CO-EXPRESSION OF HPV E6, E7 MRNA AND PD-L1 IN CERVICAL CYTOLOGY SAMPLES

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Background

HPV infection in most women is transient and clears over time. For others, the virus is persistent and can lead to pre-cancerous lesions and subsequently cervical cancer. The relatively high regression rate of cervical intraepithelial lesions (CIN) has similarly been attributed to engagement of the immune response directed against neoplastic cells. Recent advances in immuno-oncology have shown the dramatic effects of PD-1/PD-L1 inhibitors in epithelial tumors including squamous cell carcinoma and adenocarcinoma, the major cancer subtypes in the female genital tract. Here, we present a novel assay that combines RNA in situ hybridization for HPV E6, E7 mRNA, cell cycle analysis, and PD-L1 cell surface staining on epithelial cells in liquid-based cervical cytology specimens.

Methods

Forty-six residual cervical cytology specimens were obtained for this study: 25 HPV DNA-, 12 LSIL HPV DNA+, and 9 HSIL HPV DNA+. Samples underwent in-situ hybridization with E6,E7 mRNA probes and a cell cycle dye. Anti-PD-L1 antibody was added following in-situ hybridization. Samples were collected on a Beckman Coulter CytoFLEX. Samples were deemed positive or negative for E6,E7 and Post G1 expression by a dual cut-off of 3.15%. PD-L1 expression was determined based on a cut-off of 2%.

Results

	Positive % E6,E7 and Post G1	Positive % PD-L1	% Dual E6,E7 and PD-L1
HPV DNA-	24% (6 of 25)	24% (6 of 25)	83% (5 of 6)
LSIL	50% (6 of 12)	25% (3 of 12)	50% (3 of 6)
HSIL	33% (3 of 9)	11% (1 of 9)	33% (1 of 3)

Conclusion

In this study we show dual E6,E7 and PD-L1 expression on the same sample. HPV and PD-L1 expression on cell by cell basis is not currently available in a single test by any other method. It appears PD-L1 expression decreases in high grade lesions indicative of immune surveillance which could support therapeutic options.