Extracellular vesicle (EV) associated DNA: potential liquid biopsy biomarker and functional communicators in cancer

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Background & objectives

Clonal evolution and heterogeneity of tumors that survived initial therapy or acquired additional mutations independent of treatment stress is the primary cause of cancer recurrence. Therefore, sensitive and reliable methods to accurately measure mutational shifts between diagnoses during therapy and relapses could provide helpful information on disease progression. Extracellular vesicles (EVs) such as exosomes (diameter: 70–150 nm) released from tumor cells have emerged as potentially valuable biomarkers as they have been shown to carry disease-specific signatures representing the pathological state of the individual cells. In addition, EV-associated biomolecules released from cancer cells can mediate microenvironmental changes in healthy cells toward malignant traits. Therefore, our project aims to establish EV-DNA as biomarkers to detect minimal residual cancer and evaluate the functional role of EV-DNA in cancer.

Methods

EV isolation was based on differential centrifugation, ultrafiltration, and size-exclusion chromatography. The EV quality check (purity and integrity) was performed by Transmission electron microscopy (TEM). Nanoparticle tracking analysis (NTA) was performed using Zetaview to analyze EV size (diameter) and concentration in the preparation. Bead capturing and single EV detection with surface markers CD63 and CD81 was performed using FACS. Sheared EV-DNA-protein (EV-chromatin) complex was prepared using a sonicator, and the isolated chromatin was subjected to immunoprecipitation using an anti-dsDNA antibody and LC-MS analysis. EV-chromatin was imaged using atomic force microscopy. Isolated chromatin was packaged in artificial polymerosomes for function experiments to address the communication between leukemia-derived EVs with bone marrow-derived mesenchymal stromal cells in the microenvironment.

Results

We can show the potential of EV- DNA as a biomarker to detect cancer-specific mutation (1,2). Further, we can demonstrate the functional potential of EV-chromatin in downregulating key tumor suppressor protein p53 in BM-MSC as a mechanism to support the survival of leukemia cell clones in the microenvironment (3,4).

Conclusion

Our study suggests the biomarker potential of tumor EV-DNA-protein complex in early cancer detection and its functional role in cancer progression. We also highlight the limitation of EV-DNA detection in routine diagnostics, especially MRD, due to heterogeneous EV-population derived from healthy cells and tumor cells in blood samples.

References:

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