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2241 / 30 - Molecular characterization of circulating tumor cells in head and neck squamous cell carcinoma: Direct comparison of a label-independent size-based microfluidic device with EpCAM-based CTC enrichment

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Presenter/Authors

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Disclosures

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Abstract

Background: Circulating tumor cells (CTCs) heterogeneity is highly affecting the efficiency of their isolation and thus the reliability of downstream analysis. Especially in Head and Neck Squamous Cell Carcinoma (HNSCC) epithelial mesenchymal transition (EMT) is highly affecting CTC isolation and downstream analysis. We directly compared two different approaches used for CTC isolation, a label-independent size-based microfluidic-based system versus an EpCAM-based positive selection for downstream molecular characterization of CTC both at the gene expression and DNA methylation level in HNSCC.

Methods: Peripheral blood (PB) in EDTA (20mL) was collected from 50 HNSCC patients and 18 healthy donors (HD). A size-based microfluidic device (Parsortix, ANGLE) and an EpCAM-based positive immune-magnetic isolation procedure were applied in parallel, using 10mL PB in each case. Total RNA was isolated from enriched CTCs and RT-qPCR was used to study the expression levels of *CK-19*, *PD-L1*, *EGFR*, *TWIST1*, *CDH2* and *B2M*. Real time methylation specific PCR (MSP) was used to study the methylation status of *SOX17, RASSF1A* and *MLL3* genes in DNAs isolated from the same enriched CTCs.

Results: In identical blood draws, the label-free size-based CTC-isolation system was superior in terms of sensitivity when compared to the EpCAM-based CTC enrichment, since a significantly higher percentage of identical PB samples was found positive for all genes tested both at the gene expression and DNA methylation level, while the specificity was not affected.

Conclusions: In HNSCC CTC molecular characterization at the gene expression and DNA methylation level should be based on a label-free size-based isolation system.