

Genotyping of Human Papillomaviruses by a Novel One-Step Typing Method with Multiplex PCR and Clinical Applications

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Abstract

We describe here a rapid, high-throughput genotyping procedure that allows for the simultaneous detection of 16 high- and low-risk genital human papillomaviruses (HPV) with multiplex-PCR in a single reaction tube. Multiplex-PCR is based on the amplification of HPV DNA by sets of HPV genotype-specific primers, and genotypes of HPV are visually identified by the size of amplicons separated by capillary electrophoresis. The procedure does not include a hybridization step with HPV-specific probes and is rapid and labor-saving. We detected all 16 HPV genotypes (16, 58, 52, 51, 56, 31, 18, 39, 66, 59, 6, 33, 30, 35, 45, and 11) with high sensitivity and reproducibility. By using this newly developed method, we conducted a pilot study to examine the correlation between the prevalence and genotype distributions of HPV and the cytological group classifications in 547 cervical samples. Compared with the normal group (14.7%), there is a significant increase in HPV prevalence in ASCUS (61.3%), LSIL (75.8%), and HSIL (82.2%). Prevalence and genotype distribution of type 58 is correlated with cytological malignancies with the highest prevalence in HSIL. In conclusion, the novel multiplex-PCR method described in this paper appears highly suitable not only for the screening of cervical cancer precursor lesions but also for the characterization of genotype distributions in large-scale epidemiological studies and HPV vaccination trials.