

7 The presence or absence of human papillomavirus type 16 subgenomic regions in neoplasia of the uterine cervix. M. Yajima, Y. Matsushima, M. Terada, Genetics Division, National Cancer Center Research Institute, Tokyo.

Four subgenomic regions in HPV 16 genome, E6/E7, E1/E2, L1/LCR (long control region) and L2 regions were amplified by the polymerase chain reaction (PCR), and Southern blot analysis was performed using HPV 16 total genome on formalin-fixed paraffin-embedded tissues in 22 cases of cervical carcinoma and 21 cases of CIN III. Total positive rate of HPV 16 was 37% (16/43). In cases of cervical carcinoma stage I and II, HPV 16 DNA was detected in 9 out of 22 (41%) cases. In 6 out of the 9 HPV 16 positive cases, a part of or all the cases of E1/E2 and L2 regions could not be amplified, which strongly suggested that the HPV 16 viral genome in the 6 cases was truncated and integrated into human genome. In cases of CIN III, HPV 16 DNA was detected in 7 out of 21 (33%) cases. Only one out of the 7 HPV 16 positive cases showed a truncated pattern as shown above. These results indicated that the incidence of integration with deleted E1/E2 and/or L2 region greatly increased in the invasive carcinoma compared with CIN III, further the integration could also occur in severe dysplasia.

8 **Analysis of HPV16 mRNA in cervical carcinomas and cervical intraepithelial neoplasias by polymerase chain reaction with reverse transcriptase reaction.** A. Nishikawa, M. Fukushima, M. Shimada*, Y. Yamakawa, S. Shimano, Y. Sawada*, K. Fujinaga*, M. Hashimoto, Dept. Obstet. and Gynec., *Dept. Mol. Biol., Cancer Res. Inst., Sapporo Med. Col., Hokkaido, Bio Res. Lab., **Takara Shuzo Co. Ltd., Shiga.

We present the PCR detection method of HPV 16 mRNA in small amounts of specimens, and using the method, each region of HPV16 mRNA in cervical intraepithelial neoplasias (CINs) and invasive cervical carcinomas were investigated. HPV 16 E6/E7 mRNAs were identified in all of HPV 16 DNA positive samples from CINs and cervical carcinomas. There were three expression patterns in the E6/E7 region; full size E6, E6* II and predominantly E6*I in both lesions. Regarding E1, E2, E4 and E5 regions, mRNAs were detected with heterogeneous patterns. Among the HPV 16 mRNAs analyzed, full sized E2, and spliced E1⁺E2-C and E1⁺E4 were detected, and splice junctions were sequenced. In a case of CIN, regarding E6/E7 and E1 regions, both mRNA and DNA were detected. However, neither E2-E5 mRNA nor DNA were amplified suggesting deletions in the E2-E5 region in the viral genome. But no specific difference in the mRNA expression pattern were found between CINs and cervical carcinomas.

9 Detection of HPV types 16 and 18 early gene E7 messenger RNA in uterine cervical cancers by polymerase chain reaction. H. Tanimoto, N. Nagai, S. Ohta, H. Fujimoto, A. Fujiwara, K. Yoshida*, E. Tahara*, Dept. Obst. and Gynec., Hiroshima Univ. Sch. Med., *Onomichi general Hosp., **First Dept. Pathology Hiroshima Univ. Sch. Med., Hiroshima.

The expressions of mRNA for HPV types 16 and 18 genes were examined in 22 uterine cervical cancers by Northern blotting and reverse transcriptase polymerase chain reaction (RT-PCR). By Northern blotting, HPV type 16 mRNA were detected in 5 (23%) of 22 cases and HPV type 18 mRNA were detected in 5 (23%) of 22 cases. On the other hand, HPV type 16 early gene E7 were detected in 16 (73%) of 22 cases by RT-PCR. We also examined expression of early gene E5, which was detected only 2 (9%) of 22 uterine cervical cancers. These results suggest the following; 1. RT-PCR is high sensitive method compared to Northern blotting. 2. HPV type 16 E5 gene might be deleted frequently in cancer cells when HPV type 16 DNA exist as an integrated form in cancer cells or transcription of E5 gene might be suppressed, while E7 mRNA is detected frequently and might have an important role in carcinogenesis of uterine cervical cancer.