Serological Assay Of HPV 16 E6 and E7 Oncoproteins in Patients with Oral Squamous Cell Carcinoma

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Objectives: To develop an ELISA for detection of anti-HPV 16 E6 and E7 antibody from serum and to study the association between HPV 16 seropositivity and OSCC patients in Malaysia.

Methodology: HPV 16 E6 and E7 antigens were constructed in recombinant plasmids as glutathionen-s-transferase (GST) fusion proteins. A GST-capture ELISA has been established using these constructed antigens and optimized. 50 healthy control serum samples and 50 serum samples from OSCC patients were tested in this HPV ELISA. Prevalence of HPV 16 seropositivity was determined and statistical analysis was performed to determine the association of HPV 16 seropositivity and risk of oral cancer.

Results: HPV 16 E6 and E7 recombinant antigens were successfully constructed and expressed in Escherichia coli cell lysate. Expressed recombinant antigens were validated in SDS-PAGE and western blot. Using the ELISA assay established in this study, 30.0% cases and 16.0% of controls were detected to be HPV 16 E6 positive, and E7 positive in 18.0% of cases and 14.0% of controls. The variation of HPV 16 serology prevalence in male and female subjects was revealed. The adjusted risk of OSCC was elevated among those male subjects positive for HPV 16 E6 (OR=21.739, 95% CI=1.302-333.333) and HPV 16 E6 and E7 double positivity (OR=21.739, 95% CI=1.302-333.333). Furthermore, suggesting significant increased risk of OSCC was also observed in HPV 16 E6 and E7 double positivity among those non-betel quid chewers when variation of HPV 16 serology in different habit group was examined (OR=12.987, 95% CI=0.927-166.667). No significant association was found between HPV 16 seropositivity and site of tumour and survival rate.

Conclusion: In summary, a simple and convenient HPV 16 E6 and E7 ELISA assay has been established in this study. Potential association of HPV 16 E6 and E7 seropositivity with increased risk of OSCC in men subjects and non-betel quid was revealed in this study. In conjunction with previous study, this study presents additional evidence of HPV infection in OSCC in Malaysia.