

## Intratypic variation of human papillomavirus type 11 in recurrent respiratory papillomatosis

Zsófia Nagy<sup>1</sup>, Gábor Kardos<sup>1</sup>, Attila Szűcs<sup>2</sup>, Krisztina Szarka<sup>1</sup>

<sup>1</sup>Department of Medical Microbiology; <sup>2</sup>Clinic of Otorhinolaryngology and Head & Neck Surgery, University of Debrecen, Nagyterdei krt. 98., H-4032 Debrecen, Hungary

Recurrent respiratory papillomatosis (RRP) is mainly caused by low risk human papillomavirus (HPV) genotypes, HPV6 and HPV11. The infection with HPV 11 is usually associated with a more aggressive clinical appearance of the disease. Our aim was to investigate the genetic characteristics of HPV11 sequences from RRP patients with different clinical course. Our recent studies have shown that relationship is assumed between intratypic variation of HPV 11 and the different outcome of the RRP. In this study, HPV11 sequences from four new and two already known patients were analysed and compared to our previous results and GenBank data.

After nucleic acid extraction, HPV genotypes were determined in the samples and virus copy number were also estimated by real-time PCR with SYBR Green. After complete genome amplification, amplicons were purified from gel and sequenced. The assembly of the complete genome of HPV11s and phylogenetic analysis together with GenBank data were performed by CLC Gene Workbench 5.7 software.

The HPV11 genomes from recurrent papillomas were identical with the HPV11 sequences from the patients' previous recurrences. In case of a new patient, new nucleotide polymorphisms were not identified, however, in case of two additional patients, previously unidentified nucleotide polymorphisms were found. HPV11 sequences originated from three different localization of an extended papilloma were identical with each other, and the E1 open reading frame (ORF) of these genomes were identical with the E1 ORF of the reference genome derived from an extremely aggressive juvenile onset RRP. Finally, in a sample of a new patient, unique nucleotide polymorphisms causing amino acid changes in E1, E2 and E4 ORFs and T7330G polymorphism in the long control region were also identified. In addition, the previously described T7546C polymorphism, which decreases the activity of LCR significantly, was found in all newly enrolled sequences.

Two main phylogenetic lineages were distinguished in the phylogenetic analysis of the complete genomes available in the GenBank (A1 and A2); all Hungarian HPV11 sequences belong to the group A2. The existence of a sublineage with higher pathogenic potential is also assumed within group A2. This sublineage characterized by polymorphisms in E1, E2, E4, L1 ORFs and LCR; some of them may be related to the severe clinical outcome of the RRP:

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