Proteomic Profile of membrane Proteins from HCC-1954 Breast Cancer Cell line: molecules involved in adhesion mechanisms

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Introduction:
Breast cancer can disseminate to regional lymph nodes and establish distant metastases, preferentially in bone, lung, and liver tissues, resulting in poor outcome and high mortality. The molecules closely associated with these processes are predominantly present at the cell surface and in the extracellular space, establishing the first contacts with the target tissue. The HCC1954 is a hormone receptor negative, ERBB2 positive, poorly differentiated cell line derived from a primary stage IIA, grade 3, invasive ductal carcinoma, which has been the subject of several large-scale genome and transcriptome analysis. We aimed to identify cell surface proteins present in HCC1954 cell line that can be involved in the development of invasive ability and metastases.

Material and Methods:
Proteins were biotinylated and fractionated using the cell surface protein isolation kit (Pierce®), followed by in-gel digestion. The peptides obtained from all fracion were analyzed on a 1-D reversed-phase chromatography coupled to an ESI-Q-TOF and a 2-D SCX/RP chromatography coupled to a Synapt HDMS mass spectrometer (Waters, Farmington, MI, USA). The mass spectra were acquired on data-dependent scanning (DDA) experiments. For spectra processing and data analyses, ProteinLynx Global Server v.2.5 and Peaks Studio software v. 6.0 were used. Gene Ontology mapping was performed using Panther tools.

Results and Discussion:
More than 1500 proteins of the HCC-1954 breast cancer cell line were identified. The enriched membrane fraction enabled the identification of 3 times more plasma membrane proteins by both proteomic identification methodologies. Among them, molecules that plays important roles in cell migration and angiogenesis such as integrins α-1, α-3, α-v, α-6, molecules involved in cell adhesion, cell junction and cell-cell interactions as catenin α-1/cadherin-1, CD44, CD166, CEAM5, JAM-1, TBB4 were identified.

Conclusions:
This strategy allowed the identification of several cell surface proteins involved in cell-cell adhesion, cell-matrix adhesion and cell motion providing information about targets for further anti-metastatic therapies.

Key Words: cell surface proteins, plasma membrane proteins, biotinylation, HCC1954, proteomics
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