Diagnosis and Treatment of Breast Cancer based on Extracellular Vesicles

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Overview

Early detection of breast cancer (BC) and monitoring its progression, treatment response, and recurrence are crucial for survival. Current imaging methods for diagnosis are limited, and biopsies, which are the â€œgold standardâ€ for diagnosis and prognosis, are sometimes unavailable or hard to obtain and involve significant discomfort to patients. Therefore, there is a great need for non-invasive, accurate BC diagnosis and monitoring methods. Analysis of protein content of small EVs (sEV) is a promising approach; however, the lack of a standardized and reliable method for sEV purification and analysis significantly limits its clinical application. Prof. Lev and her team developed an efficient and cost-effective method for sEV purification from BC patientsâ€™ blood. Based on their analysis, they further identified specific biomarkers for BC diagnosis, staging, and evaluating the risk of relapse.

Background and Unmet Need

Early detection of BC is crucial for successful therapy and survival. Currently, screening commonly relies on mammography, ultrasound, and magnetic resonance imaging. However, these methods are not always reliable, safe, and/or cost-effective. Currently, tumor biopsies are considered the â€œgold standardâ€ of diagnosis, prognosis, and prediction of therapeutic response. However, obtaining the biopsies is often not trivial and is associated with potential morbidity and patient inconvenience. Moreover, in metastatic patients, tumor biopsy is limited by sampling a single metastatic site among many presents.

As a complement approach, increasing evidence suggests that protein contents of small circulating extracellular vesicles (EVs) derived from the plasma of BC patients represent a promising approach for early detection, diagnosis, and prognosis. Small EVs (sEV) of 30 to 100 nm size, such as exosomes or exosome-like vesicles (ELVs), are released from multiple cell types, including leukocytes, platelets, fibroblasts, adipocytes, and cancer cells, and can be found in multiple body fluids such as blood, semen, urine, saliva, breast milk, and cerebrospinal fluid. sEVs contain proteins, microRNA, mRNA, and DNA and play important roles in cell-cell communication by transferring their content to target cells. sEVs are robustly produced by cancer cells and markedly affect the primary tumor microenvironment (TME), including the immune ecosystem as well as distant metastatic niches, thereby facilitating tumor growth and metastasis. sEVs may provide unique information about the full metastatic status of tumors and allow a facile longitudinal analysis of tumor evolution in response to therapy.

A major obstacle in using sEV for clinical evaluation is that multiple methods for sEV isolation are currently used, including differential centrifugation, density gradient centrifugation, size-exclusion chromatography (SEC) and/or affinity chromatography, microfluidic devices, and synthetic polymer-based precipitation reagents. This diversity results in a lack of a standardized, reliable method for sEV purification, limiting the clinical use of sEV profiling for BC diagnosis.

The Solution

Prof. Sima Lev and her team developed an efficient approach for EVs isolation from plasma of patients with BC.

Based on their proteomic analysis, they identified specific biomarkers enabling BC diagnosis, staging, and evaluating the risk of relapse.[1]

Technology Essence

standardize the sEV purification method and overcome the challenges of plasma proteins contamination, the team established an efficient and cost-effective protocol that requires only 2 ml of plasma, resulting in high yields of sEVs. The enrichment protocol includes a filtration stage followed by size exclusion chromatography (SEC), which retains sEVs integrity and decreases plasma protein contaminants. Semiquantitative proteomic analysis was performed by the Reverse-phase protein array (RPPA) technology, a fast, cost-effective, and reliable method that covers ~300 key proteins and/or their phosphorylation status. The analysis revealed signatures of specific protein combinations and thresholds that can be used for BC diagnosis and for determining the stage of BC. Finally, the team identified specific biomarkers that correlate with the risk of BC relapse.

Applications and Advantages

- An efficient, non-invasive, and cost-effective screening method for BC diagnosis and staging
- Can be used to monitor cancer progression over time to determine responsiveness to treatment and assess risk for cancer relapse
- Provides a comprehensive assessment of the primary tumor and all metastasis

Development Status

A standardized and efficient protocol for sEV isolation from BC patients plasma was successfully developed, and a signature of specific protein expression levels was identified for BC diagnosis, staging, and risk of relapse. Based on this information, designated kits comprising specific antibodies can be made for BC diagnosis.

References

1. Vinik Y, Ortega FG, Mills GB, et al. Proteomic analysis of circulating extracellular vesicles identifies potential markers of breast cancer progression, recurrence, and response. Sci Adv. 2020;6(40):eaba5714. doi:10.1126/sciadv.aba571

Patent Status

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