Exploring the clinical relevance of circulating tumor cells and tumor-derived extracellular vesicles collected from different blood compartments in colorectal cancer patients

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Background & objectives

Circulating tumor cells (CTCs) and tumor-derived extracellular vesicles (tdEVs) have great potential for monitoring therapy response and detecting tumor relapse earlier, which could lead to personalized adjuvant therapeutic strategies. However, their low abundance in peripheral blood limits their informative value. In this study, we explored the presence of CTCs and tdEVs collected intraoperatively from a tumor-draining vein (DV) and via a central venous catheter (CVC) prior to tumor resection as an alternative to standard blood samples collected from a peripheral vein (PV).

Methods

We analyzed 371 blood samples from 290 gastrointestinal (GI) patients, and 31 samples from 20 patients with diverse benign tumours using the CellSearch system. The ACCEPT software and R scripts were used for automated detection of CTCs and tdEVs and their characterization. All 371 samples were analyzed in order to optimize the CTC and tdEV gates. The selected gate settings were applied to 199 CRC samples which included 2,159 CTCs and 13,279 tdEVs from the total collective. The corresponding parameters were obtained by numeric analysis of their fluorescence signals (e.g. size; shape; intensity) and were utilized for particle classification using machine learning tools, e.g. k-means clustering.

Results

tdEVs were generally more abundant than CTCs. Frequency of CTCs and number of CTCs and tdEVs were highest in DV. CTCs and tdEVs in PV were associated with tumor-spread while DV-CTCs associated with tumor size (univariate $p < 0.01$). In all three compartments, uni- and multi-variate Kaplan-Meier analyses revealed superior relation of tdEVs (univariate Logrank test $p < 0.05$) with shorter survival compared to CTCs. In DV, CTCs and tdEVs were

characterized by elevated parameters concerning the cytokeratin (CK) intensity and size ($p < 0.0001$). Interestingly, ROC and k-means analyses identified sub collectives of tdEVs defined by stronger CK intensities which were particularly linked to poor survival outcomes.

Conclusion

Our study indicates that collecting CTCs and tdEVs from DV and CVC sites provides more valuable information than from PV. tdEVs have higher clinical and prognostic value than CTCs, and analyzing both types suggests significant phenotypical heterogeneity. These results have important implications for personalized adjuvant therapies for colorectal cancer patients.