 Albanian cervical specimens with low- or high-grade cervical intraepithelial lesions (SILs) from Obstetrics and Gynecology clinics of the 20 different hospitals. Age ranges were between 21...
and 66 (mean age 35.4) and 19 and 67 (mean age 32.2) in Turkish and Albanian group, respectively. Each participant gave written-informed consent before she was enrolled and the study was approved by the institution. DNA was extracted using a commercial kit (Magnesia Viral Nucleic Acid Extraction kit, Anatolia, Istanbul, Turkey). Extracted DNA was amplified using a multiplex F-HPV PCR with a set of 15 fluorochrome-labeled primers recognizing HPV types within the E6 and E7 regions of the HPV genome. The F-HPV amplification was performed in a final PCR volume of 25 μL containing 20 μL of reaction mixture and 5 μL of extracted DNA for 35 repeat cycles of 30 s at 95°C, 30 s at 64°C, and 30 s at 72°C. Products were analyzed using capillary electrophoresis on an ABI 3500xl genetic analyzer and GeneMapper 5.0 Software (Applied Biosystems).

**RESULTS**

HPV DNA was positive in 850 of 2,234 (38.05%) Turkish cervical samples. Although 544 samples (64%) a single HPV type was genotyped, more than one HPV type was identified in 306 samples (36%) as shown in Table 1. HPV 16 as the most frequent type accounting for 143 of 850 typed cases (16.82%). HPV 6 was the second most common type being detected in 68 of 850 cases (7.58%). HPV 59 was the fourth with 54 of 850 cases (6.59%), HPV 52 and 18 were detected in 56 and 54 cases, respectively (6.4; 6.35%), and were followed by HPV 56 and 66 (50 cases, 5.88%). These eight types constitute about 70% of the total.

In Albanian group, HPV DNA was positive in 161 of 357 (45.09%). Single HPV type was calculated 67.08% (108 cases), multiple HPV type was calculated 32.92% (58 cases) as shown in Table 2. HPV 16 as the most frequent type accounting for 35 of 161 typed cases (21.74%). HPV 6 was the second most common type being detected in 22 of 161 typed cases (13.66%). HPV 18 is the third with 15 of 161 typed cases (9.32%).

**DISCUSSION**

In Turkey, HPV prevalence has been reported between 2.0% and 46.0% in some regional studies.[11-13] However, these studies had a limitation that they were conducted in a single center. With this study HPV type distribution, and the relationship between HPV positivity were studied from the cervical smear samples of women from 20 hospitals in different regions of Turkey. In Albania, HPV prevalence has been reported 15.1–43.9%.[14,15] In a study with a very large numbers of Turkish patients, 25% of women were found to be HPV positive.[16] This ratio is not compatible with our study (38.05%). This was possible due to the use of a multiplex PCR-based method that showed a high sensitivity and specificity. This HPV detection assay is also

| Table 1a: HPV positivity rate and prevalence of HPV types among 2,234 cases in Turkey. Positivity rate |
|---------------------------------------------------------------|---------------|
| **Single** | **Multiple** | **Total** | **%** |
| Negative | 1,384 | 61.95 |
| Positive | 544 | 306 | 850 | 38.05 |

| Table 1b: HPV positivity rate and prevalence of HPV types among 2,234 cases in Turkey. Prevalence of HPV types |
|---------------------------------------------------------------|---------------|
| **HPV Type** | **%** | **Single (n)** | **Multiple (n)** | **With 6 (n)** | **With 16 (n)** |
| 6 | 12.59 | 67 | 40 | 14 | 0 |
| 11 | 5.18 | 22 | 22 | 6 | 2 |
| 16 | 16.82 | 83 | 60 | 0 | 12 |
| 18 | 6.35 | 26 | 28 | 6 | 4 |
| 31 | 5.46 | 36 | 10 | 4 | 0 |
| 33 | 2.82 | 22 | 2 | 0 | 2 |
| 35 | 4 | 28 | 6 | 2 | 2 |
| 39 | 7.58 | 50 | 14 | 2 | 2 |
| 45 | 3.76 | 20 | 12 | 2 | 2 |
| 51 | 4 | 24 | 10 | 5 | 2 |
| 52 | 6.4 | 32 | 24 | 6 | 2 |
| 56 | 5.88 | 24 | 26 | 4 | 2 |
| 58 | 3.11 | 16 | 10 | 2 | 2 |
| 59 | 6.59 | 34 | 22 | 4 | 2 |
| 66 | 5.88 | 34 | 16 | 2 | 4 |
| 68 | 3.58 | 26 | 4 | 0 | 0 |

| Table 2a: HPV positivity rate and prevalence of HPV types among 357 cases in Albania |
|---------------------------------------------------------------|---------------|
| **Single** | **Multiple** | **Total** | **%** |
| Negative | 196 | 54.91 |
| Positive | 108 | 53 | 161 | 45.09 |

| Table 2b: HPV positivity rate and prevalence of HPV types among 357 cases in Albania |
|---------------------------------------------------------------|---------------|
| **HPV Type** | **%** | **Single (n)** | **Multiple (n)** | **With 6 (n)** | **With 16 (n)** |
| 6 | 13.66 | 14 | 8 | - | 1 |
| 11 | 3.1 | 3 | 2 | 1 | 0 |
| 16 | 21.74 | 28 | 7 | 1 | - |
| 18 | 9.32 | 10 | 5 | 1 | 1 |
| 31 | 6.83 | 4 | 7 | 0 | 2 |
| 33 | 4.97 | 4 | 4 | 0 | 1 |
| 35 | 4.97 | 6 | 2 | 1 | 0 |
| 39 | 3.1 | 3 | 2 | 0 | 1 |
| 45 | 1.24 | 0 | 2 | 1 | 0 |
| 51 | 3.1 | 4 | 1 | 0 | 0 |
| 52 | 3.73 | 4 | 2 | 0 | 2 |
| 56 | 4.35 | 5 | 2 | 0 | 0 |
| 58 | 4.35 | 4 | 3 | 0 | 0 |
| 59 | 5.6 | 6 | 3 | 2 | 0 |
| 66 | 8.07 | 11 | 2 | 0 | 0 |
| 68 | 1.86 | 2 | 1 | 0 | 0 |
very sensitive to detect multiple infections as it is based on the use of type-specific primers, rather than consensus/degenerated primers. In general, the rate of HPV positivity is low in studies using sequencing after PCR with degenerate primers as a method. In the presence of multiple infections, DNA sequencing is unable to resolve. There are some studies supporting this result and also compatible with the results of our study from Turkey. In a microarray-based study, 44.7% of all patients were HPV positive. In another study with a linear array method, HPV positivity was calculated 46.3%. Duran AC et al. reported that, in 100 of 261 genital samples, HPV DNA were positive using real time PCR method. This ratio, which is 38.3%, is almost the same as our result (38.05%).

Filipi et al. reported 15.1% of the cohort were found to be HPV positive. This rate is lower than both as a result of our study and also another HPV study from Albania. We think that the reason of this difference is due to using of the sequencing method, as discussed above. In a recent study from Albania, HPV prevalence has been reported 43.9% but there is no any molecular methods in this study to identify of the HPV type.

**CONCLUSION**

In summary, our findings show that the most common type was HPV 16, which is similar to data from lots of other studies. On the other hand, there are differences between the frequency and order of the HPV genotypes detected at the second and subsequent frequencies. This may be explained by differences in the quality and type of samples analyzed, as well as the HPV detection methods.

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**Conflicts of interest**

There are no conflicts of interest.

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