

HPV 16 L1L2 GENE EXPRESSIONS, PROTEIN SYNTHESIS AND INTERACTION IN HUMAN CELL CULTURE

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At least 15 distinct Human Papillomavirus (HPV) types are described to be involved in genital, mouth, throat, and skin cancers. More than 50% of cervical cancers worldwide are attributed to HPV16. L1 is the HPV major and L2 is the minor capsid protein. We are producing HPV16 L1L2 VLPs for investigating the mechanisms by which virus-cell infection cause cancer. Cell cultures of 293T human embryonic kidney cell line were transfected with the DNA constructs encoding for humanized L1, (L1h) and L2h antigen of HPV16, sub cloned into the mammalian expression vectors pUF3L1h and pUF3L2h. Western blotting to control protein expression, immunofluorescence in laser scanning confocal microscopy (LSCM), negative staining and gold immunolabeling for VLPs analyses by transmission electron microscopy (TEM) were used. Pathogen-host cell interaction assays using HPV16 L1L2 VLPs were performed. Recombinant L1L2 DNA was expressed in 293T cells in a high efficiency. At least 85% of cells expressed intracellular L1L2 and VLPs, detected by LSCM and TEM. The HPV16 L1L2 VLPs produced in this study interacted with non-transfected 293T cell line confirmed by LSCM. We are establishing a methodology for an efficient system of recombinant protein expression. The production of HPV16 L1L2 VLPs by transfected 293T cells opens the possibility for new basic studies concerning to HPV-cell interactions and carcinogenesis mechanisms.

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